

Sea-derived microalgae leads to healthier red meat and reduced methane emissions

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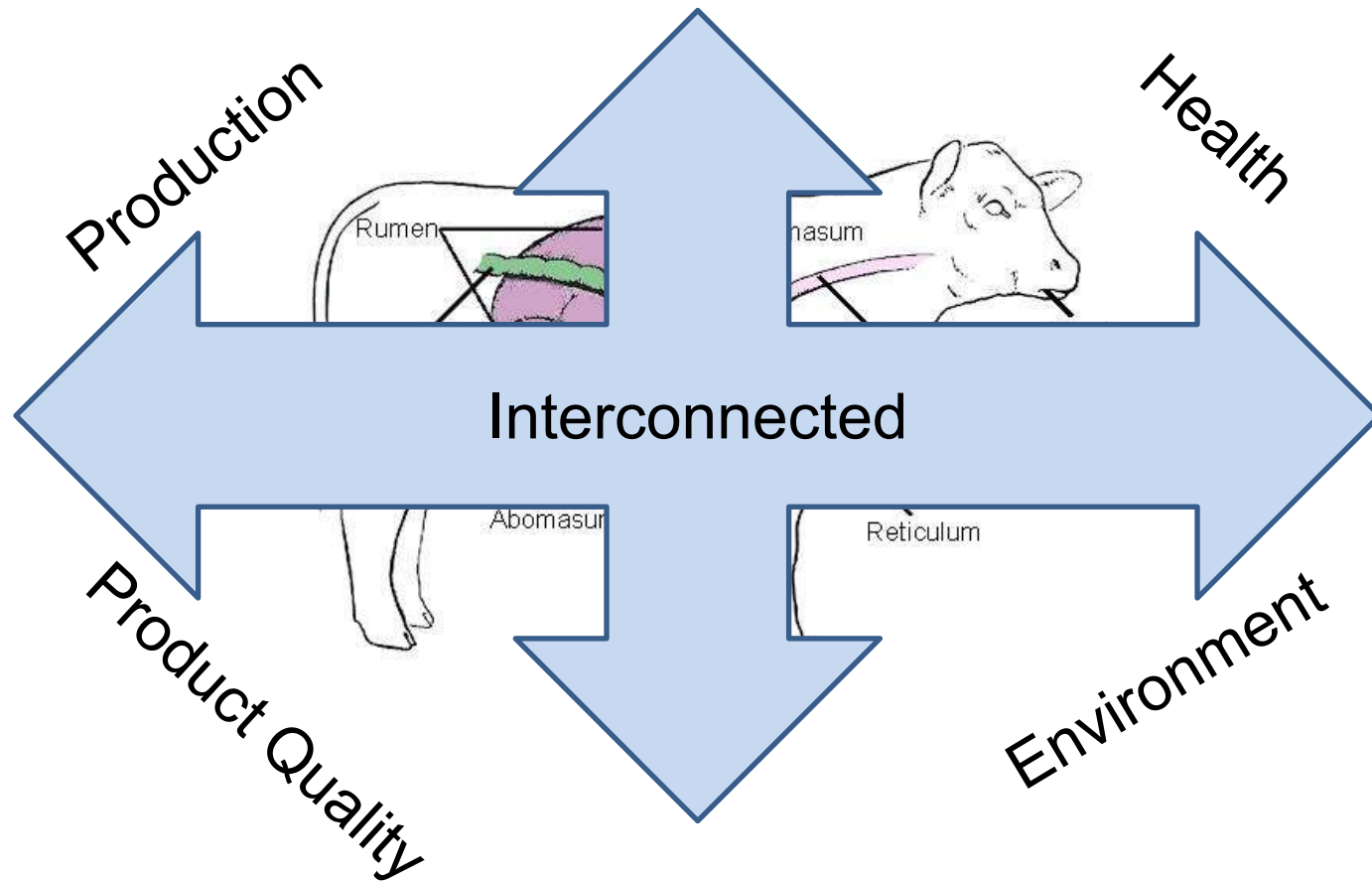


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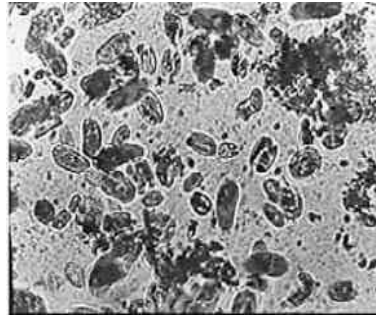
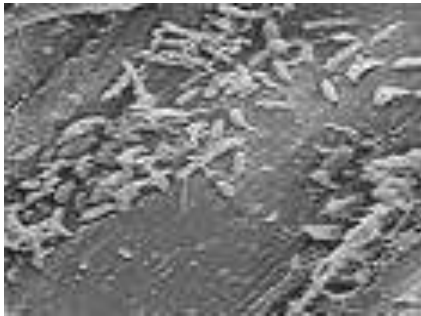
IGFS

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Ruminant Sector Challenges



Rumen Microbiome Central to the Challenges



Bacteria

10^9 - 10^{10} /mL

Protozoa

10^4 - 10^6 /mL

Fungi

10^3 - 10^4 /mL

Archaea
(methanogens)

Approx. 10^4 /mL

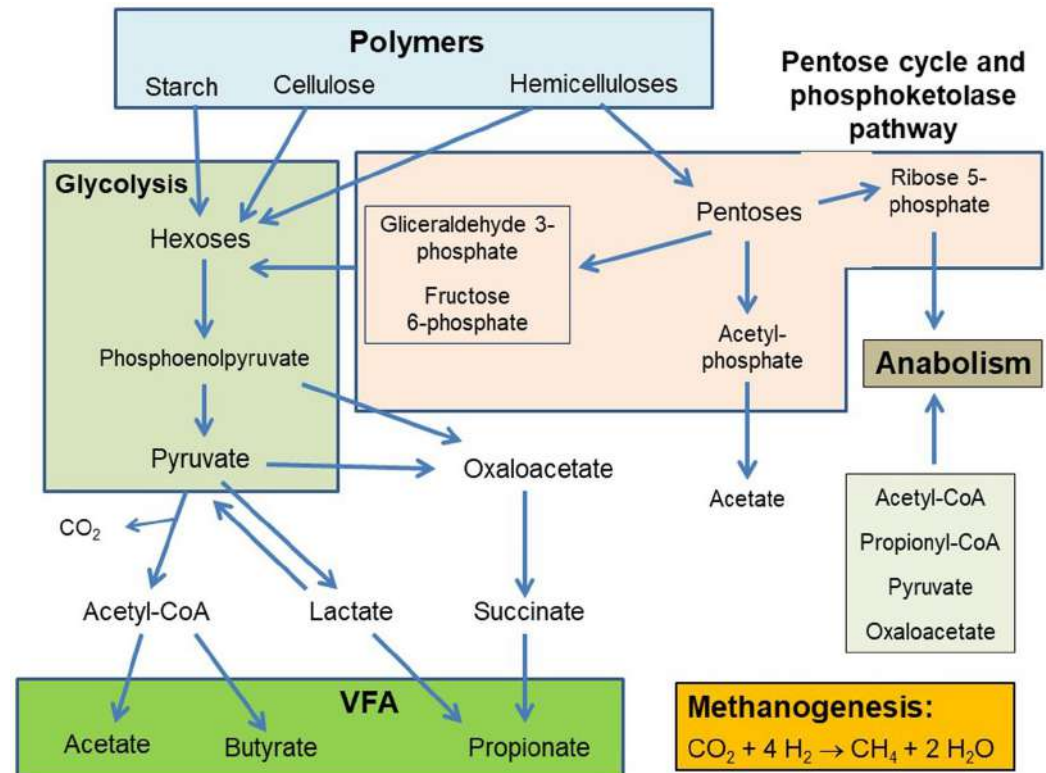
Huws et al. 2018. Addressing Global Ruminant Agricultural Challenges Through Understanding the Rumen Microbiome: Past, Present, and Future. *Frontiers in Microbiology*. 9:2161. doi: 10.3389/fmicb.2018.02161.

The rumen microbiome in action



Rumen fermentation

- Cellulose/hemicellulose to Volatile fatty acids: Energy source but source of released H.
- Volatile fatty acids:
 Acetate, lactate, butyrate, succinate, propionate
- Hydrogen sinks:
 Pyruvate to lactate: utilizes 2H
 Lactate to propionate: utilizes 2H
 Acetyl-CoA to butyrate : utilizes 2H

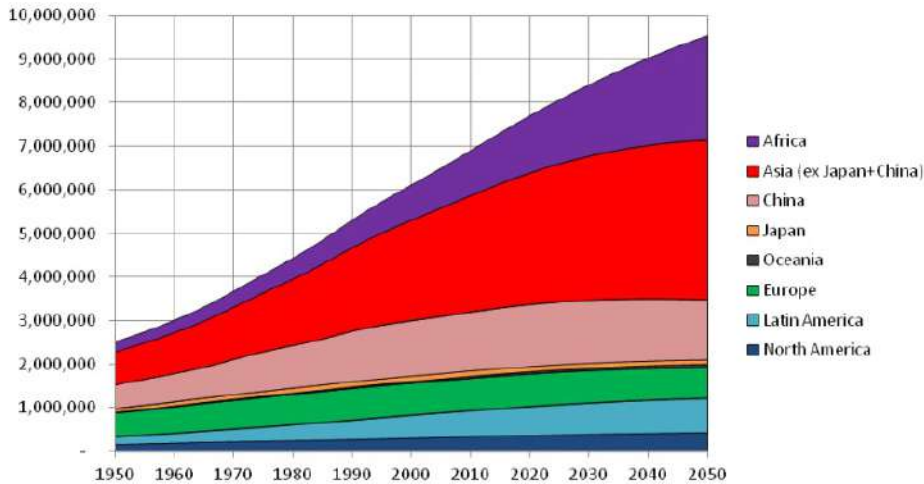


Source: Ungerfeld EM. Metabolic Hydrogen Flows in Rumen Fermentation: Principles and Possibilities of Interventions. *Frontiers in Microbiology*.11:589. doi: 10.3389/fmicb.2020.00589.

