

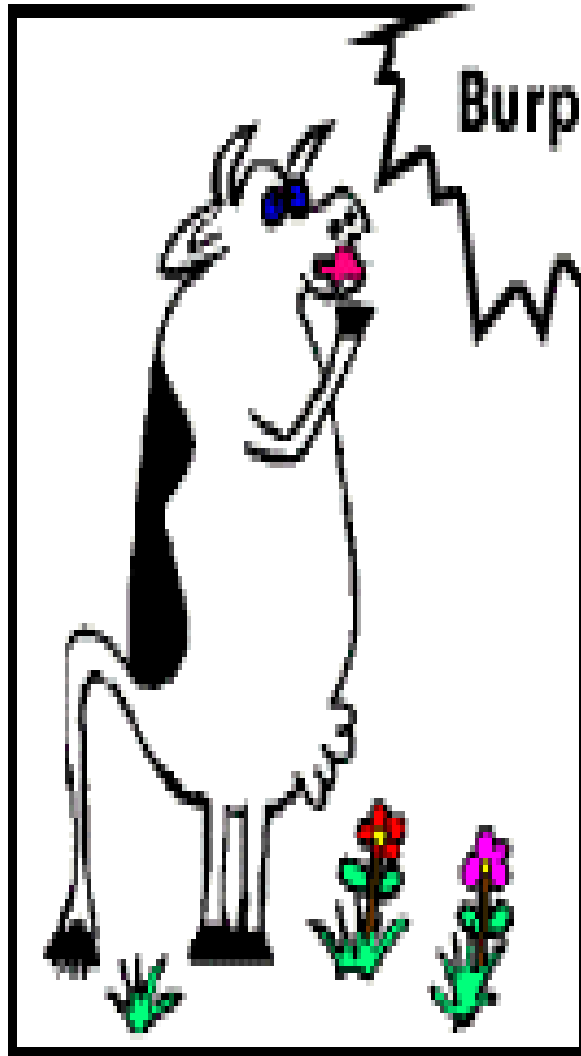
Redressing the balance - "less belching cows"

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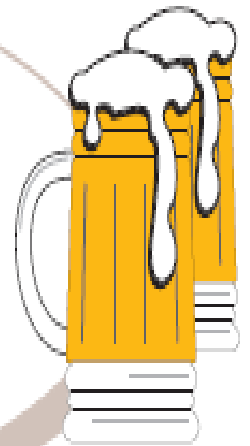
- ❖ Average human output 500- 1500 ml/ d
- ❖ Only 50% of people produce methane
- ❖ From 10 to 50 % methane
- ❖ 50 – 750 ml of methane per day



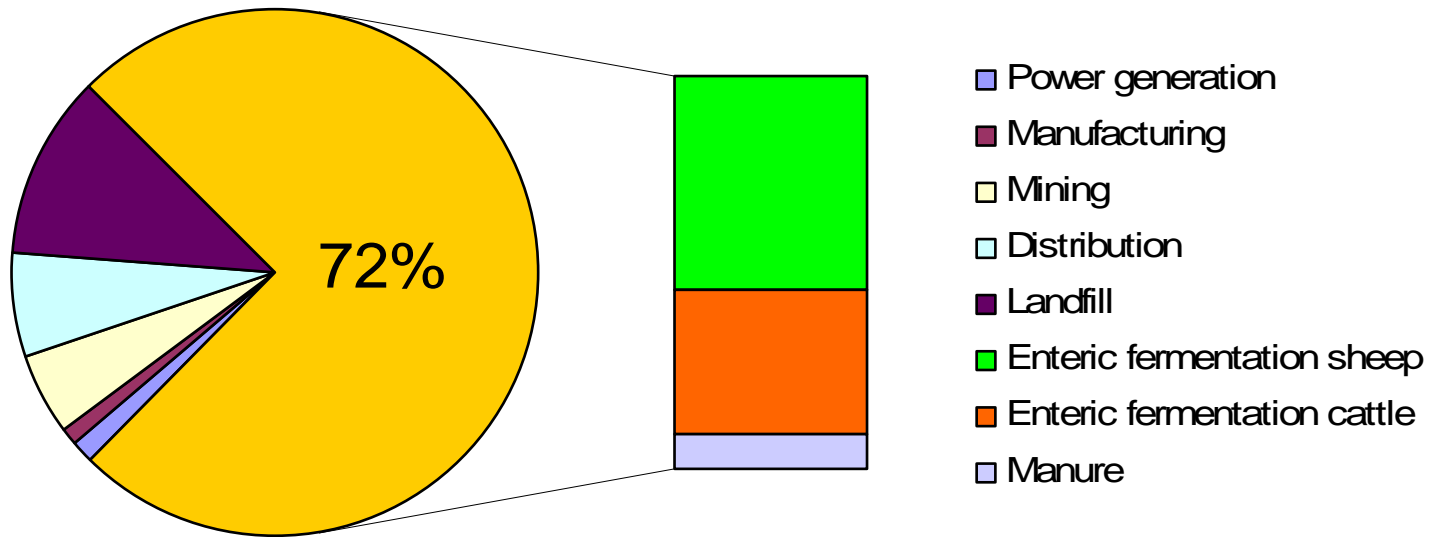


One cow can produce
between 500 and 600 litres of
methane per day

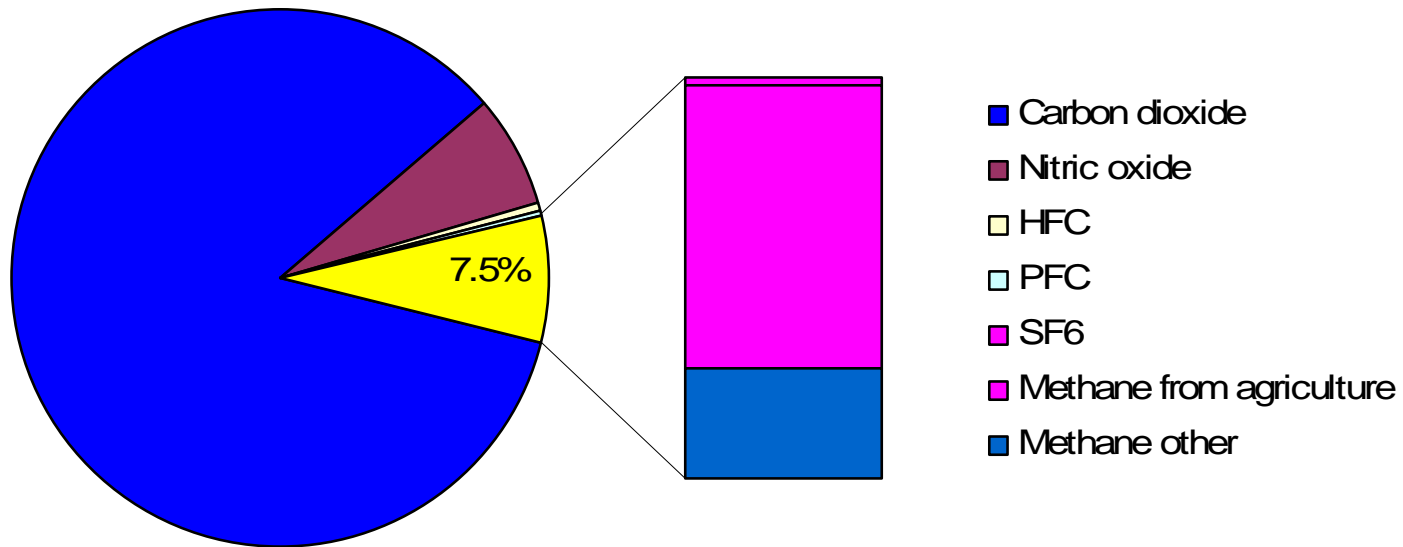
The equivalent to almost 1000
pints of beer



Sources of methane in Wales



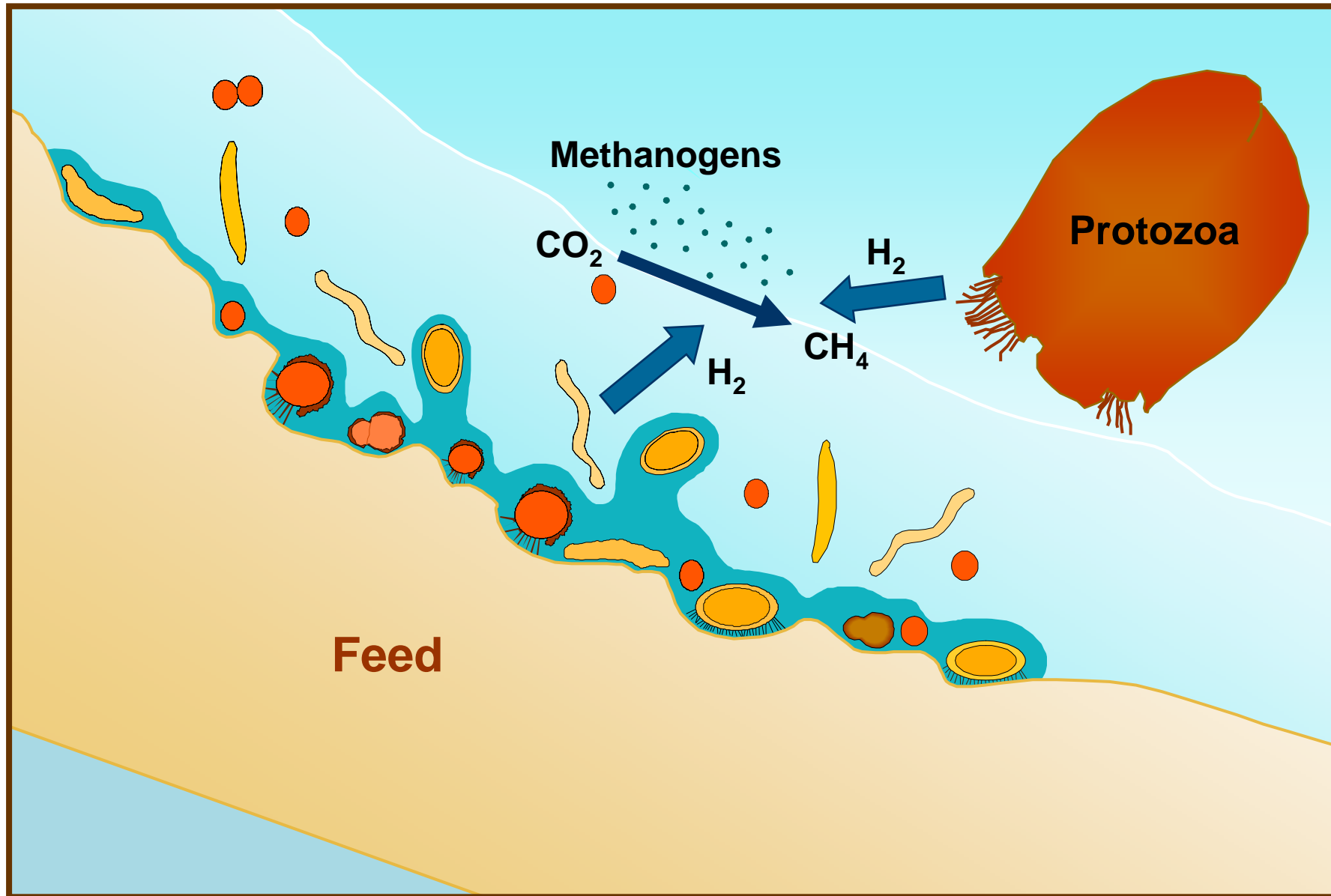
Contribution to greenhouse gas emissions (GWP-equivalents)



