## Sharon Huws

Ruminant Methane Mitigation Conference

Art of the possible by 2030 and beyond

#### #MethaneBFS23

## The Rumen and it's Future

## **Professor Sharon Huws**

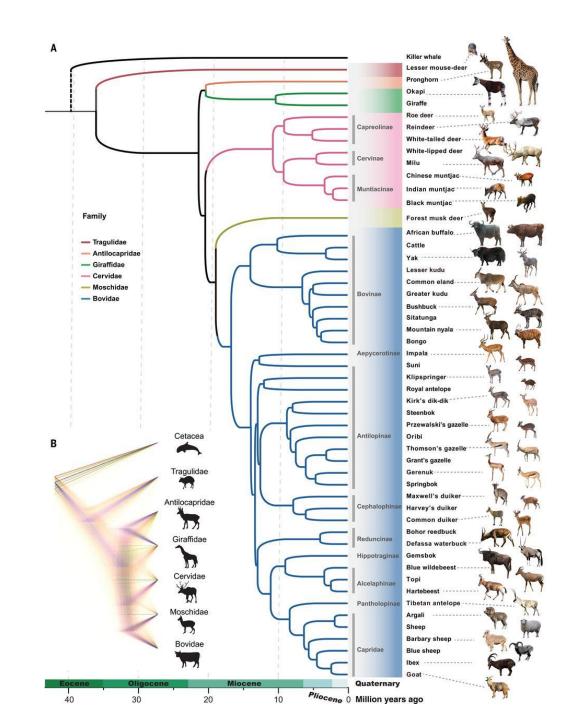


# Ruminants and their Evolution

- The first ruminants evolved about 50 million years ago and were small forest-dwelling omnivores.
- We now have almost 200 living ruminant species in 6 families: Tragulidae, Antilocapridae, Giraffidae, Moschidae, Cervidae, and Bovidae.
- Most species reside within the family Bovidae, with at least 143 species, which includes important livestock animals

Hackmann TJ, Spain JN. J Dairy Sci. 2010 Apr;93(4):1320-34. doi: 10.3168/jds.2009-2071.

Chen et al. Science. 2019 Jun 21;364(6446):eaav6202. doi: 10.1126/science.aav6202. PMID: 31221828.

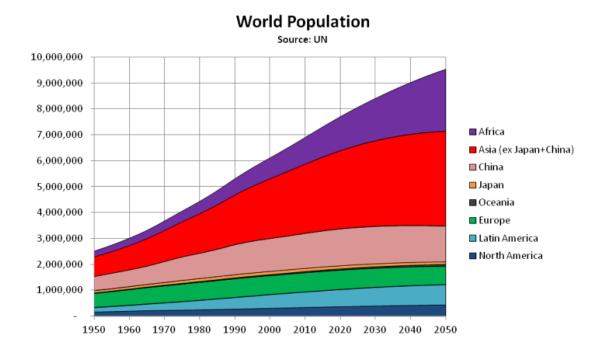


#### Ruminant Food Security challenges due to the Growing Human Population

FAO predict World population will increase to 10.4 billion by 2050.

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

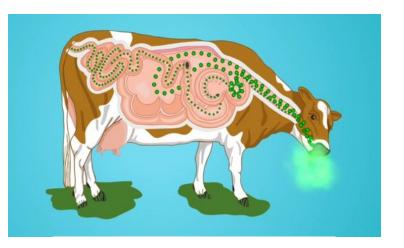
Set against the need to address climate change