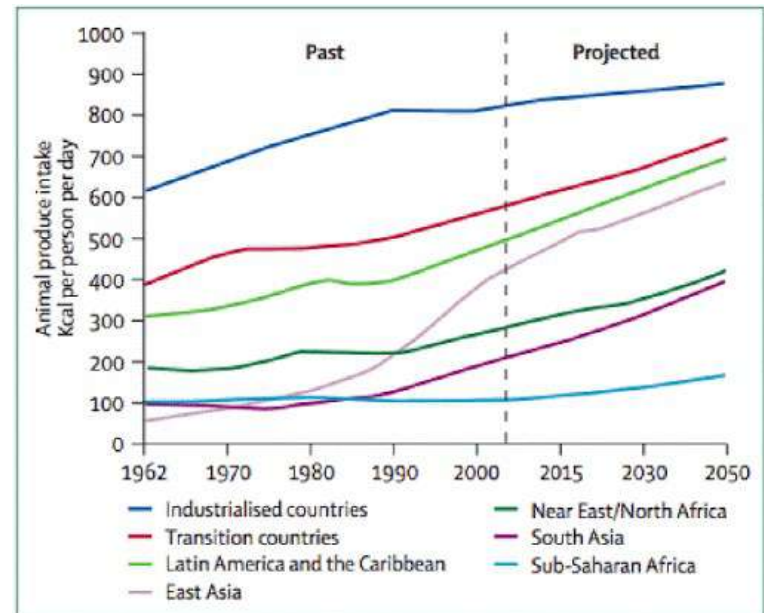
Food Security

World Population

Source: UN



Graphic: deconstructingrisk.com

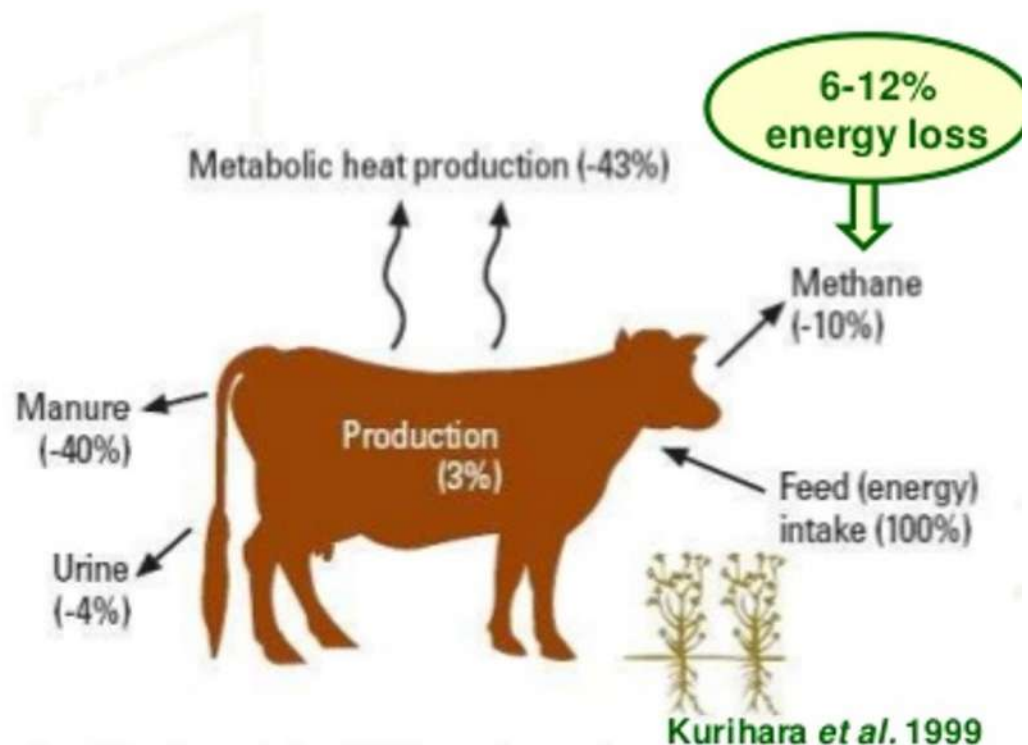


FAO predict meat and dairy production will have to increase by 76% and 63% respectively by 2050

Methane and Productivity

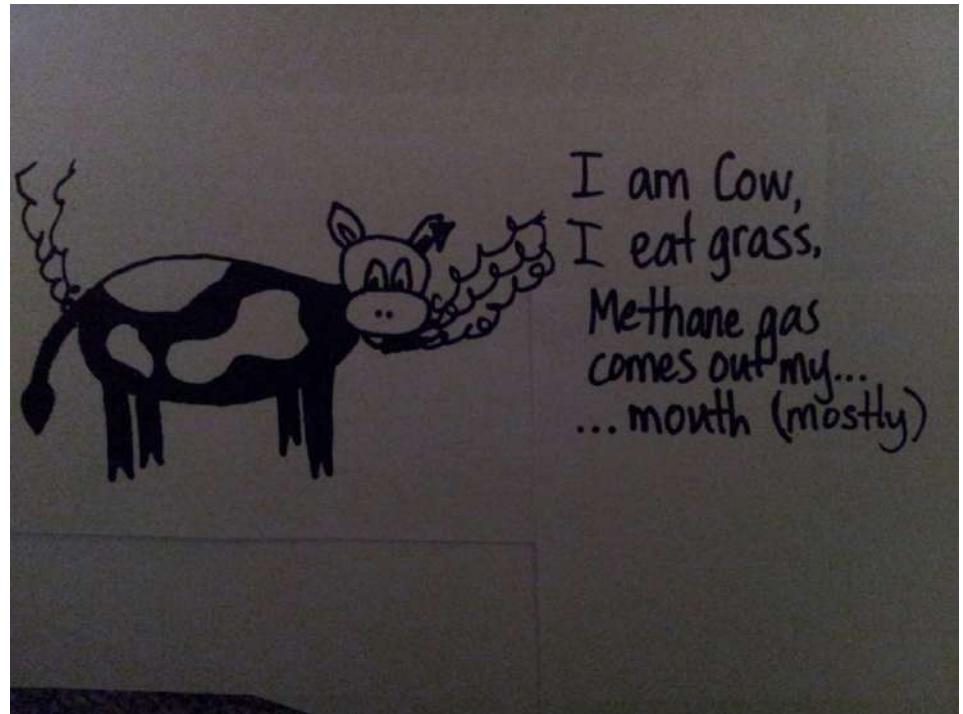
Data has shown that ruminants which release less methane are more productive.

Some dietary interventions to reduce methane may increase productivity.



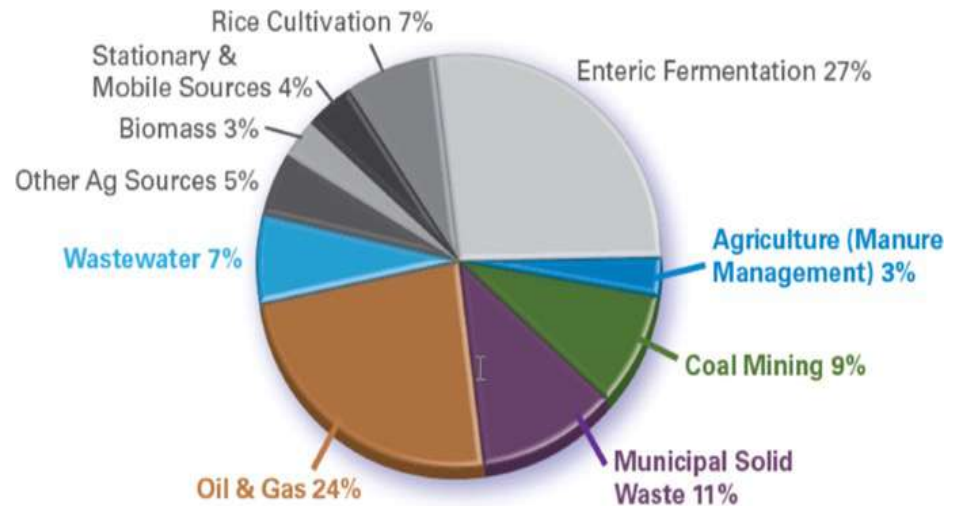
Ruminants and Methane

- FAO state that livestock agriculture responsible for approx. 18% of GHG emissions, mainly in the form of methane.
- Paris agreement: Limit global warming to less than 2%. A 45% reduction in methane emissions could reduce global warming by 0.3°C.
- Recent US-EU climate pledge to reduce methane emissions by 30% by 2030.