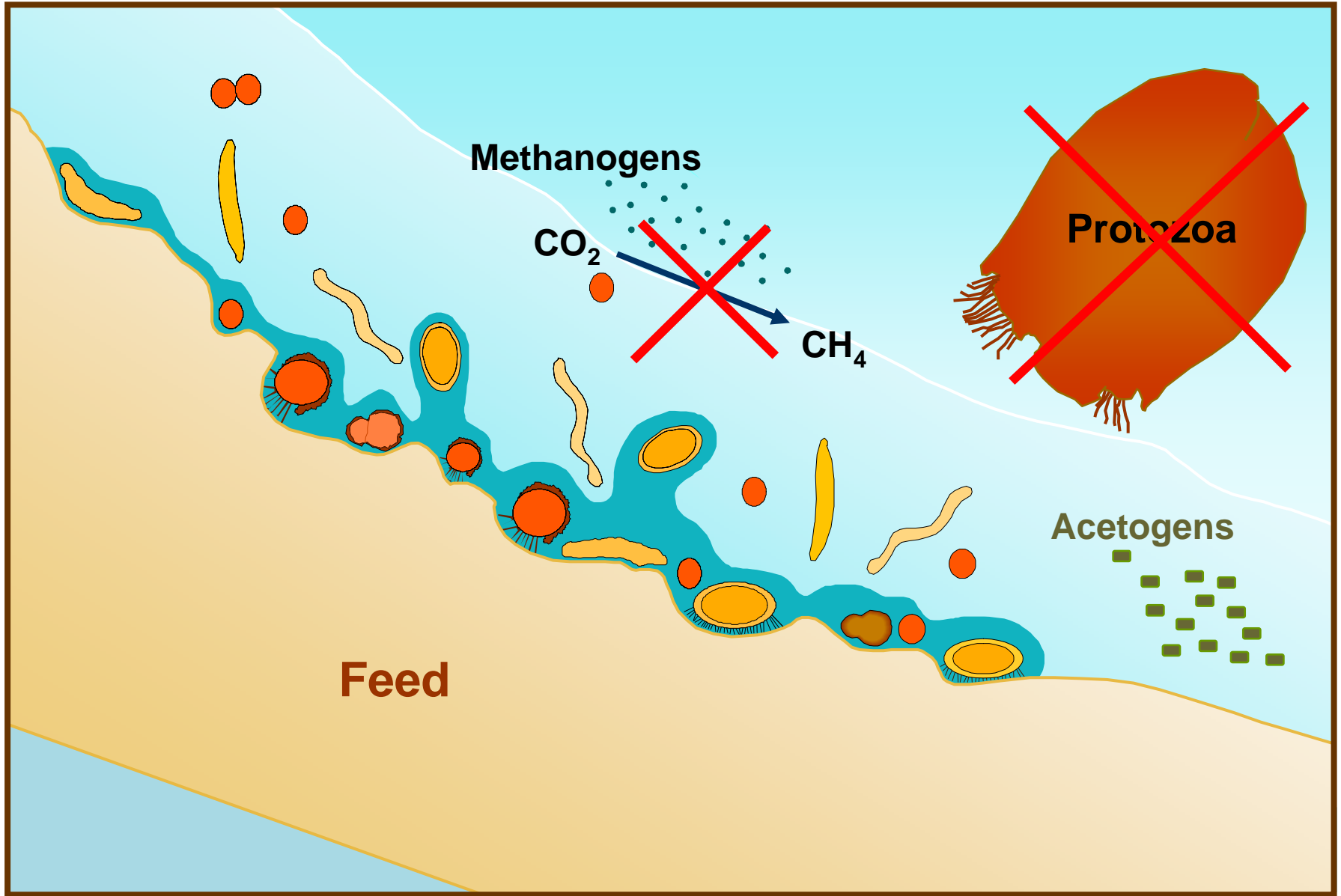
An opportunity

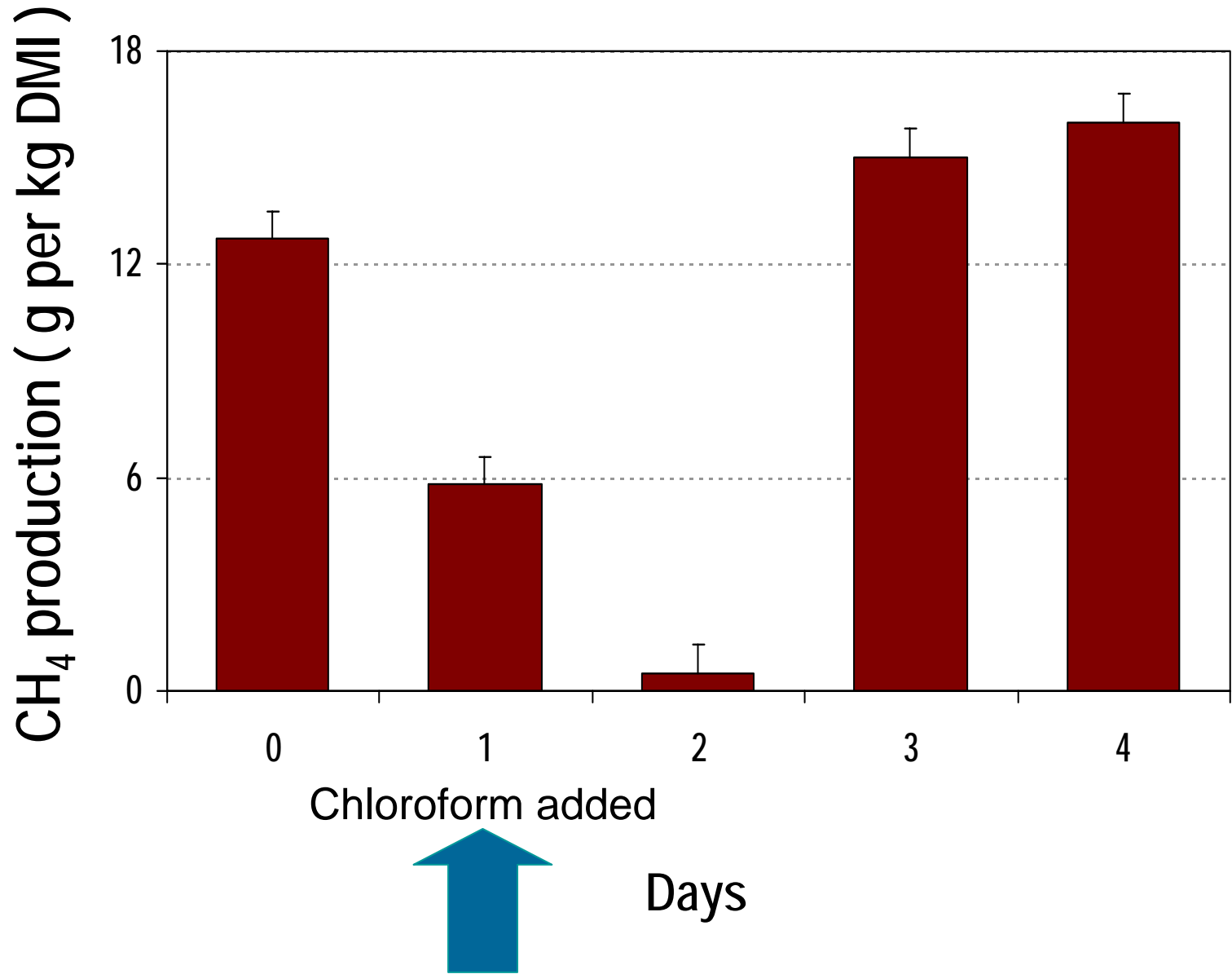


Methane production: a microbially driven process to remove hydrogen

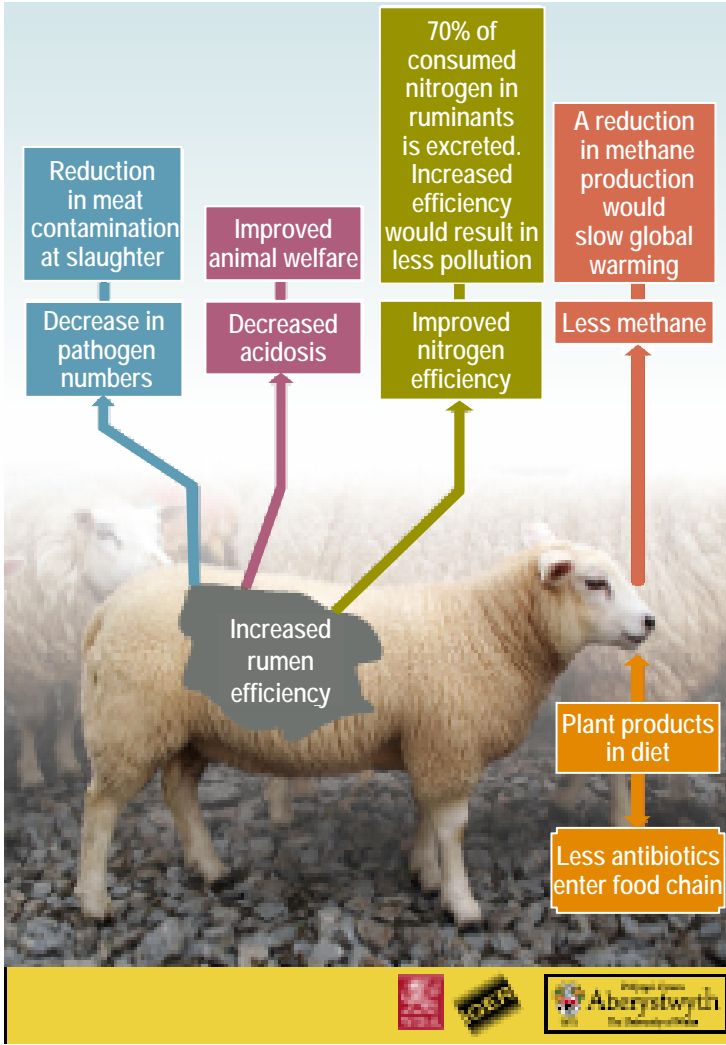


Methods of methane mitigation:  Direct inhibition of methanogenesis



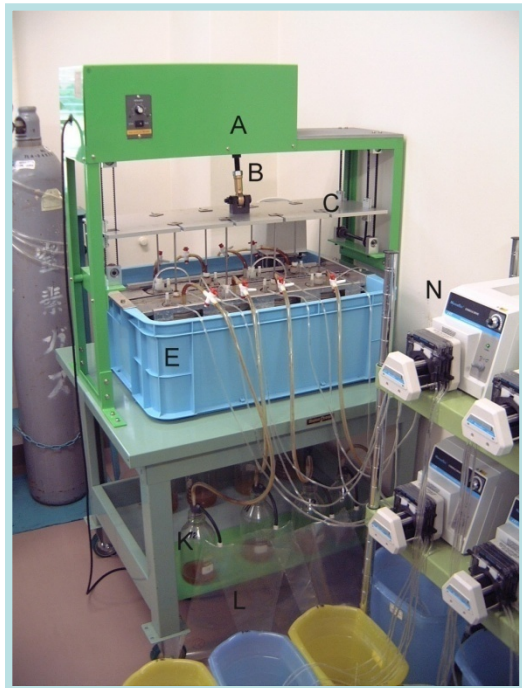


Plant extracts

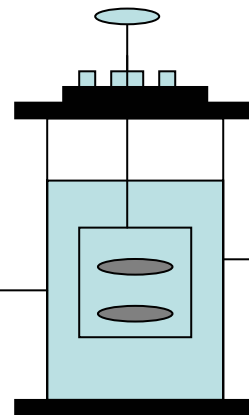


Rumen Simulation Technique (Rusitec)

Rusitec



Allicin
0, 2 or 20 $\mu\text{g/mL}$



- 250 mL ruminal fluid
- 250 mL artificial saliva
- 250 mL water

Dilution rate 26.5 mL/h

- Feed: 20 g/d
- 39° C for 17 d

(Hart et al., 2006)

VFA production (mmol per day)

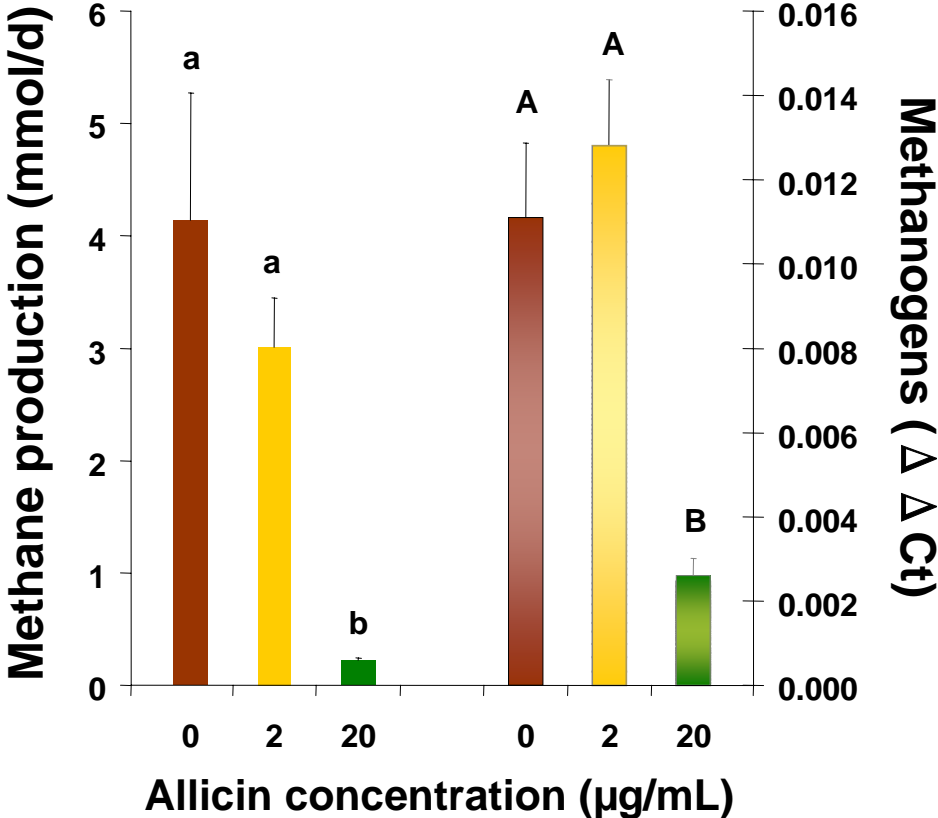
VFA	Allicin concentration ($\mu\text{g/mL}$)			s.e.m.	<i>P</i>
	0	2	20		
Acetate	17.5	17.4	17.9	1.84	0.964
Propionate	7.1	7.2	7.1	0.66	0.966