Graphic:: deconstructingrisk.com

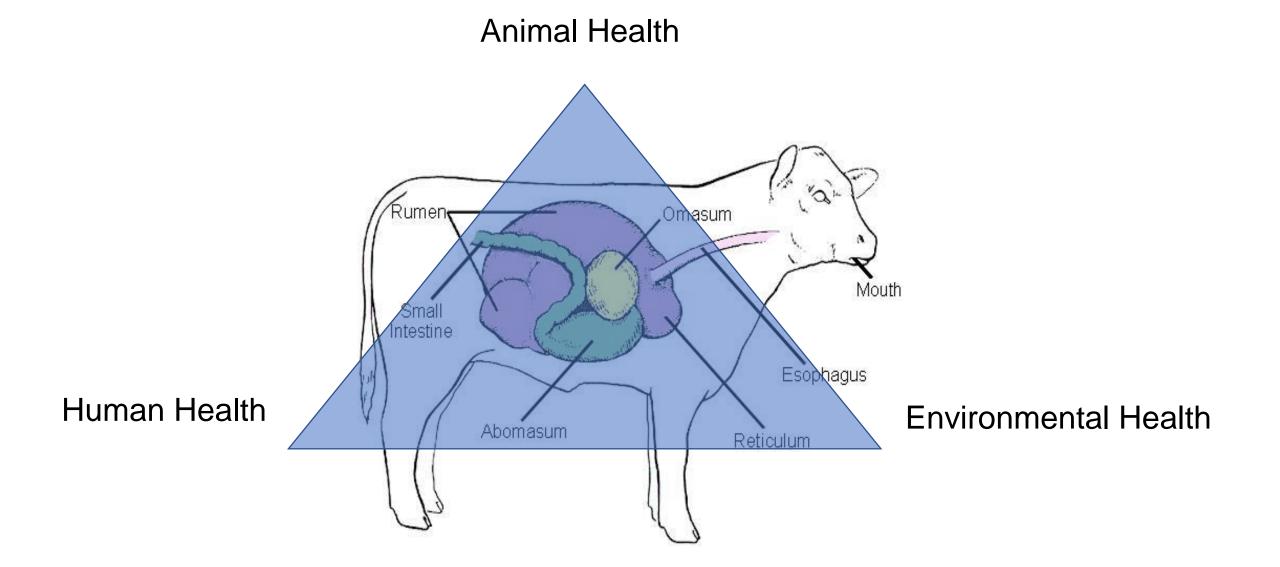
#### Ruminant Food Security and Environmental Health Challenges

- FAO state that livestock agriculture responsible for approx. 18% of GHG emissions globally, 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.
- Many countries aim for net zero by 2050. Northern Ireland bill aims to reduce methane emissions by 46%.
- Data suggests Global Warming Potential (GWP) methane much less than predicted.

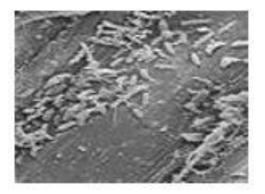




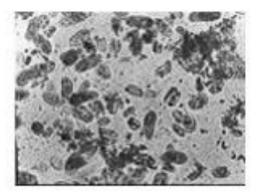
#### Rumen Microbiome Central for Planetary Health