Cross-Sectorial GHG emissions

- Fossil fuel industry are the greatest producers of CO₂ which remains in the atmosphere for over 100 years.
- Livestock mainly produce methane which has a half life of less than 10 years.
- Irrespective need to decrease environmental impact of livestock by 2050.



A large green graphic on the left side of the slide. It consists of a stylized plant stem with a single leaf at the top. Below the stem is a large, dark grey lowercase letter 'm' enclosed within a green circular outline. At the bottom of the stem, there are three wavy green lines representing roots or soil.

THE MASTER PLAN: Microbiome Applications for Sustainable food systems through Technologies and EnteRprise

Project Co-ordinator:

Prof Paul Cotter, Teagasc, Ireland



<http://www.master-h2020.eu/>

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the master consortium

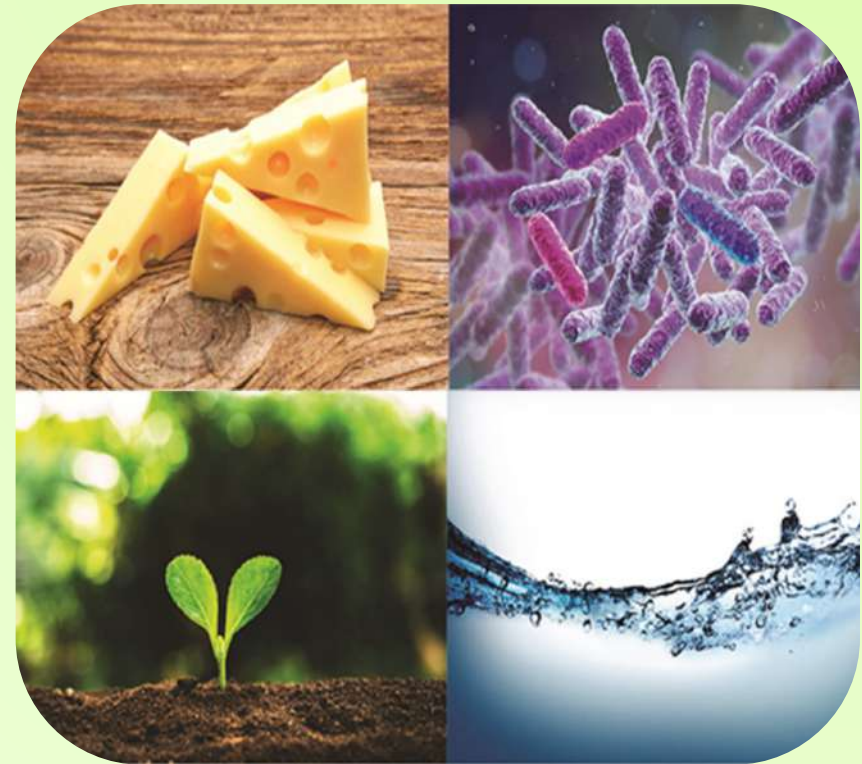
A Ireland Teagasc CIRCA UCC Devenish 4D Pharma ICBF	H France INRA Novolyze IFREMER Anadiag Danone
B Spain CSIC ULE	I Germany Qiagen INOQ
C Finland LUKE	J Denmark CH
D Austria FFOQSI AIT	K Belgium AB-Inbev
E Italy UNINA UNITN	L Netherlands WU BASECLEAR
F UK QUB AFBI ONT	M Iceland MATIS
G Romania TTC	N Norway FVG

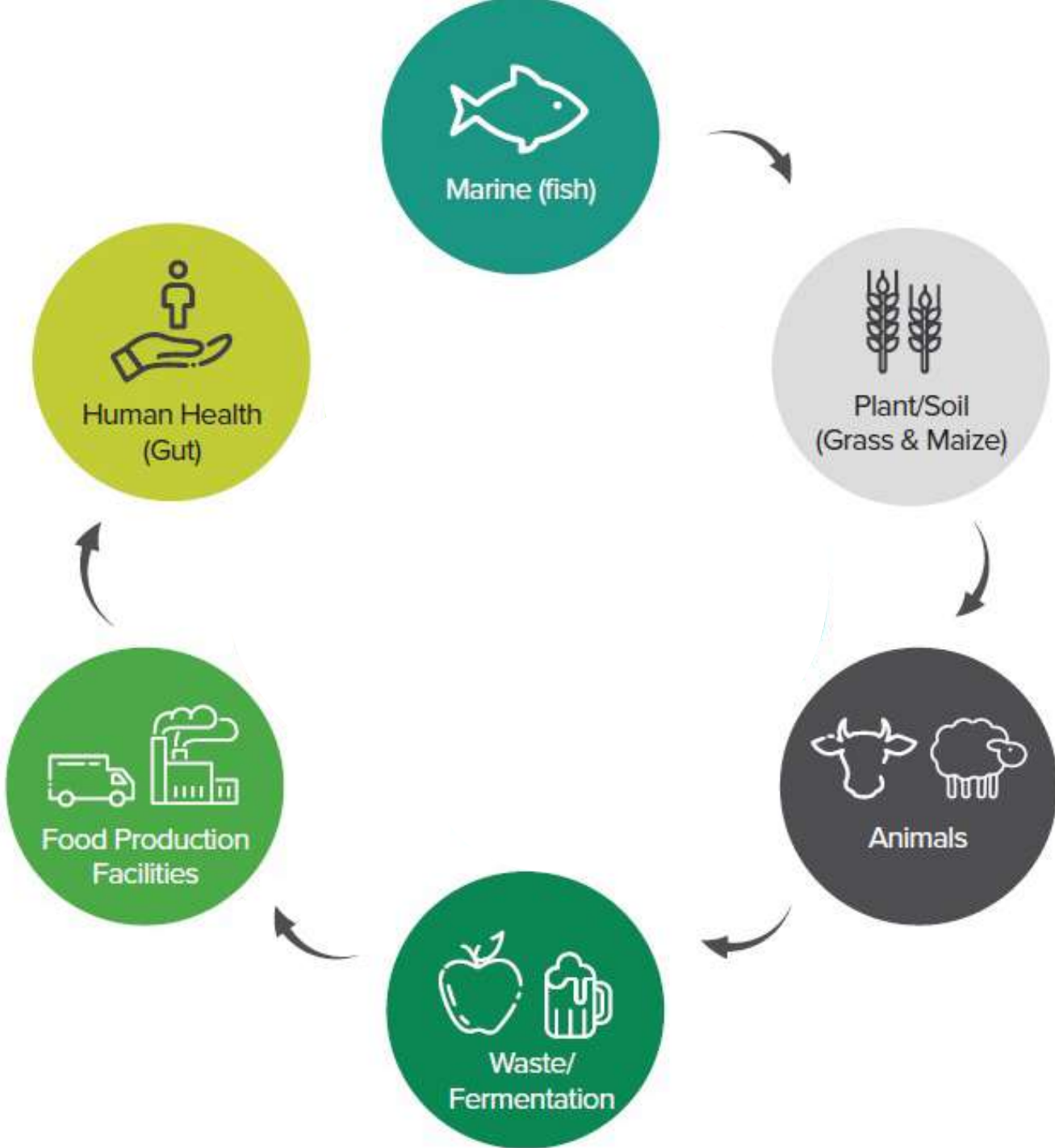


Microbiome Applications for Sustainable Food Systems through Technologies and Enterprise

MASTER takes a global approach to the development of microbiome products, foods/feeds, services or processes with high commercial potential.

This will benefit society through **improving the quantity, quality and safety of food** across multiple food chains. These include marine, plant, soil, rumen, meat, brewing, fruit and vegetable waste, and fermented foods.





Rumen microbiome

WP3: Rumen Microbiome – Improving animal production and reducing environmental impact through manipulation of the rumen microbiome *Lead QUB (Huws), Co-Lead CSIC (Yanez-Ruiz), partners Teagasc, LUKE, INRA, Devenish, ICBF, AFBI,*

Objective:

- Utilise host genomics to alter the rumen microbiome and animal phenotype.
- Define the rumen microbiome (different life stages) to improve efficiency, reduce greenhouse gases emissions & improve the fatty acid content of meat and milk.
- Develop microbiome-based tools to predict animal phenotype.



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Beyond Nutrition



Integrated microbiome technologies

WP5: Integrated Microbiome Technologies for the Food Chain *Lead UNITN (Segata), Co-lead QUB (Creevey), partners WU, Teagasc, UCC, 4DC, ULE, ONT, FFOQSI, Novolyse, Qiagen, Baseclear, Danone in close interaction with WPs 1, 2, 3, 4 and 6*

WP5 will provide the technological, computational, and analytical tools to (i) support the other WPs, (ii) establish standardized tools and procedures for companies in the food chain, (iii) meta-analyse the produced data, and (iv) build user- and company-friendly resources to support all the microbial tasks associated with the food chain.