<i>n</i> -Butyrate	8.2	7.6	7.5	0.87	0.729
<i>iso</i> -Butyrate	0.29	0.32	0.33	0.038	0.552
<i>n</i> -Valerate	2.8	2.6	2.7	0.21	0.519
<i>iso</i> -Valerate	1.2	1.2	1.4	0.14	0.387
Total VFA	37.1	36.3	36.9	3.53	0.971

Fermentation parameters

Item	Allicin concentration ($\mu\text{g/mL}$)			s.e.m.	<i>P</i>
	0	2	20		
Vessel pH	6.9	6.9	6.7	0.14	0.367
NH ₃ (mmol/d)	43.9	42.0	46.5	3.20	0.412
CH ₄ (mmol/d)	4.14	3.01	0.22	1.16	0.022
24-h DMD (%)	22.8	28.6	23.2	6.39	0.615
48-h DMD (%)	34.1	34.0	31.2	7.39	0.905
Bacterial counts					
Total (CFU/mL)	10×10^9	5×10^9	8×10^9	4.7×10^9	0.353
		8.22	7.89		0.845
Cellulolytic*	8.16	(16.6×10^7)	(7.8×10^7)		

* \log_{10} CFU/mL (values in parentheses are back-transformed means/mL).

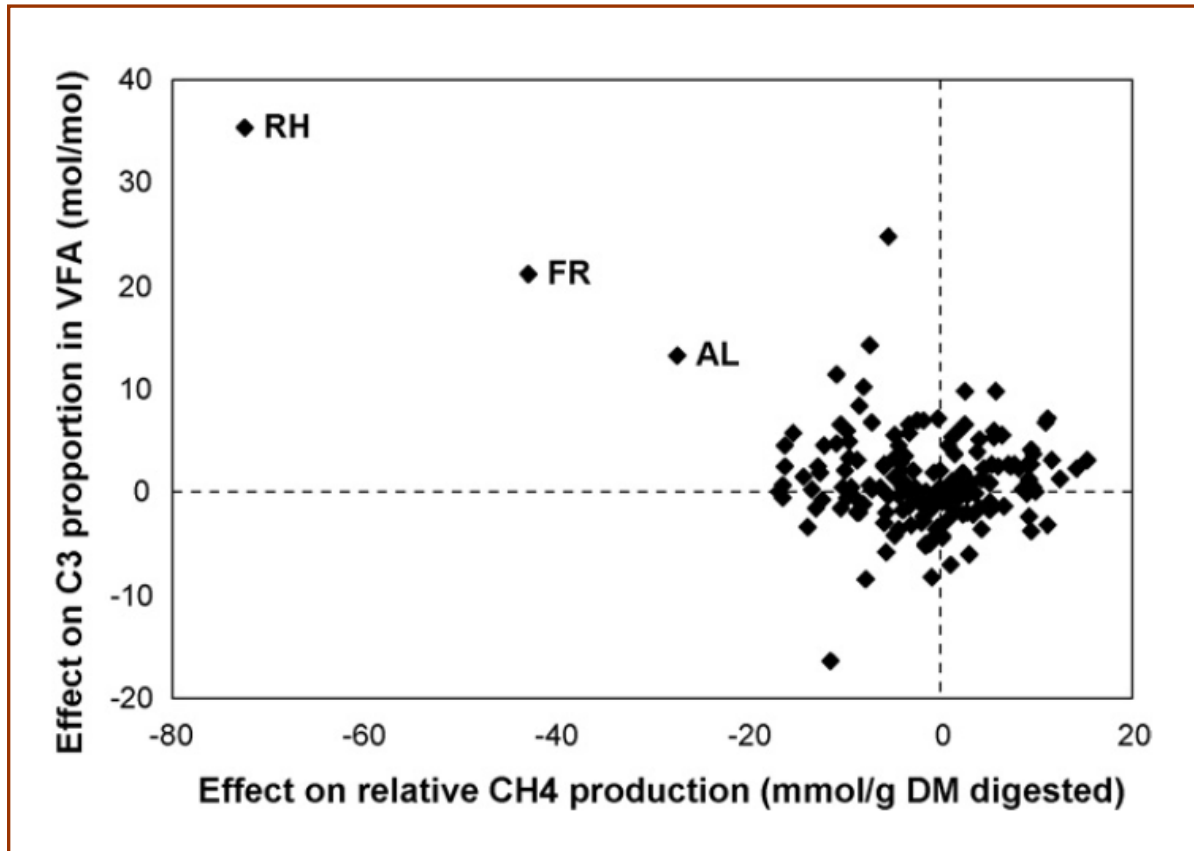
Garlic and methane



a,b or A,B: Means differ at ($P < 0.05$).