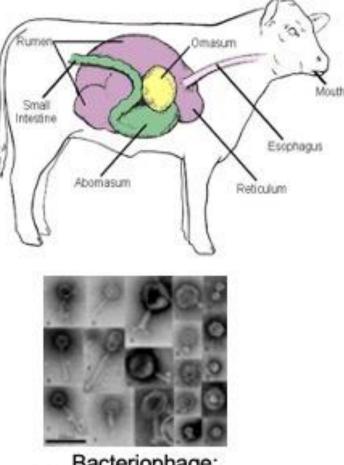
### The Rumen Microbiome

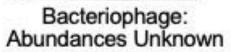


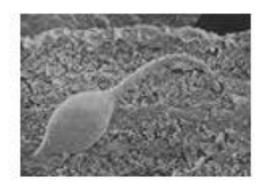
Bacteria: 109-1010/mL



Protozoa: 104-106/mL





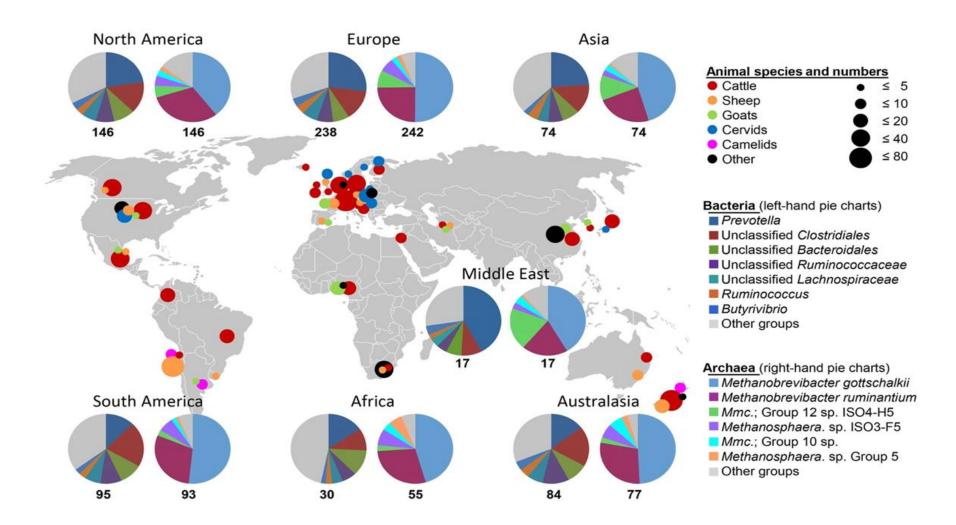


Fungi: 103-104/mL



Archaea (methanogens) :104/mL

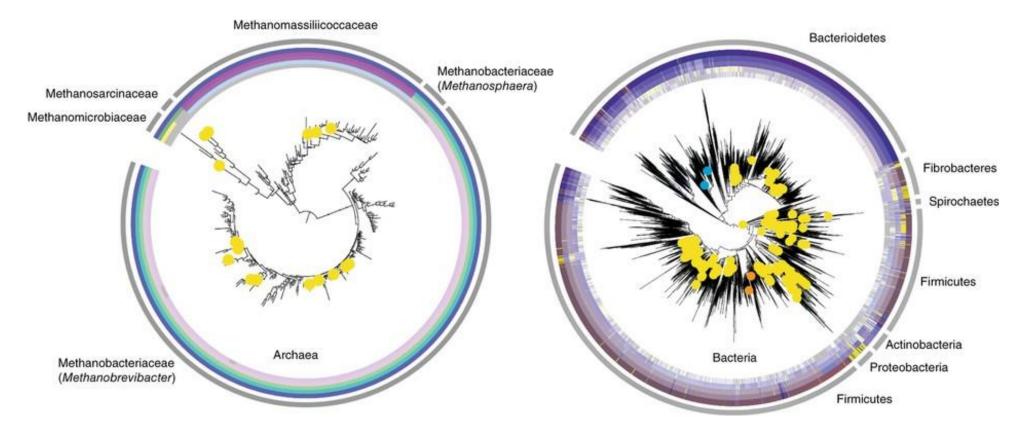
#### Recent Advances in Bacteria and Archaea: Global rumen Census



Henderson et al. Sci Rep. 2016;6:19175.

## Recent Advances in Bacteria and Archaea: Hungate Collection

Hungate Collection: 410 bacteria and archaeal genomes sequenced: But many gaps to be filled



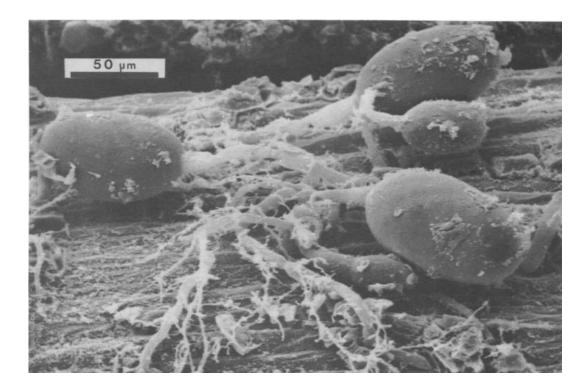
Seshadri et al. 2018 Nat Biotechnol. 36(4):359-367. doi: 10.1038/nbt.4110.