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UCC
Coláiste na hOllscoile Corcaigh, Éire
University College Cork, Ireland



WP5: Integrated Microbiome Technologies for the Food Chain



SOPs

SOP for validated sample collection and storage

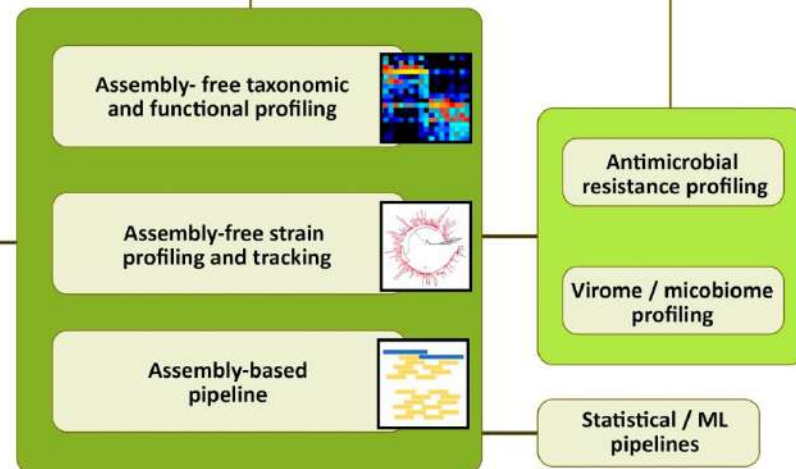
SOP for validated 16S rRNA-based microbiome processing

SOP for validated shotgun metagenomics based microbiome processing

Pipelines/Tools



MASTER and public metagenomes
Pre-processing



WP3: Objectives



3.1: Utilise host genomics to beneficially alter the rumen microbiome and consequently animal phenotype.

3.2: Employ novel feeding technologies to define the rumen microbiome at different stages of life to improve efficiency, reduce greenhouse gas emissions and improve the fatty acid content of meat and milk.

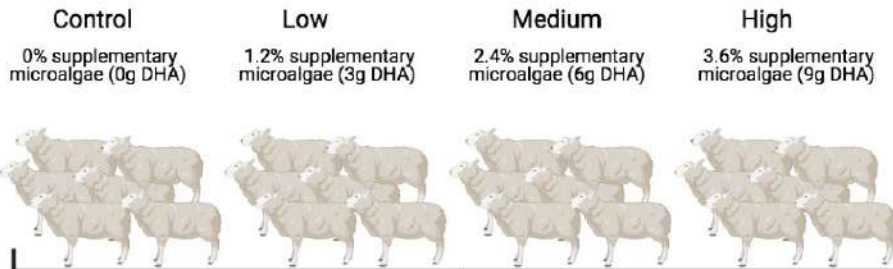
3.3: Develop microbiome-based tools and mathematical models to predict animal phenotype.

3.2: Dietary manipulations of the rumen microbiome for improved animal phenotype.

3.2.1: Feed technologies in adult animals 2) Microalgae (link to WP 2)

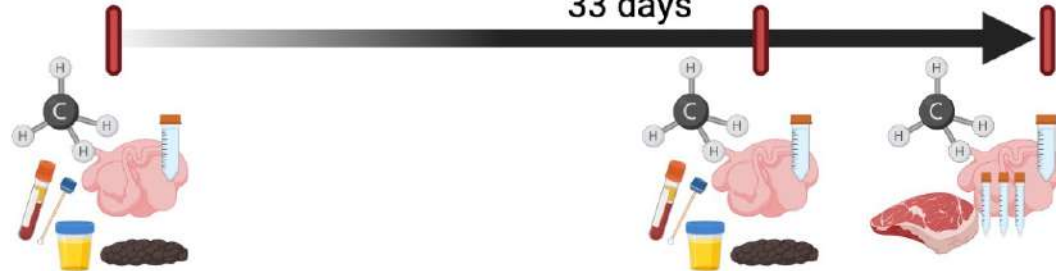
50:50 isoenergetic diet
(grass silage:concentrate)

Microalgae supplementation



56/24 Finishing Texel X Scottish black face lambs

33 days



56 animals

- 24 Chambers trial
- 32 Feed trial

Before and after samples

- Methane
- Oral swab
- Blood
- Ruminal liquid
- Urine (digestibility)
- Faeces (digestibility/ microbiome)

Slaughter samples

- Ruminal liquid
- Small intestine
- Large intestine
- Cecum
- Meat (2 and 7 days): loins, shoulder, legs



3.2: Dietary manipulations of the rumen microbiome for improved animal phenotype.

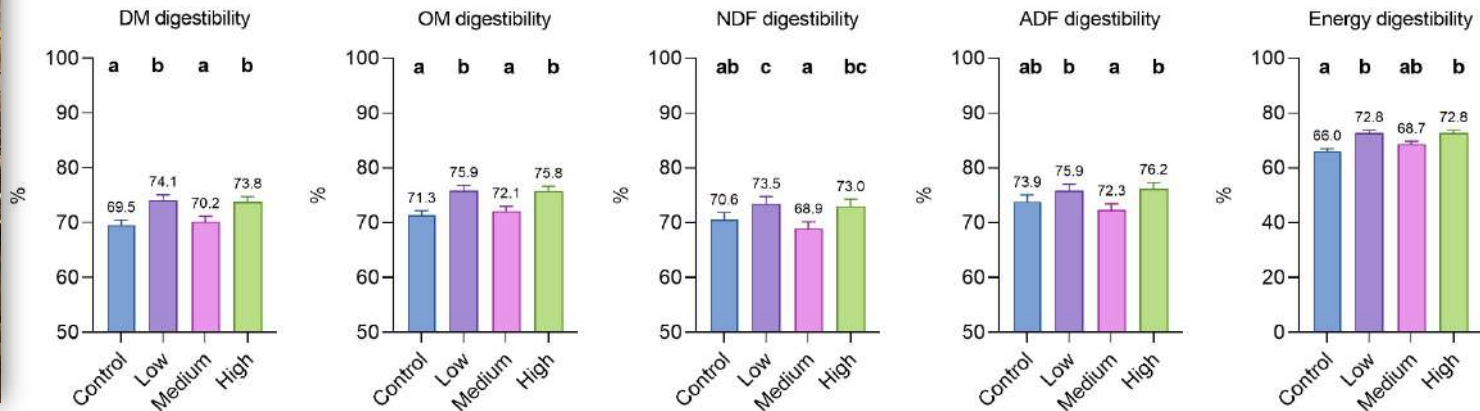
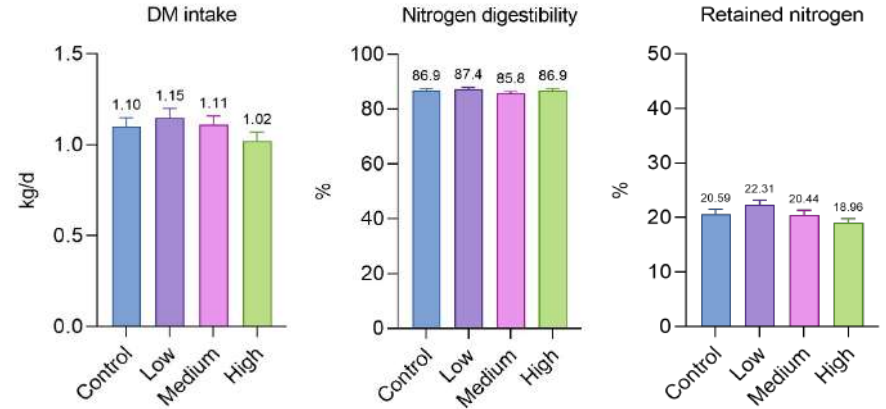


3.2: Dietary manipulations of the rumen microbiome for improved animal phenotype.