Plant additives

Increased propionate



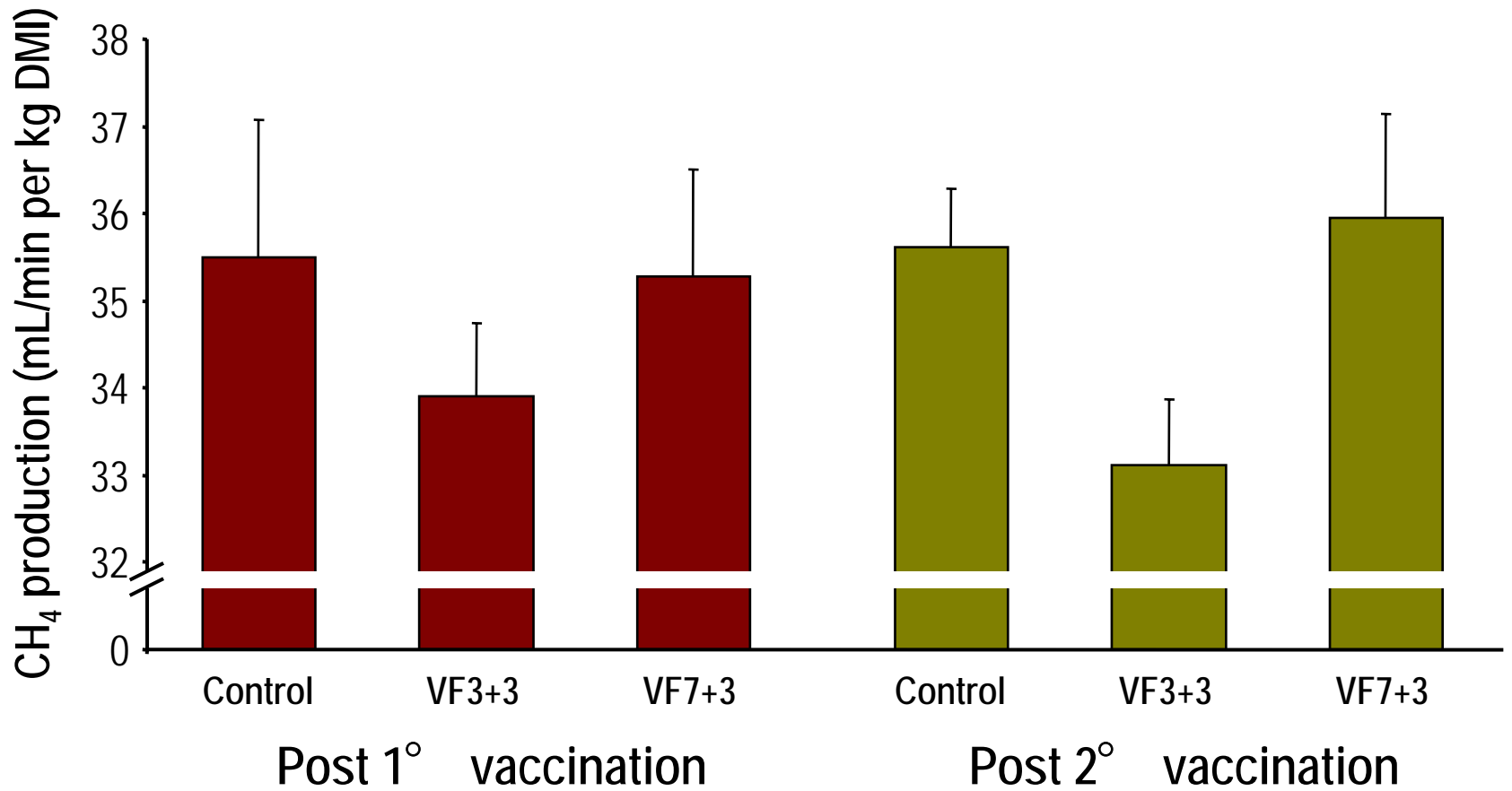
RH: *Rheum officinale*
(rhizomes)

FR: *Frangula alnus*
(bark)

AL: *Allium sativum*
(bulbs)

◆ = Individual agent tested. Changes are expressed as % of corresponding control values.

Decreased CH₄ production

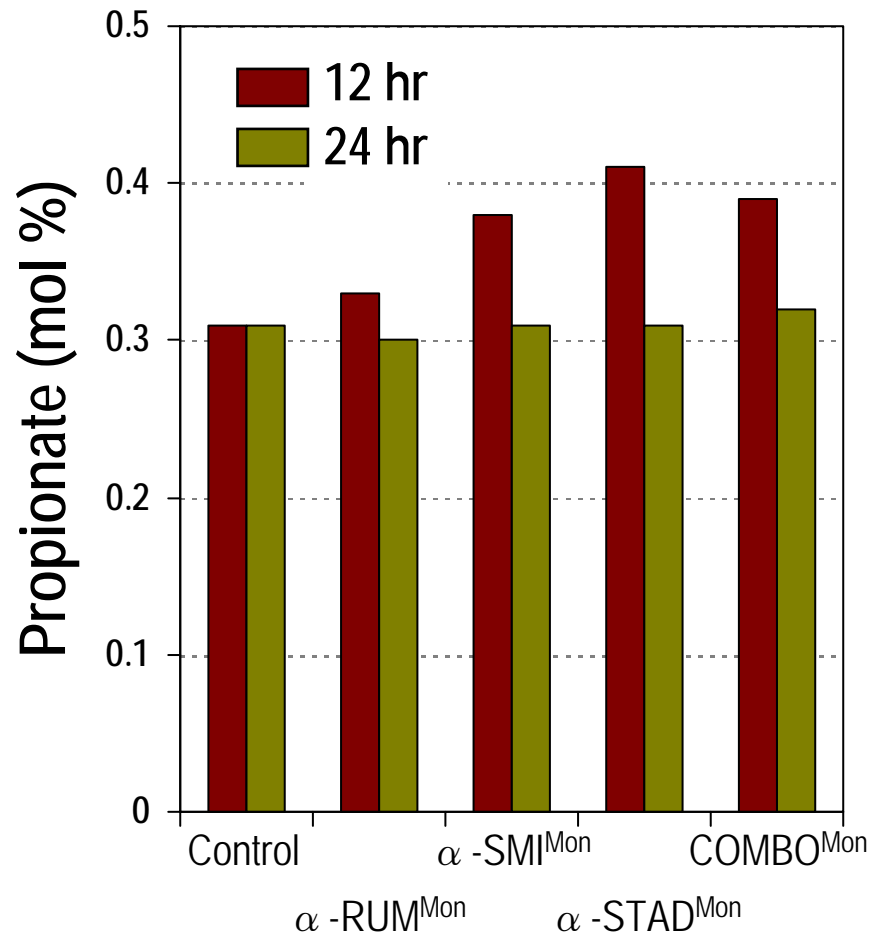
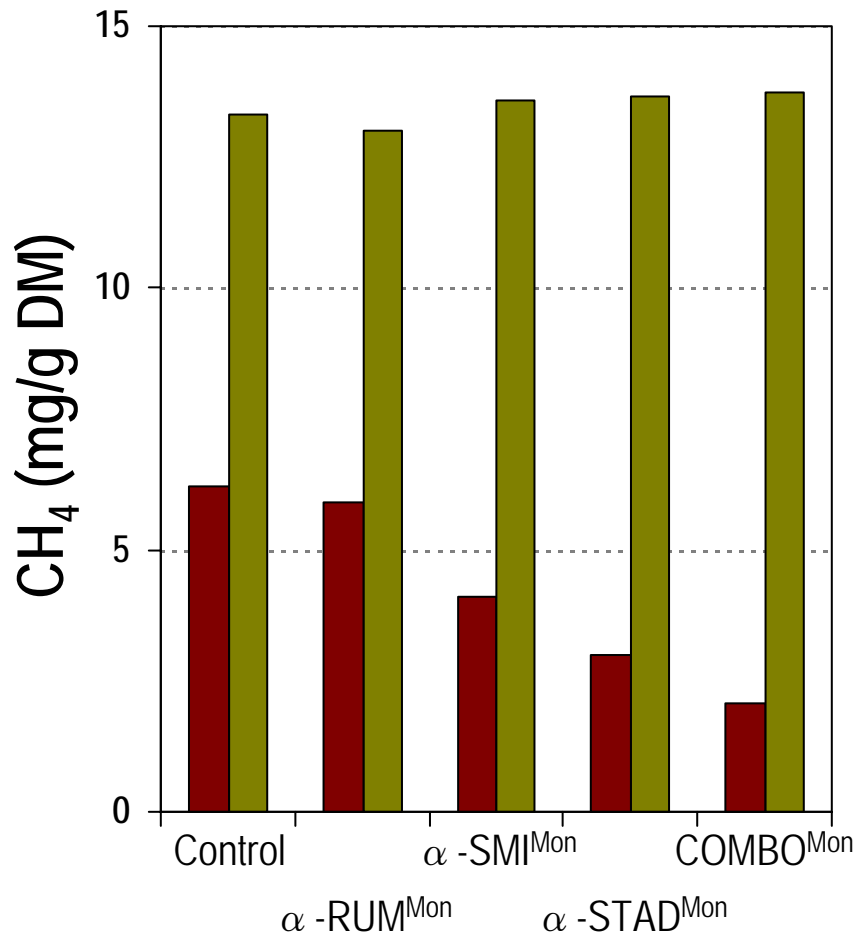