## Recent Advances: Rumen Fungi

- Play important role in cellulose breakdown.
- One class (Neocallimastigomycetes), one order (Neocallimastigales) and four families described :

Anaeromycetaceae, Caecomycetaceae, Neocallimastigaceae and Piromycetaceae

• Produce hydrogen so rumen archaea associate closely with them.



Hess et al. 2020 Oct 21;11:584893. doi: 10.3389/fmicb.2020.584893.

### Recent Advances: Rumen Fungi

#### Number of genomes available in 2020:

Organism	Strain	Host	Genome size <sup>a</sup> [base pairs]	Gene count <sup>a</sup>	CAZyme count <sup>a</sup>	References
Anaeromyces robustus	S4	Sheep	71,685,009	12,832	1,766	Haitjema et al. (2017)
Caecomyces churrovis	<del></del>	Sheep	165,495,782	15,009	ND <sup>b</sup>	Henske et al. (2017)
Neocallimastix californiae	G1	Goat	193,495,782	20,219	2,743	Haitjema et al. (2017)
Pecoramyces ruminantium	C1A	Cow	100,954,185	18,936	2,029	Youssef et al. (2013)
(formerly Orpinomyces sp.)						
Piromyces finnis	Pirfi3	Horse	56,455,805	10,992	1,463	Haitjema et al. (2017)
Piromyces sp.	E2	Elephant	71,019,055	14,648	3,819	Haitjema et al. (2017)

<sup>a</sup>https://mycocosm.jgi.doe.gov (Grigoriev et al., 2013). <sup>b</sup>Not determined.

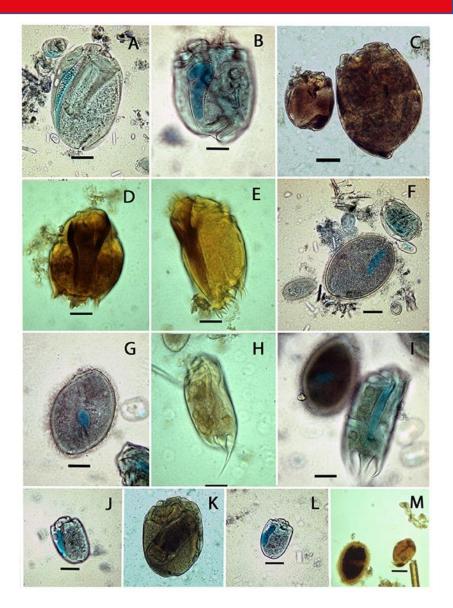
#### Now expanded to 14!

Hess et al. 2020 Oct 21;11:584893. doi: 10.3389/fmicb.2020.584893.

### Recent Advances: Rumen Protozoa

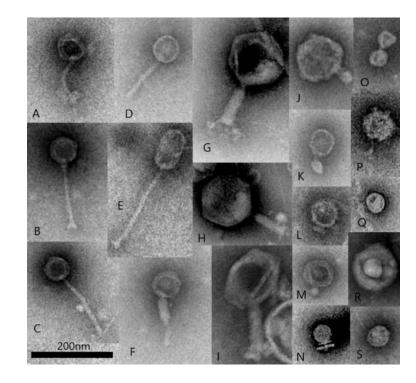
- Two groups: Entodinomorphids and Holotrichs.
- Entodinomorphs have cellulolytic capacity, whereas holotrichs utlise simple sugars.
- Possess micro and macronucleus (macronucleus contains genomic information).
- Possess hydrogenosomes: Generate H<sub>2</sub> causing methanogens to strongly associate with them.
- Cannot be grown for long periods outside the rumen.
- Can be removed from the rumen by defaunation.
- Removal reduces methane emissions by 11% and can increase average daily gain.

Williams et al. Front Microbiol. 2020 Apr 29;11:720. doi: 10.3389/fmicb.2020.00720. Newbold et al. Front Microbiol. 2015 Nov 26;6:1313. doi: 10.3389/fmicb.2015.01313.