Animal Performance, digestibility and methane emission

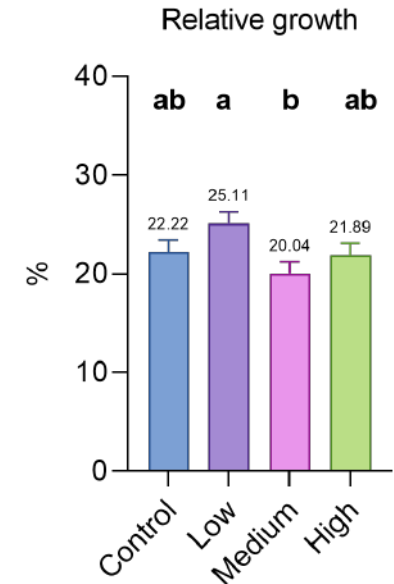
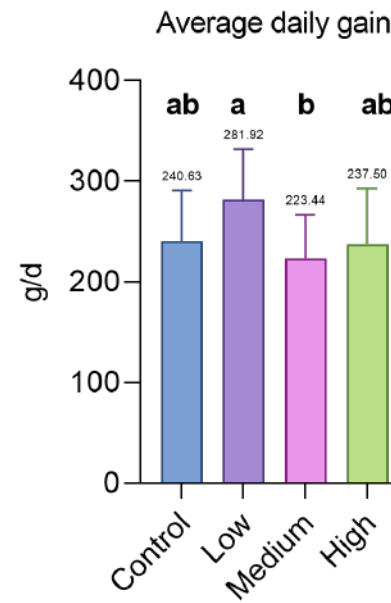
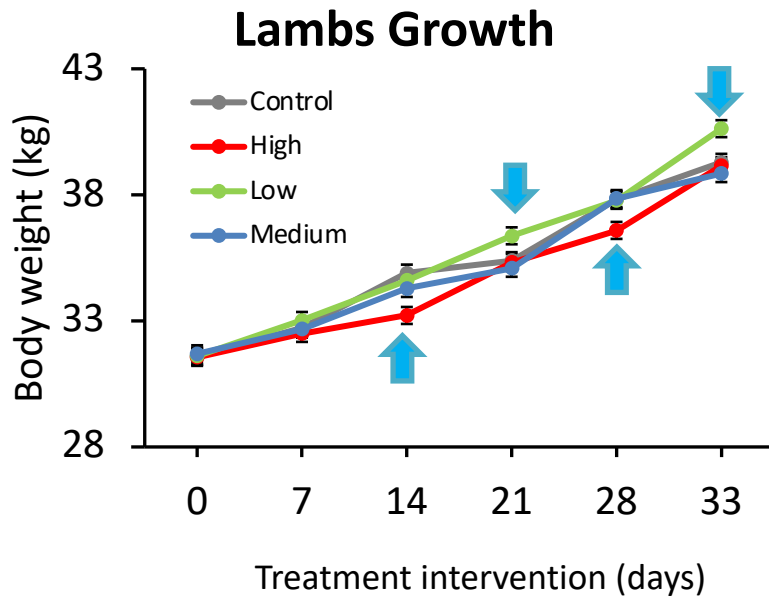
- The DMI tended to decrease ($P=0.078$) in High treatment compared to the other diets ($n=24$).
- Digestibility of DM, OM, NDF, ADF and Energy were greater ($P\leq 0.030$) in Low and High treatment than in Control and Medium treatment, while N digestibility and retention were similar ($P\geq 0.325$) between treatments ($n=24$).



3.2: Dietary manipulations of the rumen microbiome for improved animal phenotype.

Animal Performance, digestibility and methane emission

- No differences ($P \geq 0.453$) between Control and Microalgae-fed lambs were observed for initial body weight (BW) (n=56). Differences in BW were seen at different time points (see arrows below)
- The average daily gain (ADG) was greater ($P=0.020$), and the relative growth tended ($P=0.068$) to be greater in Low than in Medium treatment (n=56).



$P\text{-trt} < 0.001$; $P\text{-tim} < 0.001$; $P\text{-int} = 0.004$

*RG (%) = $(W2 - W1) / ((W1 + W2) / 2) * 100$; W1 = Initial BW, W2 = Final BW

3.2: Dietary manipulations of the rumen microbiome for improved animal phenotype.

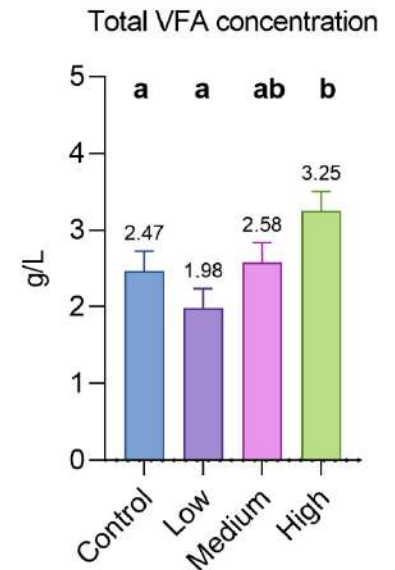
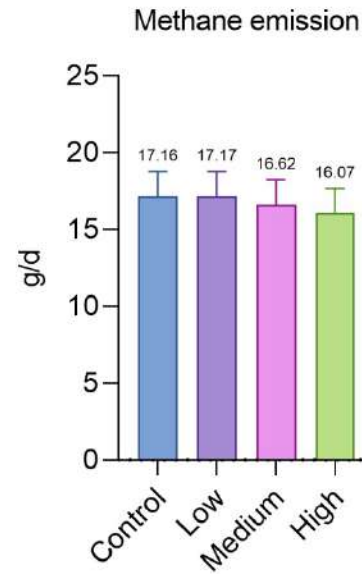
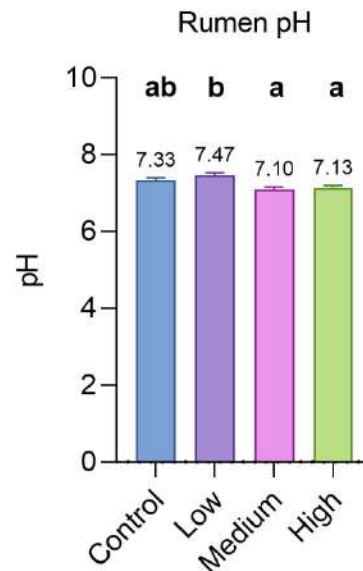


Animal Performance, digestibility and methane emission

Indirect open-circuit respiration calorimeter chambers



- Ruminal pH was lower ($P=0.024$) in Medium and High treatment than in Low treatment.
- Total VFA concentration was greater ($P=0.012$) in High than in Control and Low treatment.
- Methane emissions and methane/DMI were similar ($P\geq 0.143$) between treatments, however an approx. 8% reduction was seen with High treatment.



3.2: Dietary manipulations of the rumen microbiome for improved animal phenotype.

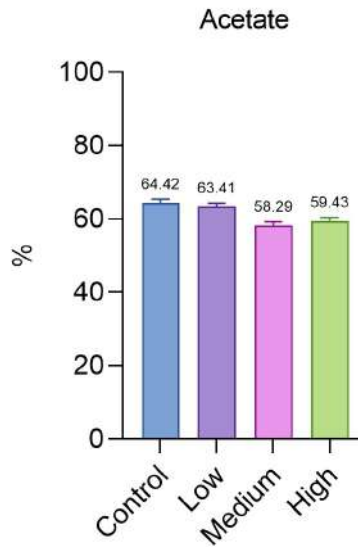


Animal Performance, digestibility and methane emission

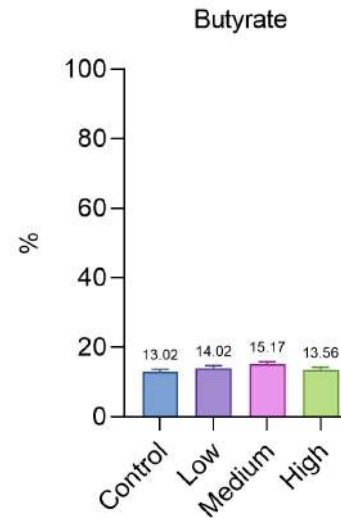
Indirect open-circuit respiration calorimeter chambers



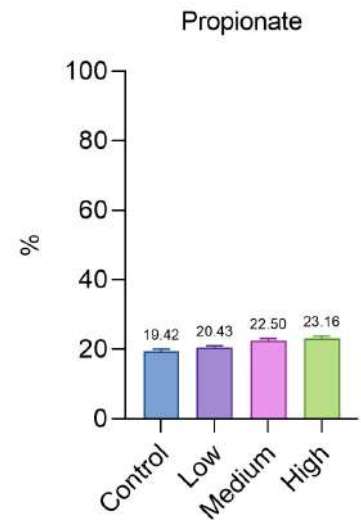
- Tendency for acetate to be lower in lambs fed medium and high levels of microalgae.
- Whilst not significant modest increases in propionate were also seen in lambs fed medium and high levels of microalgae.