From Wright et al. (2004)

Passive immunization

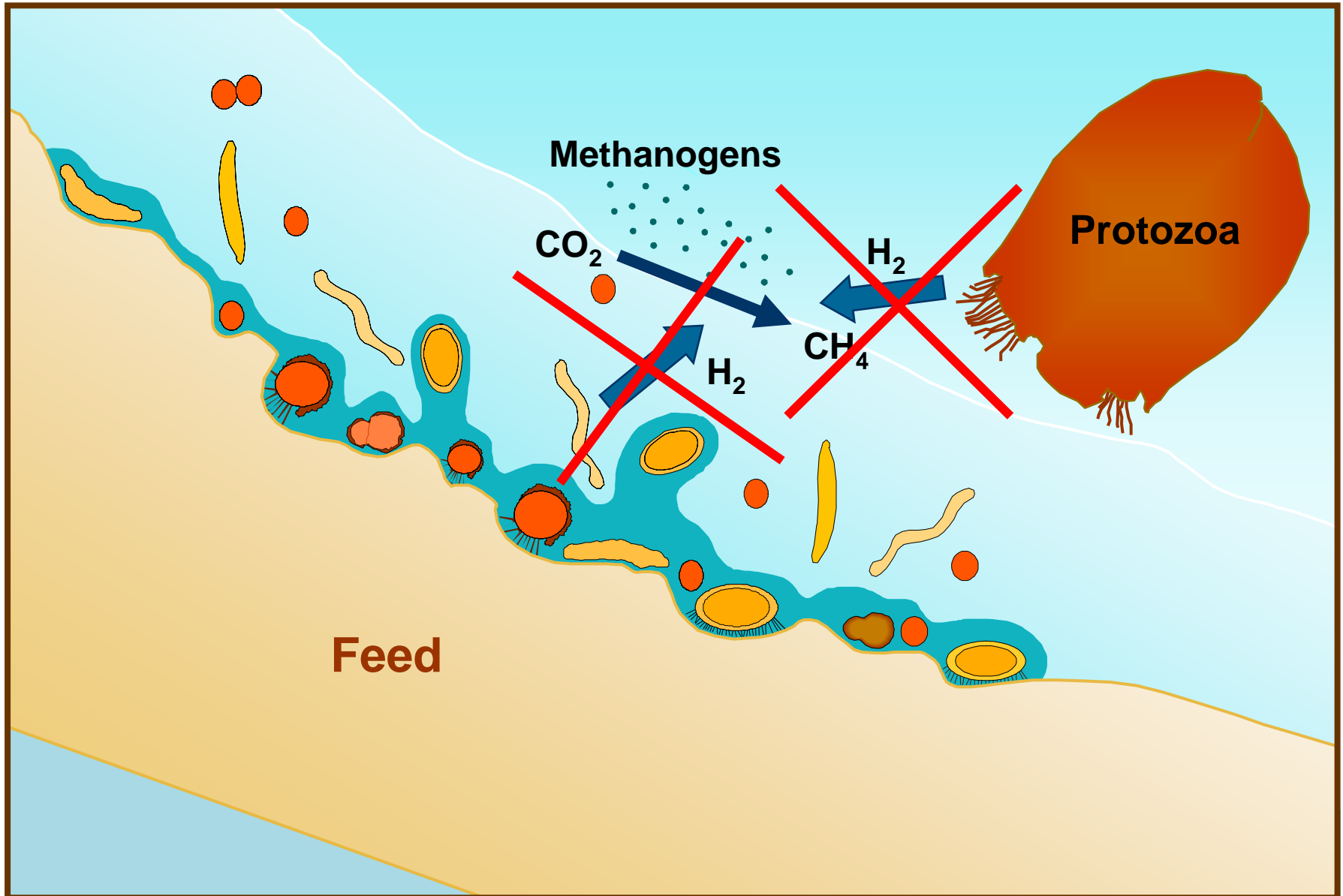
- Hens immunized with methanogenic bacteria:
 - *Methanobrevibacter smithii*
 - *Methanobrevibacter ruminantium*
 - *Methanosarcina stadtmanae*
- Studied as spray-dried whole egg or as semi-purified IgY
- Included in *in vitro* batch culture incubations
 - Treatments: α -SMI, α -RUM, α -STAD or combination
 - Control: egg powder from non-immunized hens
 - Negative control: no egg powder

Anti-methanogen egg powder: *in vitro* effects

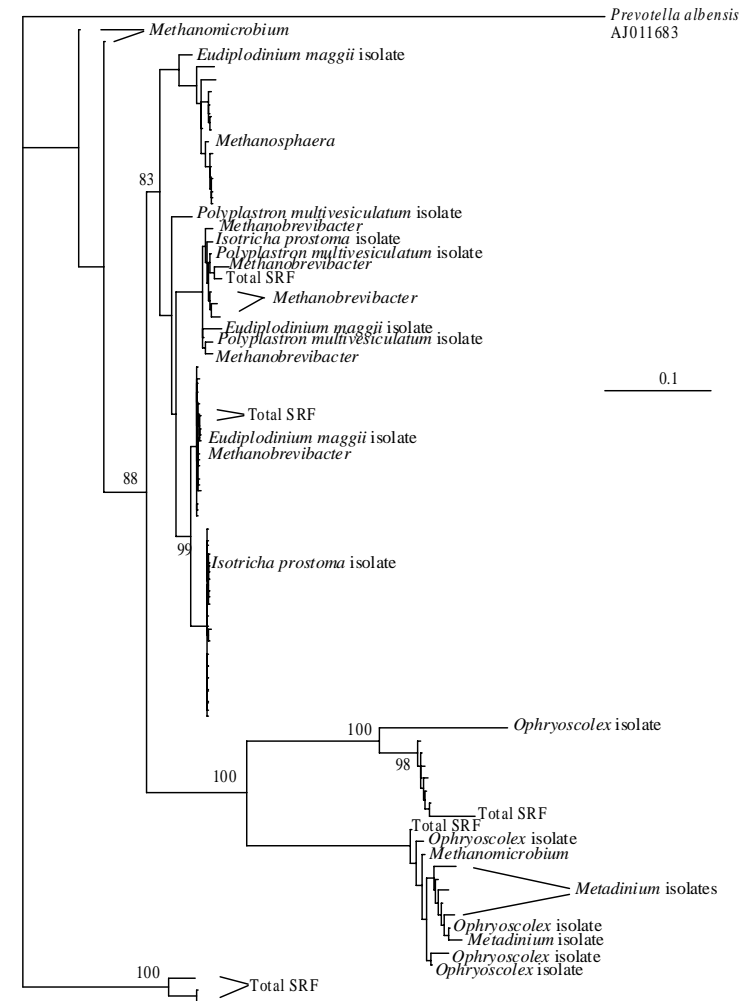
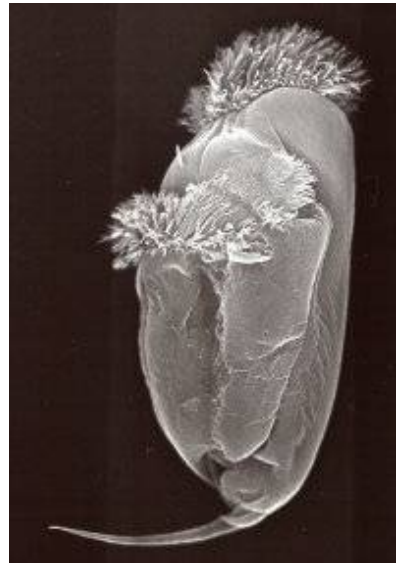
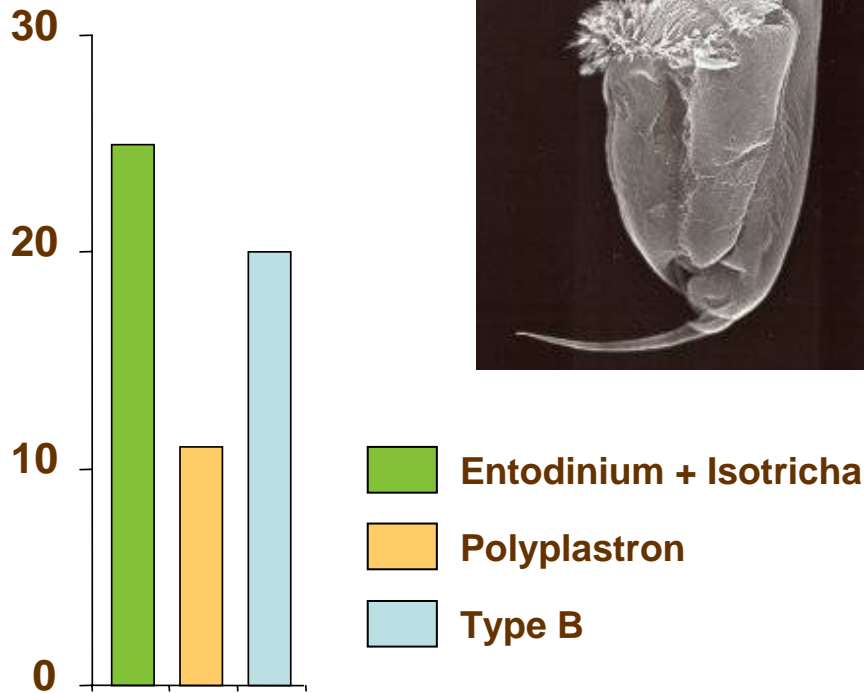