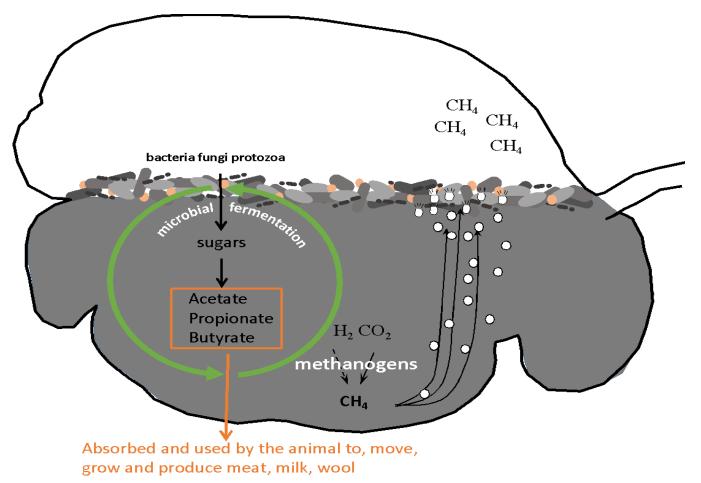
### Recent Advances: Bacteriophages

- Bacteriophages are viruses which infect bacteria.
- They can either integrate into the host genome or lyse their target bacteria.
- Therefore, they can affect the bacterial community substantially – effects on function and animal phenotype?

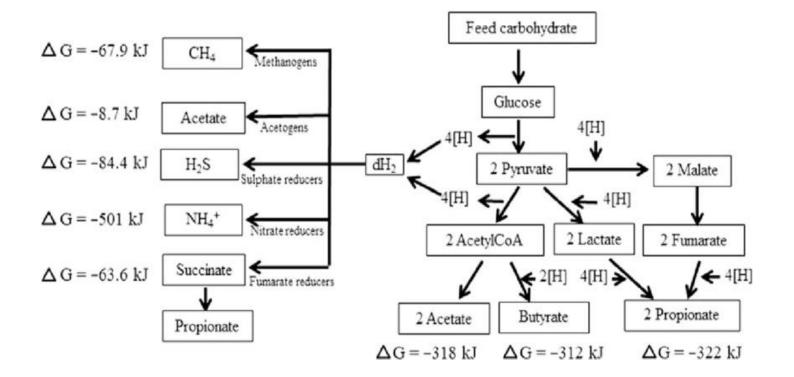


#### Role of these Microbes in Rumen Biochemistry

- Cellulose/hemicellulose to Volatile fatty acids (source of energy).
- Volatile fatty acids: Acetate, lactate, butyrate, succinate, propionate
- VFAs provide energy source but their production causes release of H.
- Results in methanogenesis and CH<sub>4</sub> release.



### **Rumen Hydrogen Dynamics**



Some VFAs are H sinks:

Pyruvate to lactate: utilizes 4H Lactate to propionate: utilizes 4H Acetyl-CoA to butyrate : utilizes 2H

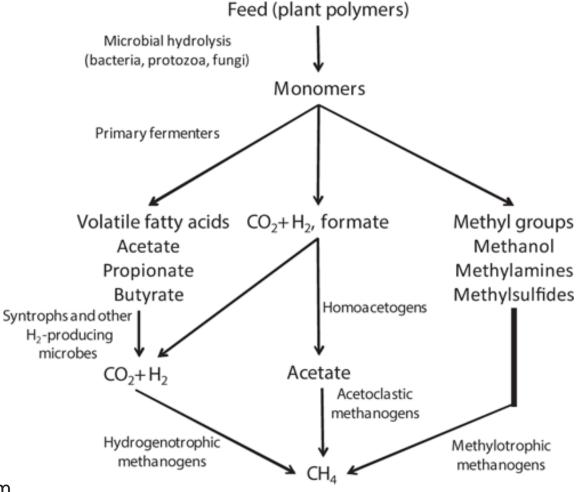
Beauchemin KA, Ungerfeld EM, Eckard RJ, Wang M. Review: Animal. 2020 Mar;14(S1):s2-s16. doi: 10.1017/S1751731119003100.