P=0.078



P=0.157



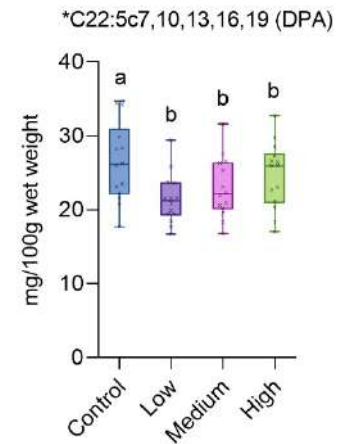
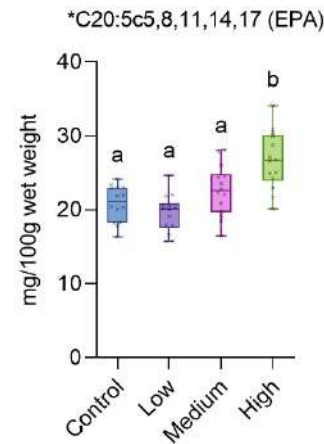
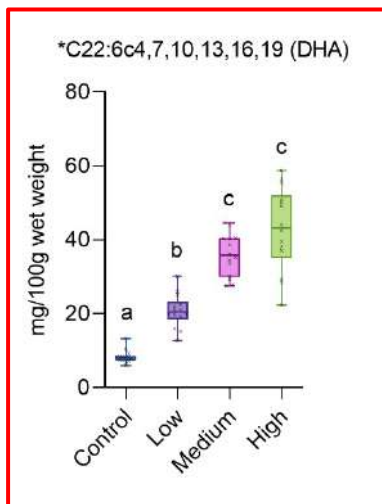
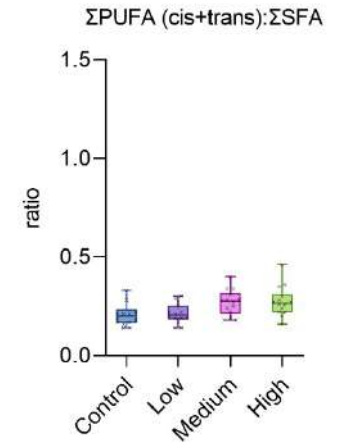
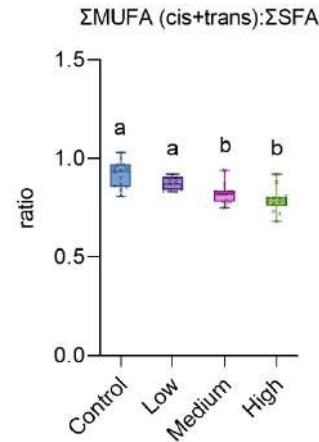
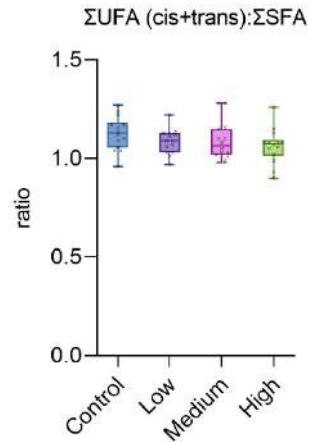
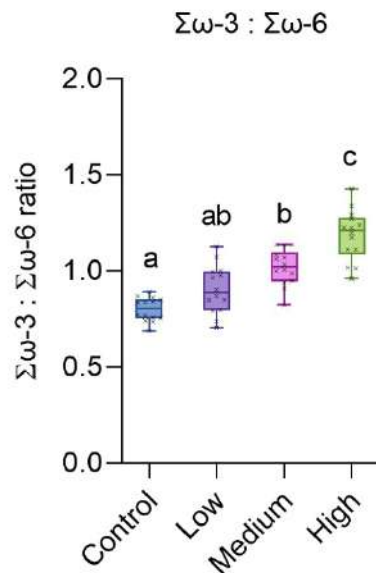
P=0.533

3.2: Dietary manipulations of the rumen microbiome for improved animal phenotype.



Meat quality and consumption

- DHA concentration increased in the **loins** as the microalgae content on the lamb's diet increased ($P < 0.05$).



* $P < 0.05$

3.2: Dietary manipulations of the rumen microbiome for improved animal phenotype.



Meat quality and consumption

- Microbiology testing (food standards):**

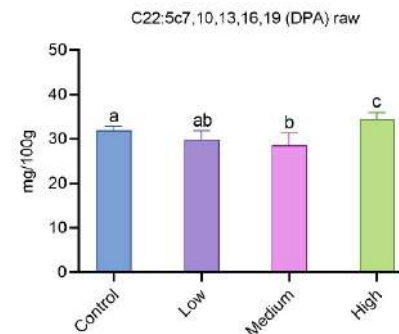
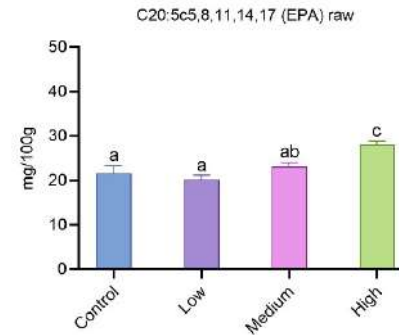
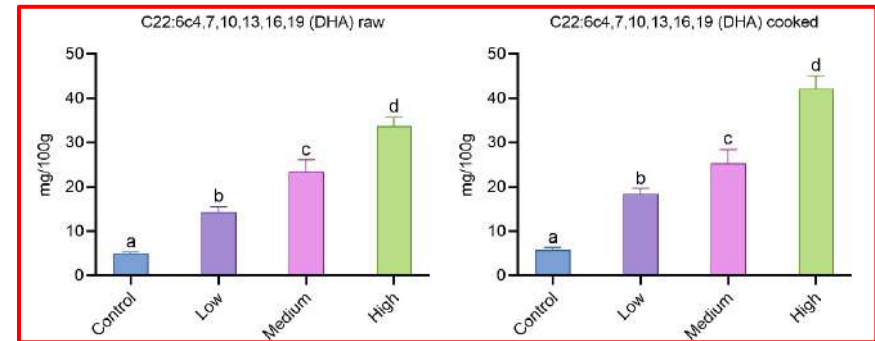
ALS: Presumptive Coliforms; *Escherichia coli*; Thermotolerant *Campylobacter*; Aerobic colony count; β -Glucuronidase + *E. coli*; *Salmonella*; *Listeria* spp.

- Fatty acid concentration increased during the cooking (water loss of 37.8%)



Home-sensorial trial in course

Raw and cooked fatty acid of legs and shoulder patties (50:50%)



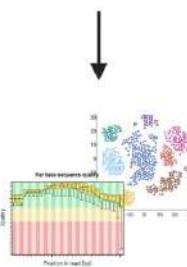
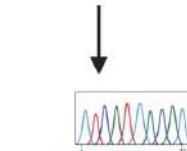
*P < 0.05

3.2: Dietary manipulations of the rumen microbiome for improved animal phenotype.



Animal microbiome and ruminal fermentation

Shotgun metagenomic sequencing



Illumina Nova seq 6000 S4 300 flow cell
(150bp PEat ~>6.2GB/ sample).

- The average number of reads per sample after quality control was 23,553,087
- A total of 4712 metagenome associated genomes (MAGs) were extracted and characterized, functionally and taxonomically with an average number of 1,955 ORFs and average N50 metric was 23,937.

Abundance percentages across samples of the most abundant genera

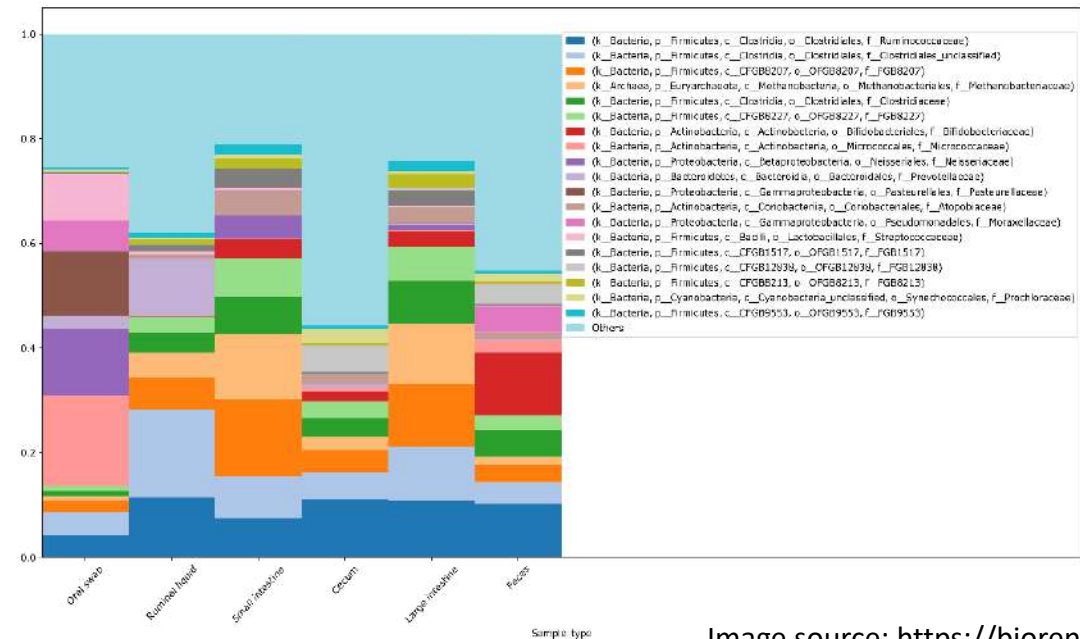
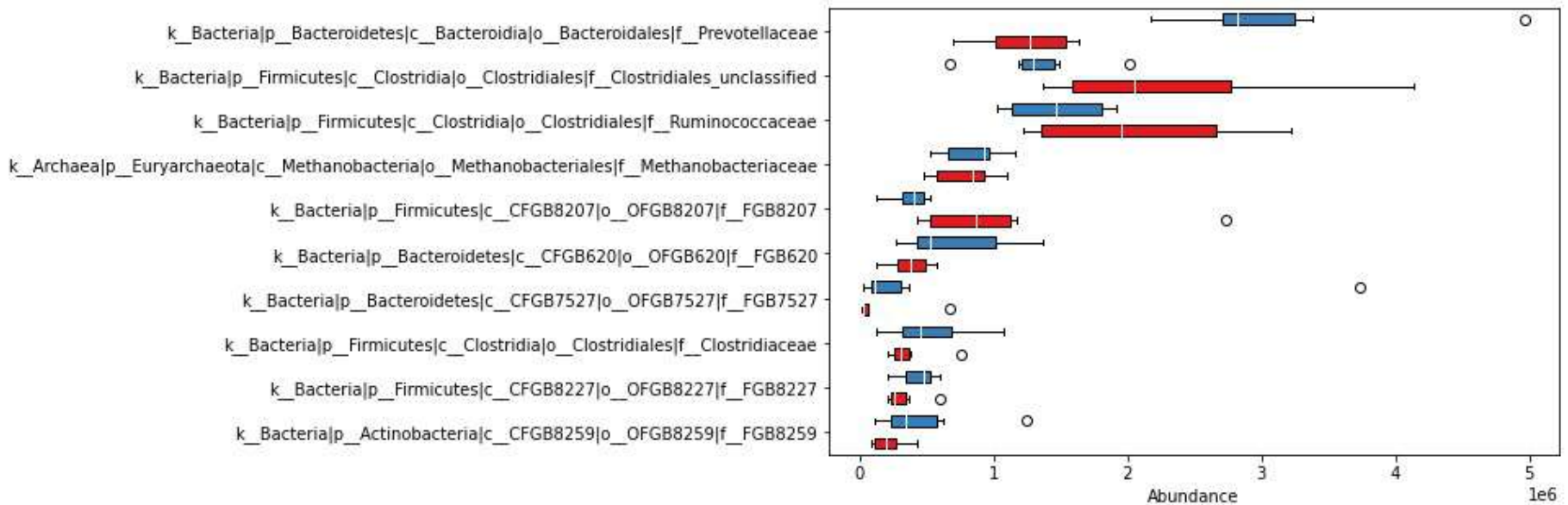