Treatment

Methods of methane mitigation: \rightarrow Decrease H_2 production

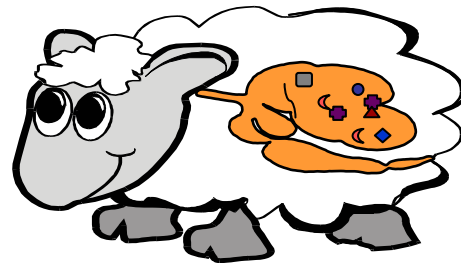


Methanogens and protozoa

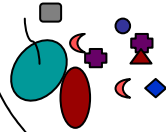


Methanogenesis associated with protozoa (%)

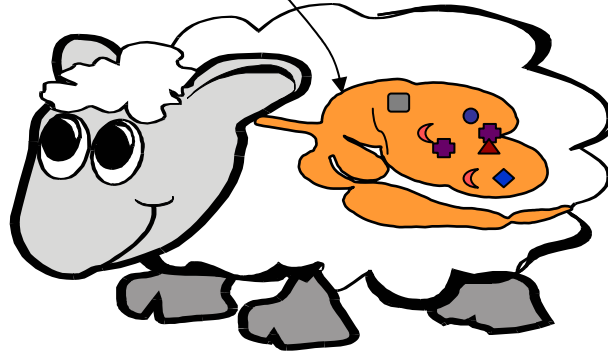
8 weeks



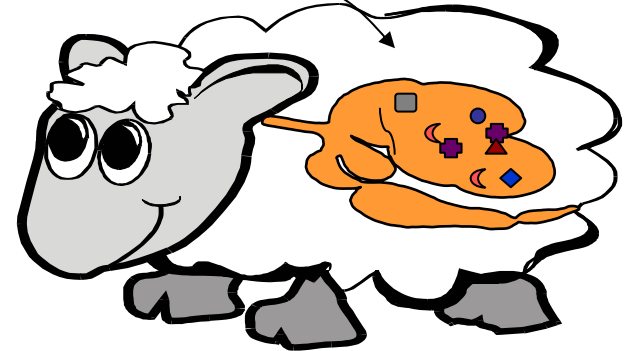
$n = 12$



5 months

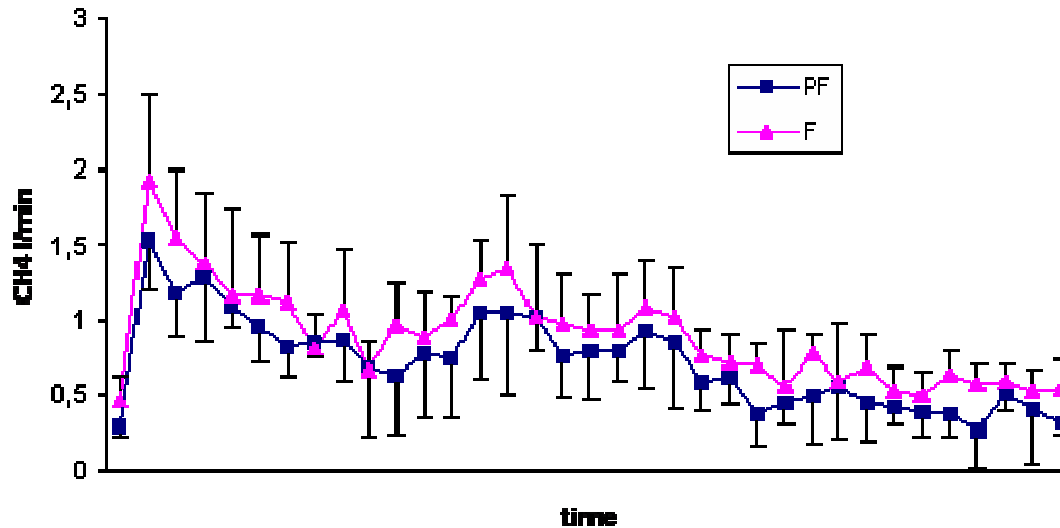


Faunated (F)
($n = 6$)



Protozoa-free (PF)
($n = 6$)

Methane emissions



CH ₄ production	PF	F	s.e.m.	<i>P</i>
L per day	26.0	35.2	2.82	0.049
L per kg LW	0.52	0.71	0.044	0.024
L per kg DMI	21.6	29.0	1.41	0.006

PF: protozoa-free lambs; F: faunated lambs.
LW: liveweight; DMI: dry matter intake

Bacterial numbers (qPCR/MPN) whole contents

Parameter	PF	F	s.e.m.	<i>P</i>
Total bacteria (ng DNA/mL)	51.7	38.4	7.86	0.257
Total methanogens ($\Delta \Delta$ Ct)	0.14	0.36	0.008	0.078