## Pathways to Rumen Methane Generation

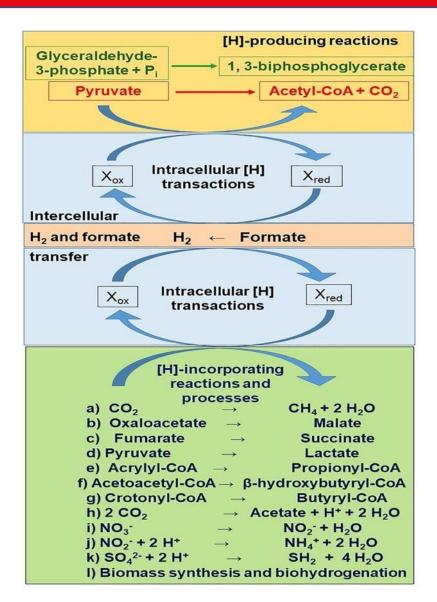
- Methane is produced in the rumen through 3 potential biochemical pathways:
- Hydrogenotrophic pathway (most dominant – catlysed by McrA enzyme)
- Methylotrophic pathway

Acetoclastic pathway

Pereira AM, de Lurdes Nunes Enes Dapkevicius M, Borba AES. Anim Microbiome. 2022 Jan 6;4(1):5. doi: 10.1186/s42523-021-00153-w.



### Formate Production Important?

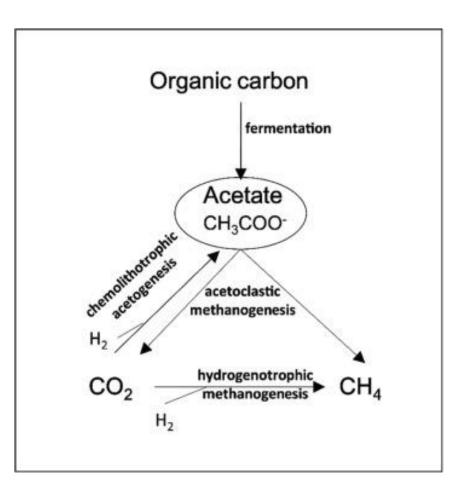


- Formate is another VFA formed in the rumen (approx. 10% of total VFAs).
- 18% of rumen methane is also derived from formate.
- Formate can be easily utilised by methanogens to produce CH<sub>4</sub> directly, or degraded to CO<sub>2</sub> and H<sub>2</sub> and then converted into CH<sub>4</sub> by methanogens.
- Reducing formate production a good strategy?

Hungate et al. J Bacteriol. 1970 May;102(2):389-97. doi: 10.1128/jb.102.2.389-397. He et al.. Animal. 2019 Jan;13(1):90-97. doi: 10.1017/S1751731118000691.

## Reductive Acetogenesis in the Rumen

- Kangaroos produce little methane due to the action of their reductive acetogens, which capture H<sub>2</sub> to produce acetate, thereby taking it away from methanogenesis pathways.
- Reductive rumen acetogens can capture up to 26%
   H2 to make acetate resulting in re-direction energy.
- However, methanogenesis is thermodynamically favoured and reductive acetogenesis only occurs when methanogenesis is low in occurrence



Li et al. Front Microbiol. 2020 Jun 30;11:1311. doi: 10.3389/fmicb.2020.01311.

#### Language Advances: Electron Transfer

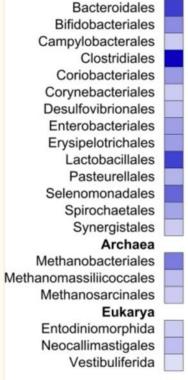
- At a more detailed level, rumen fermentation involves a series of reactions, where electrons are released during oxidation of substrates.
- Microbes then dispose of these electrons via production of the fermentation intermediates lactate, ethanol, and/or VFAs.
- Protons (H<sup>+</sup>) and CO<sub>2</sub> also serve as terminal electron acceptors and are reduced to H<sub>2</sub> and formate, respectively and so act as external electron carriers.