Image source: <https://biorender.com>

3.2: Dietary manipulations of the rumen microbiome for improved animal phenotype.



Animal microbiome and ruminal fermentation



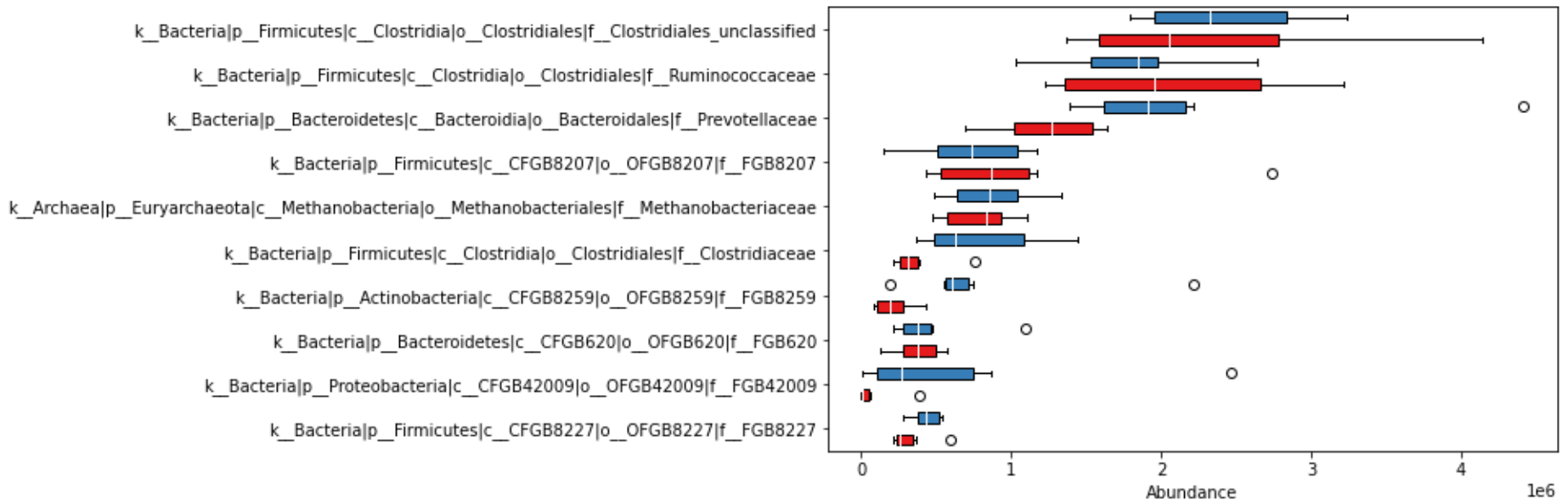
No differences seen in Archaea

■ Control treatment ■ Medium treatment

3.2: Dietary manipulations of the rumen microbiome for improved animal phenotype.



Animal microbiome and ruminal fermentation



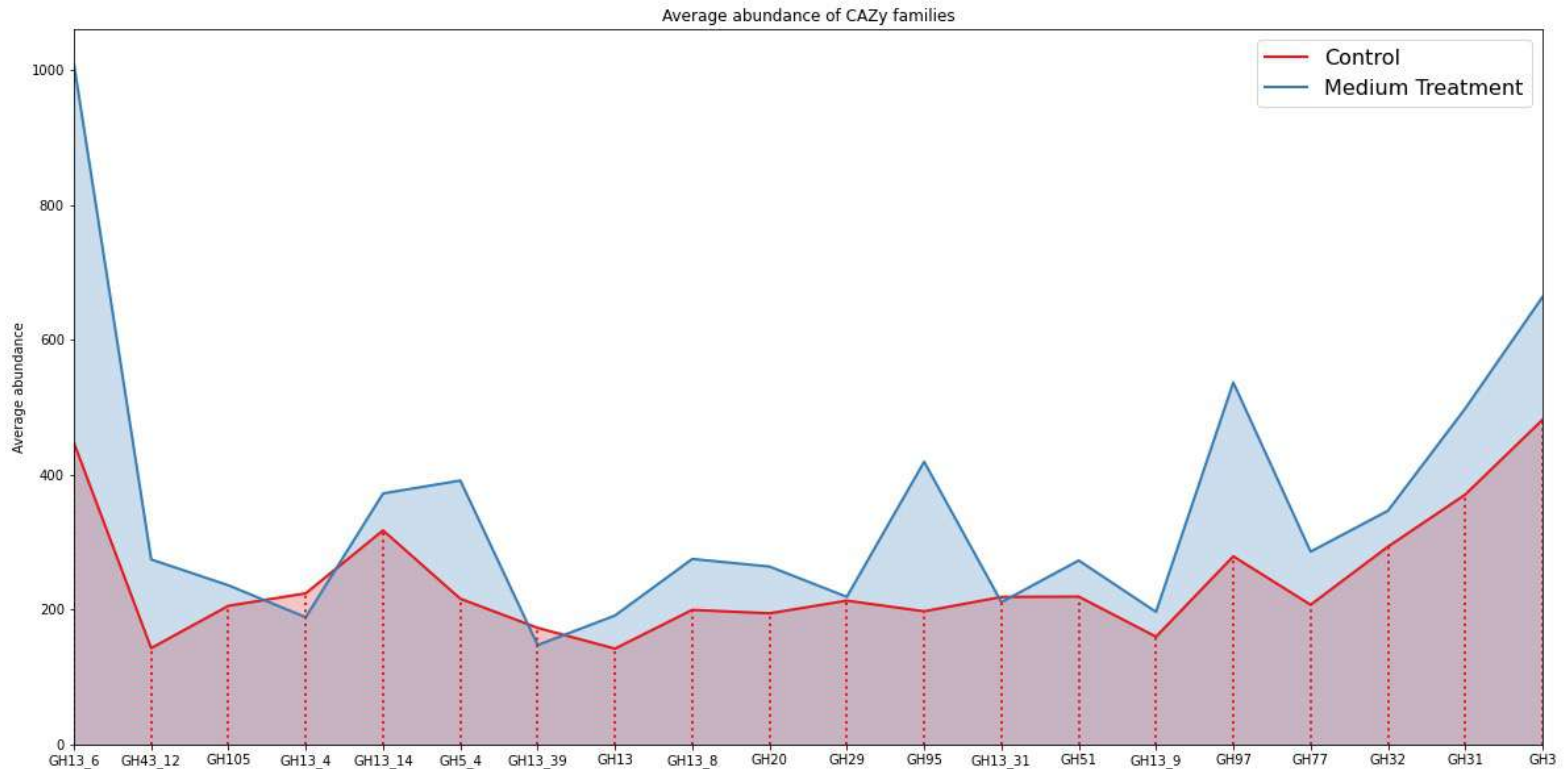
No differences seen in Archaea

Control treatment High treatment

3.2: Dietary manipulations of the rumen microbiome for improved animal phenotype.