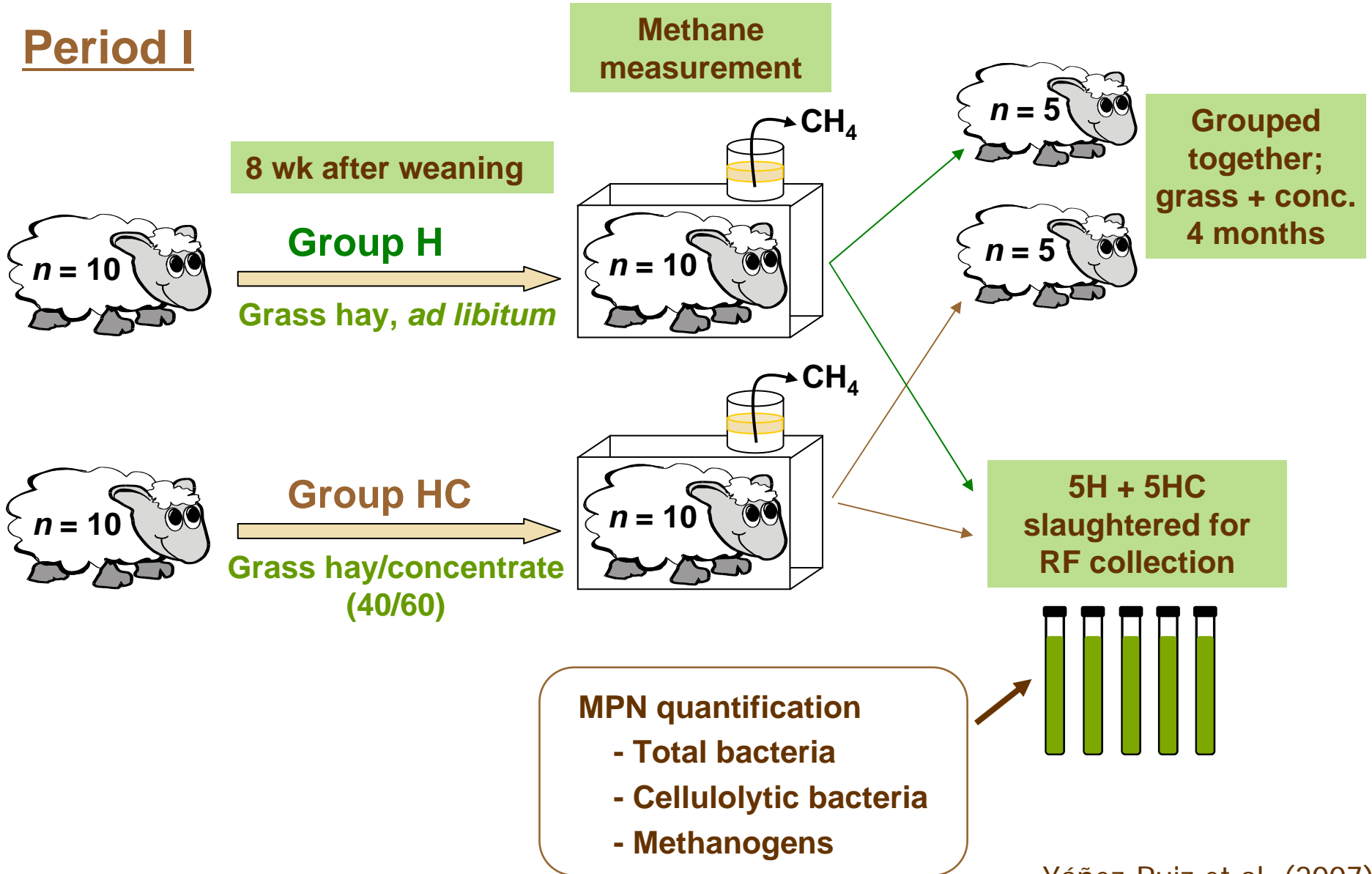
PF: Protozoa-free lambs

F: Faunated lambs



Effect of diet at weaning

Period I



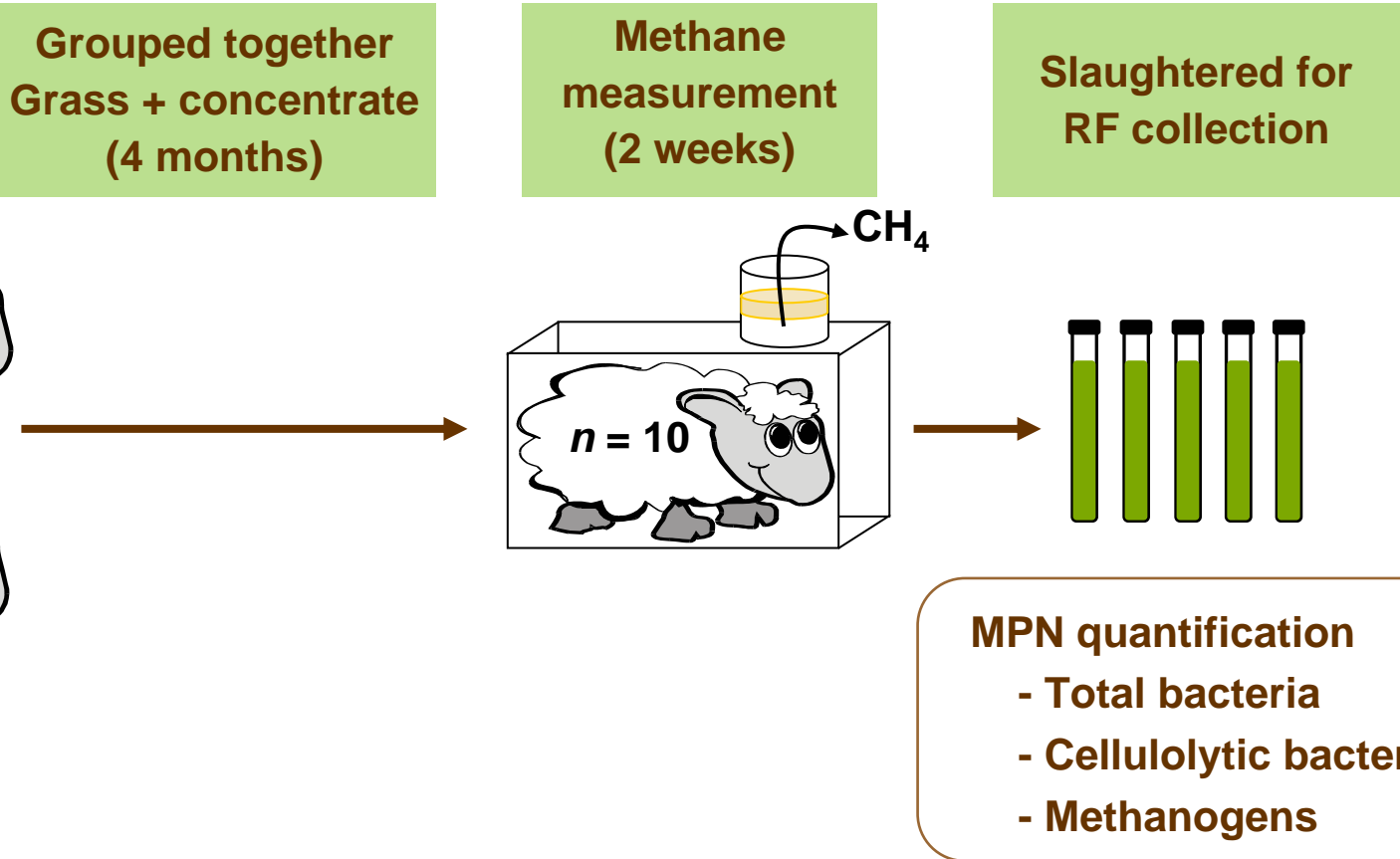
Lambs fed hay or hay/concentrate only

Measurement	Measurements taken 8 weeks after weaning		
	H lambs	HC lambs	<i>P</i> ^a
CH ₄ (L per kg DMI)	26.0	22.5	0.04
Microbial populations (per mL) ^b			
Total bacteria	10.2 × 10 ¹⁰	6.1 × 10 ¹⁰	0.084
Cellulolytic bacteria	40.6 × 10 ⁹	10 × 10 ⁹	0.098
Methanogenic bacteria	37 × 10 ⁹	19 × 10 ⁹	0.113
Acetate:propionate ratio	3.00	2.35	0.089

^aProbability (*P* value) of a difference between treatment means.
^bDetermined by Most Probable Number.

Effect of diet continued...

Period II



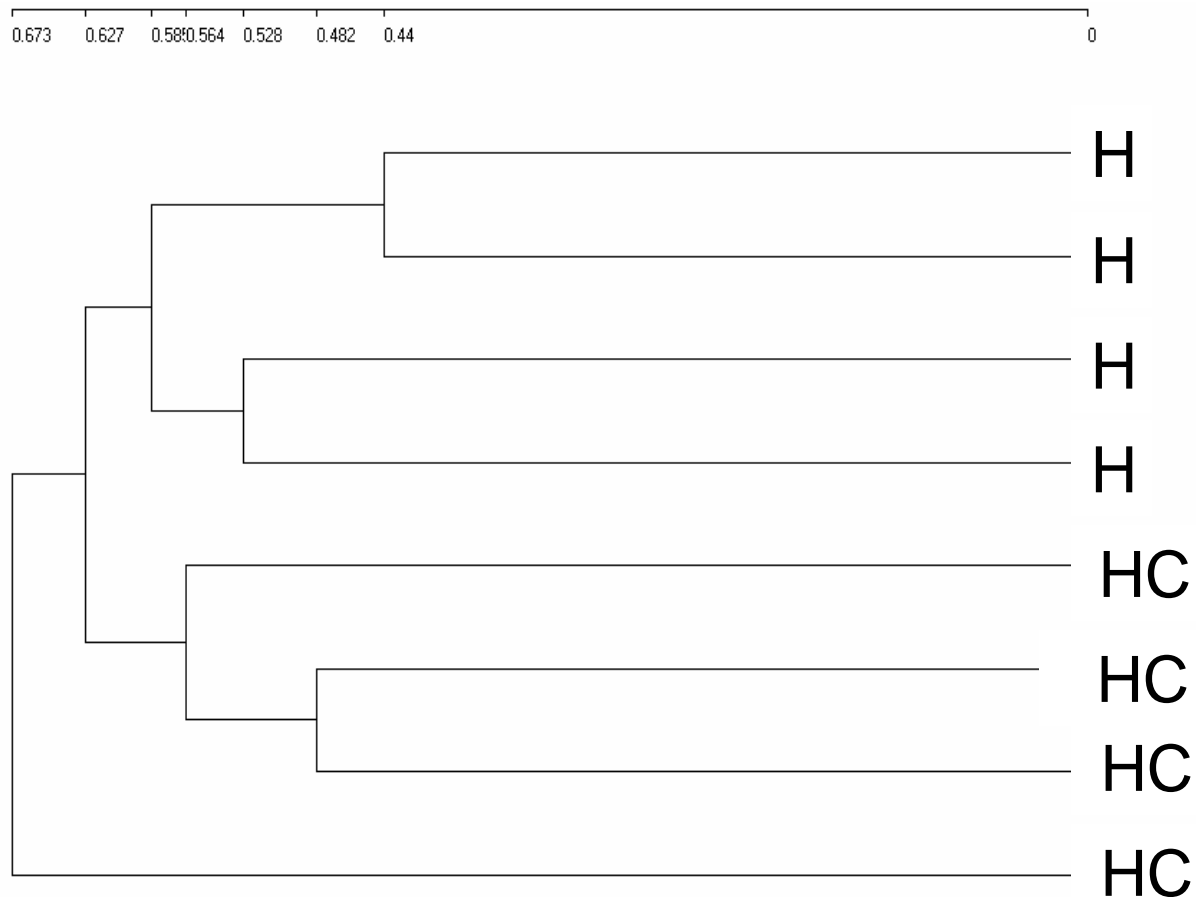
Lambs fed hay or hay/concentrate

Measurement	Measurements taken 8 weeks after weaning			Measurements after 4 months of identical diets		
	H lambs	HC lambs	<i>P</i> ^a	H lambs	HC lambs	<i>P</i> ^a
CH ₄ (L per kg DMI)	26.0	22.5	0.04	25.7	26.6	0.591
Microbial populations (per mL) ^b						
Total bacteria	10.2 × 10 ¹⁰	6.1 × 10 ¹⁰	0.084	4 × 10 ¹⁰	11 × 10 ¹⁰	0.939
Cellulolytic bacteria	40.6 × 10 ⁹	10 × 10 ⁹	0.098	6.5 × 10 ⁹	20.3 × 10 ⁹	0.294
Methanogenic bacteria	37 × 10 ⁹	19 × 10 ⁹	0.113	25 × 10 ⁹	22.3 × 10 ⁹	0.73
Acetate:propionate ratio	3.00	2.35	0.089	2.87	2.81	0.659