Leahy et al. Trends Microbiol. 2022 Mar;30(3):209-212. doi: 10.1016/j.tim.2021.12.005.

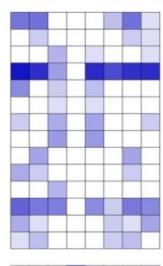
## **Electron transfer**

- Electron transfer via H<sub>2</sub> formation is catalysed by hydrogenase enzymes.
- Recent study showed many rumen bacteria posses hydrogenases and some methanogens too.
- Controlling hydrogenase activity may enable control of H<sub>2</sub> production and therefore methane production.

Greening et al. ISME J. 2019 Oct;13(10):2617-2632. doi: 10.1038/s41396-019-0464-2.

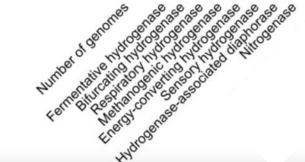


Bacteria









# Future Advancements in the Understanding of the Rumen Microbiome

Current/Ongoing projects (national and international):

- DAERA Excellence & Innovation funding
- MethAbate
- SUREFoodi co-centre
- Holoruminant
- Foundation for Food and Agriculture Research (FFAR)
- RUMEN Gateway











# Global Research Alliance Flagship Project RUMEN GATEWAY



#### BACKGROUND

#### Culture Collection Challenge

Further understanding and manipulation of the rumen microbiome are hindered by poor culture collections and a lack of effort placed on culturing.



#### Hungate Collection

The Hungate collection project sequenced 410 rumen microbes (2018), providing a step-change in our understanding of the rumen microbiome.





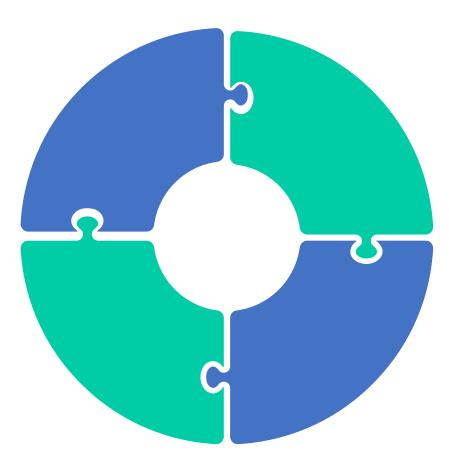
#### WHAT DO WE GAIN?

#### **Rumen function**

Ability to understand rumen microbial function

#### **Feed interaction**

Mechanistic understanding of the action of feed interventions to mitigate methane



#### Hinder methanogensis

Provision of potential direct-fed microbials to redirect hydrogen away from methanogenesis

#### Discovery

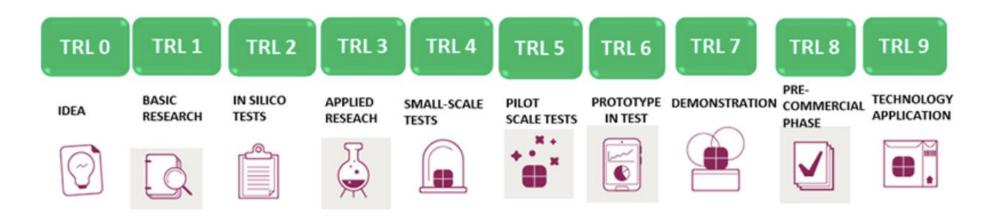
Microbial resource availability for bioactive compound discovery.





## Summary

- Rumen microbes and their function is complicated with much remaining unknown.
- Fundamental understanding of rumen microbes and their function will lead to targeted innovative strategies to reduce methane and enhance sustainable production e.g Bovaer developed from structural information on McrA gene.
- The global RUMEN Gateway project will enhance our fundamental understanding which will aid innovation, such as development of methane vaccines etc.



## Thank you for Listening

and

# Welcome to Queen's University Belfast