CAZyme abundances

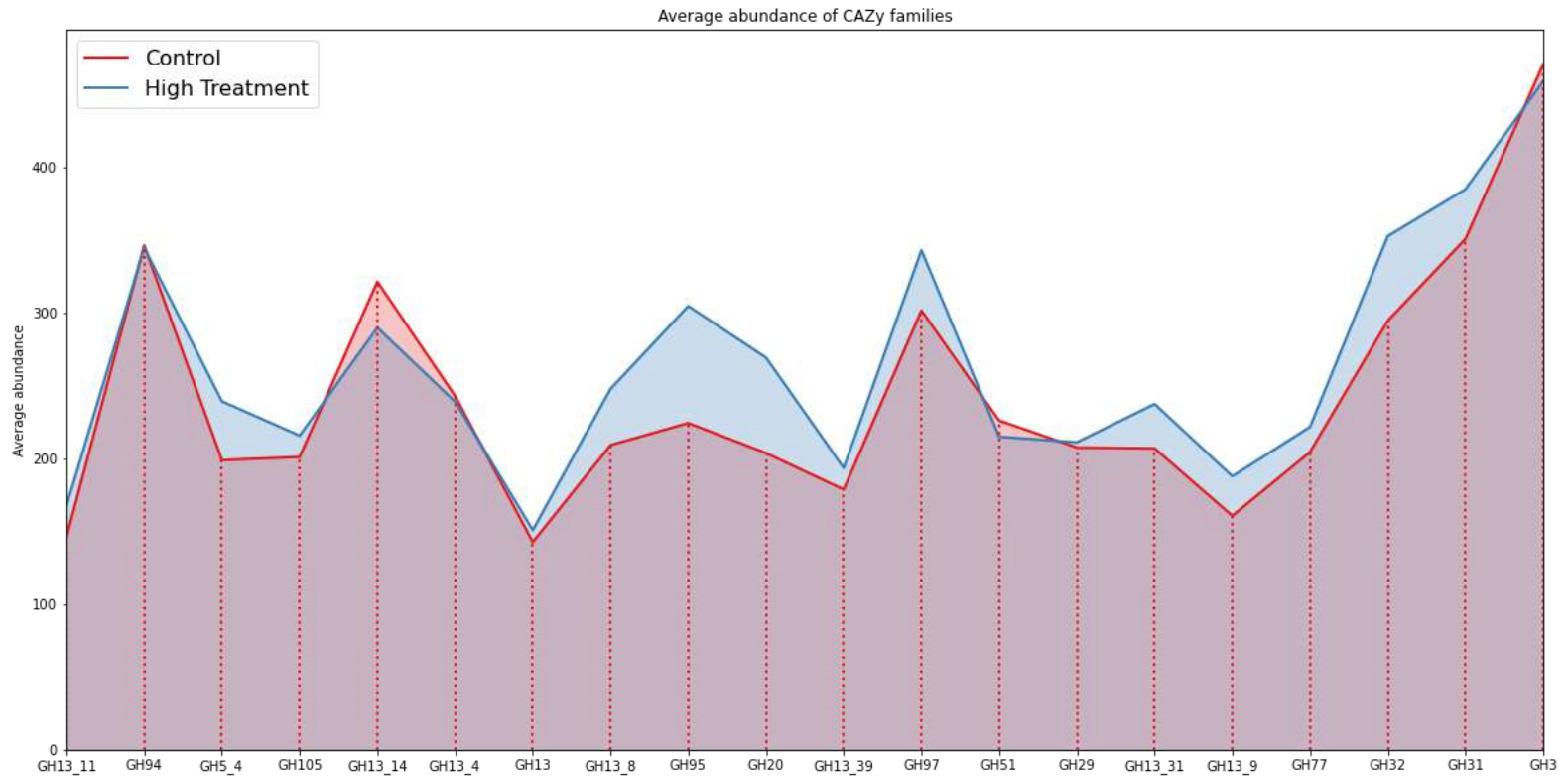


Higher abundances of many of these enzymes families when lambs fed medium levels of microalgae

3.2: Dietary manipulations of the rumen microbiome for improved animal phenotype.



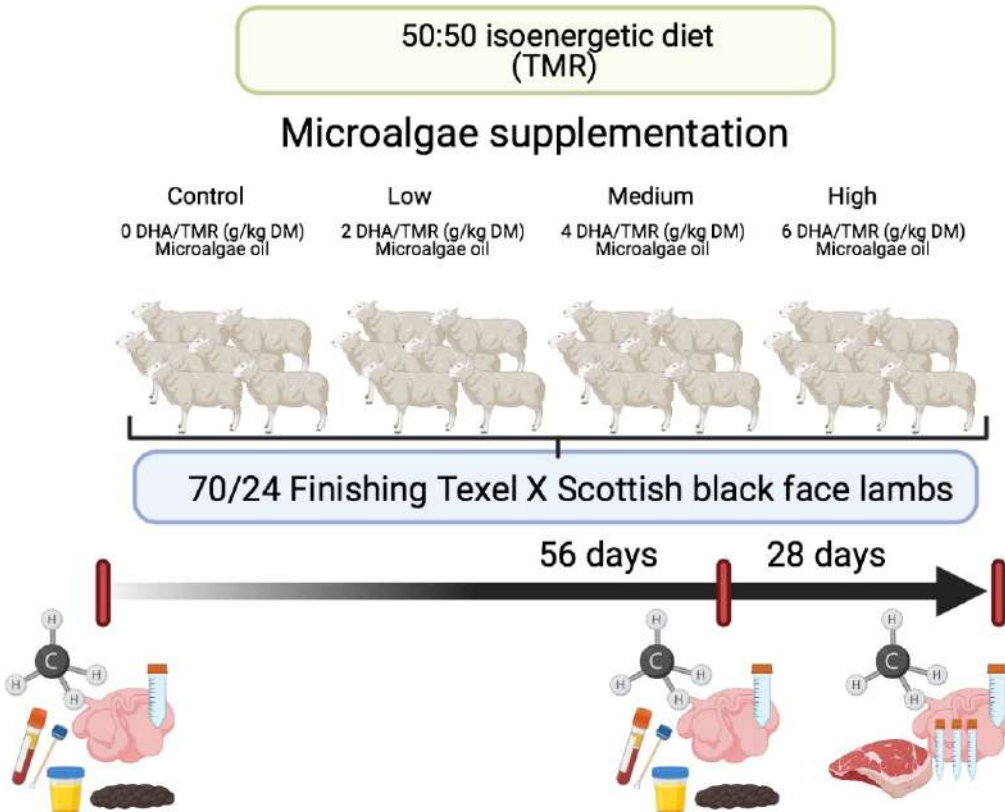
CAZyme abundances



Higher abundances of some of these enzymes families when lambs fed high levels of microalgae

3.2: Dietary manipulations of the rumen microbiome for improved animal phenotype.

3.2.1: Feed technologies in adult animals 2) **Microalgae oil** (link to WP 2)



68 animals

24 Chambers trial

44 Feed trial

Before and after samples

- Methane
- Oral swab
- Blood
- Ruminal liquid
- Urine (digestibility)
- Faeces (digestibility/ microbiome)
- Diet (fatty acid/microbiome)

Slaughter samples

- Ruminal liquid
- Small intestine
- Large intestine
- Cecum
- Meat (2 and 7 days): loins, shoulder, legs

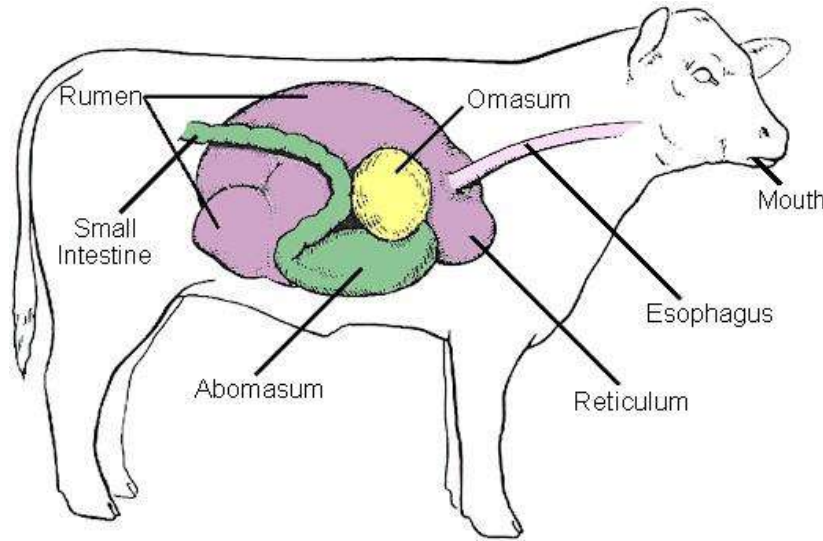
Conclusion

- Feeding freeze-dried microalgae at low, medium and high levels had **no detrimental affect of animal health.**
- Feeding freeze-dried microalgae at low, medium and high levels **significantly increased DHA content of meat and burgers** (pre and post cooking) with a small reduction in methane emissions (8%) seen following feeding on 4.5g/day of the microalgae (High)- **human health benefits.**
- **Modest reductions seen in methane emissions (8%)** likely due to changes in the bacterial fraction leading to the tendencies towards beneficial volatile fatty acid changes.
- **No major effects on animal productivity.**
- Gastrointestinal tract microbiome analysis ongoing with initial interesting data.
- Microalgae oil experiments ongoing.

Ruminant Sector Challenges



Production



Health



Product Quality



Environment



Acknowledgements



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Dr Tianhai Yan, AFBI
Dr Aurelie Aubry, QUB

The MASTER Consortium



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Thank you



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