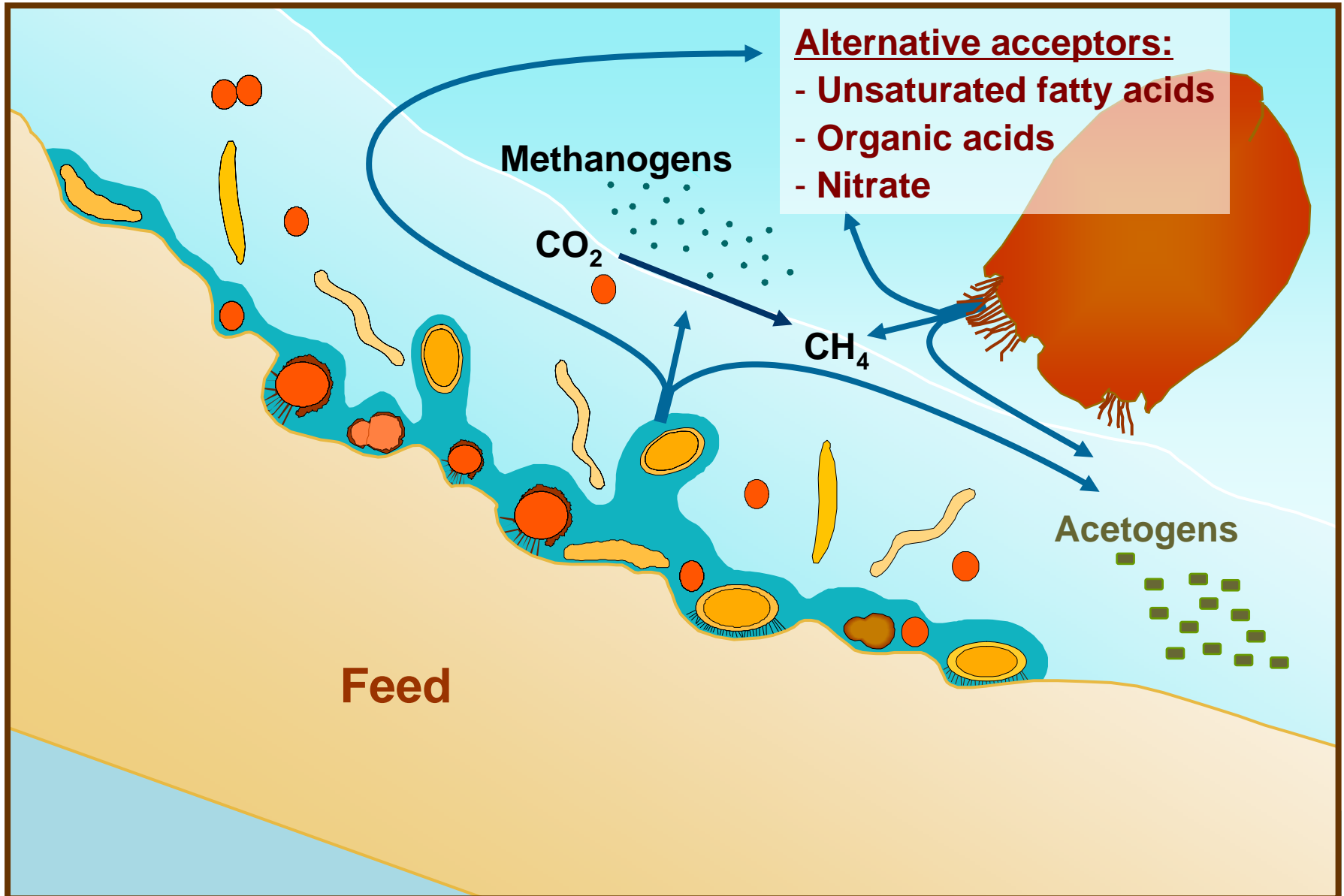
^aProbability (*P* value) of a difference between treatment means.

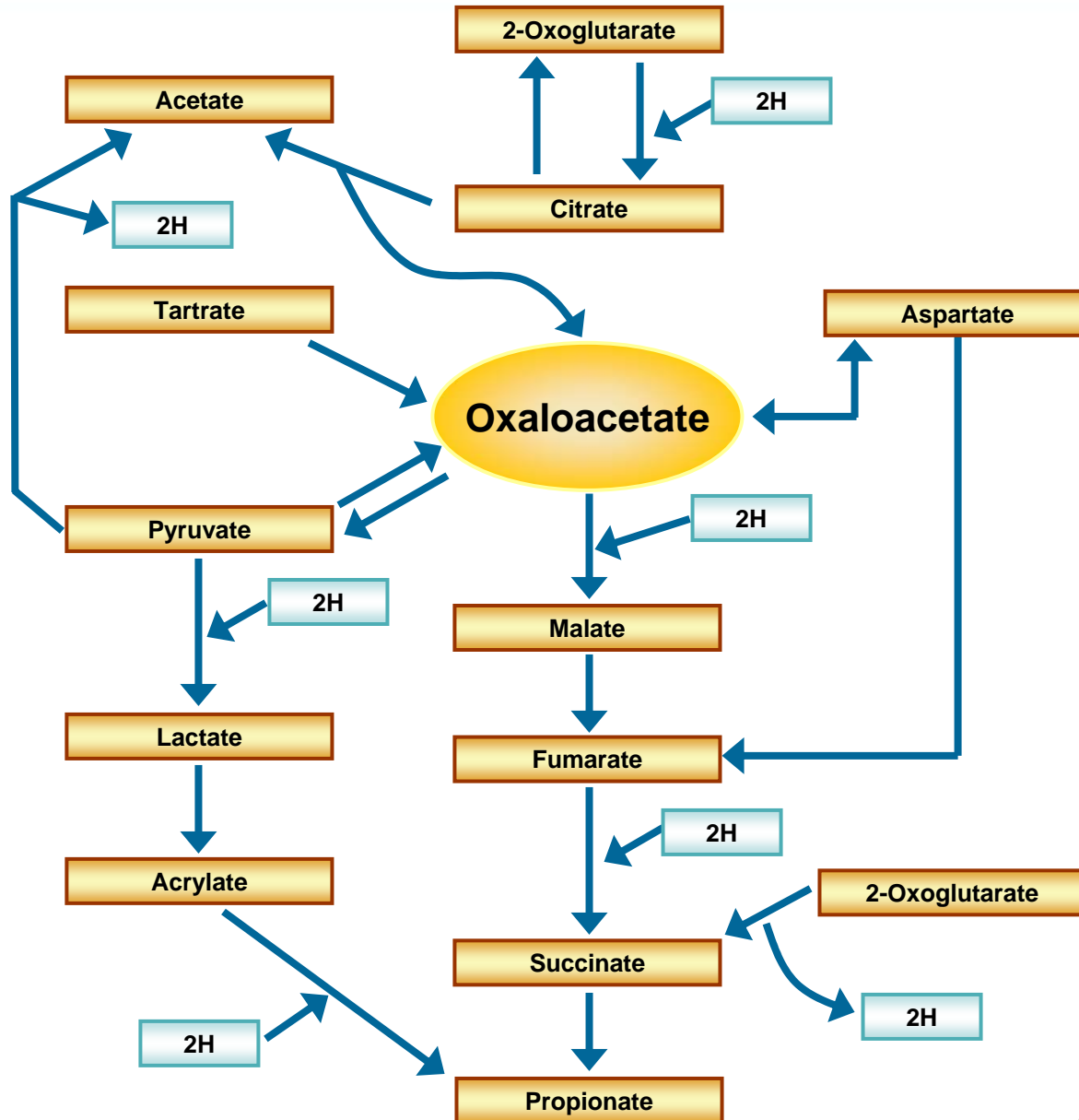
^bDetermined by Most Probable Number.

Bacterial profile determined by T-FRLP of the 16S rDNA gene after 4 months on identical diets

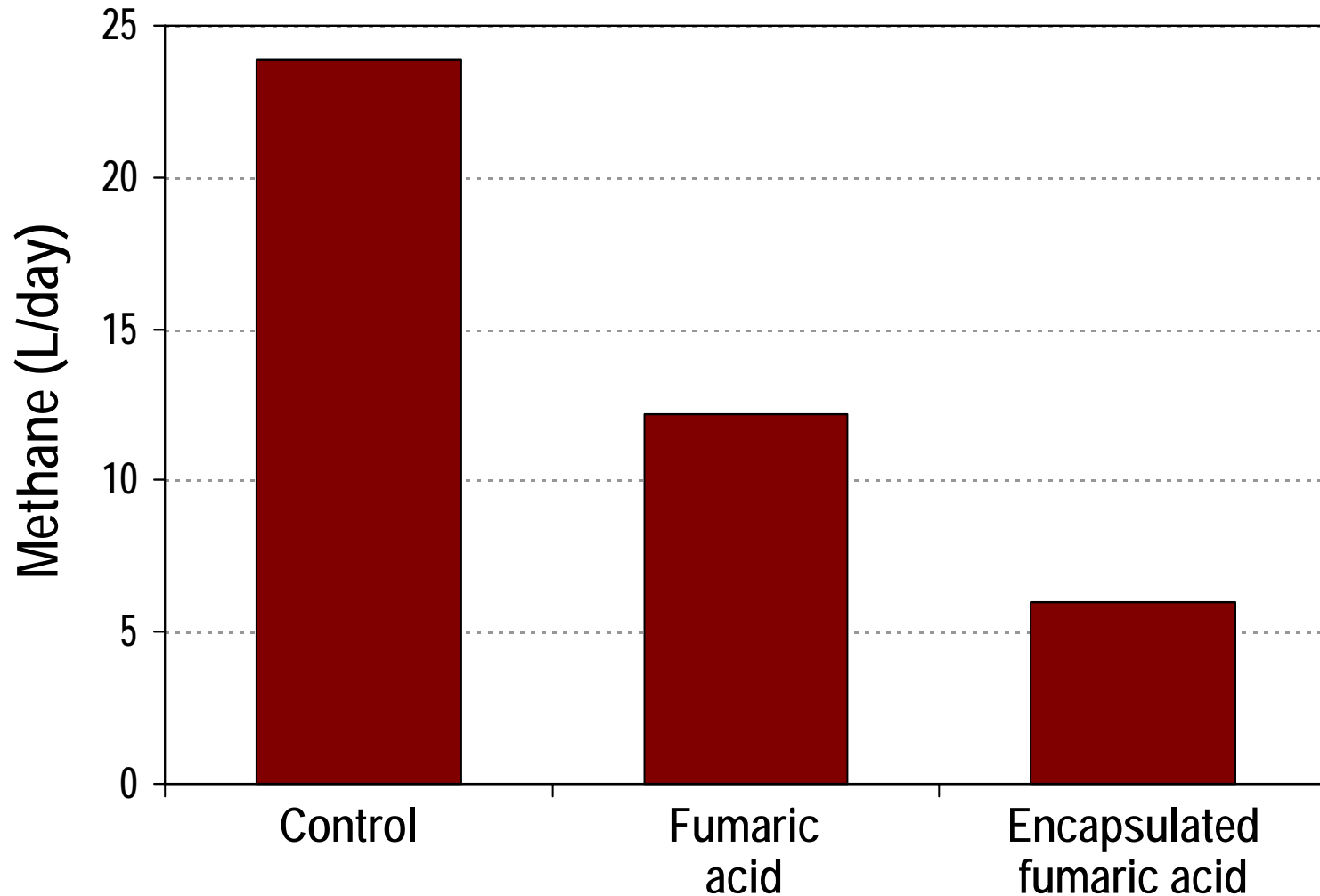


Methods of methane mitigation: \Rightarrow Redirect H_2 flow





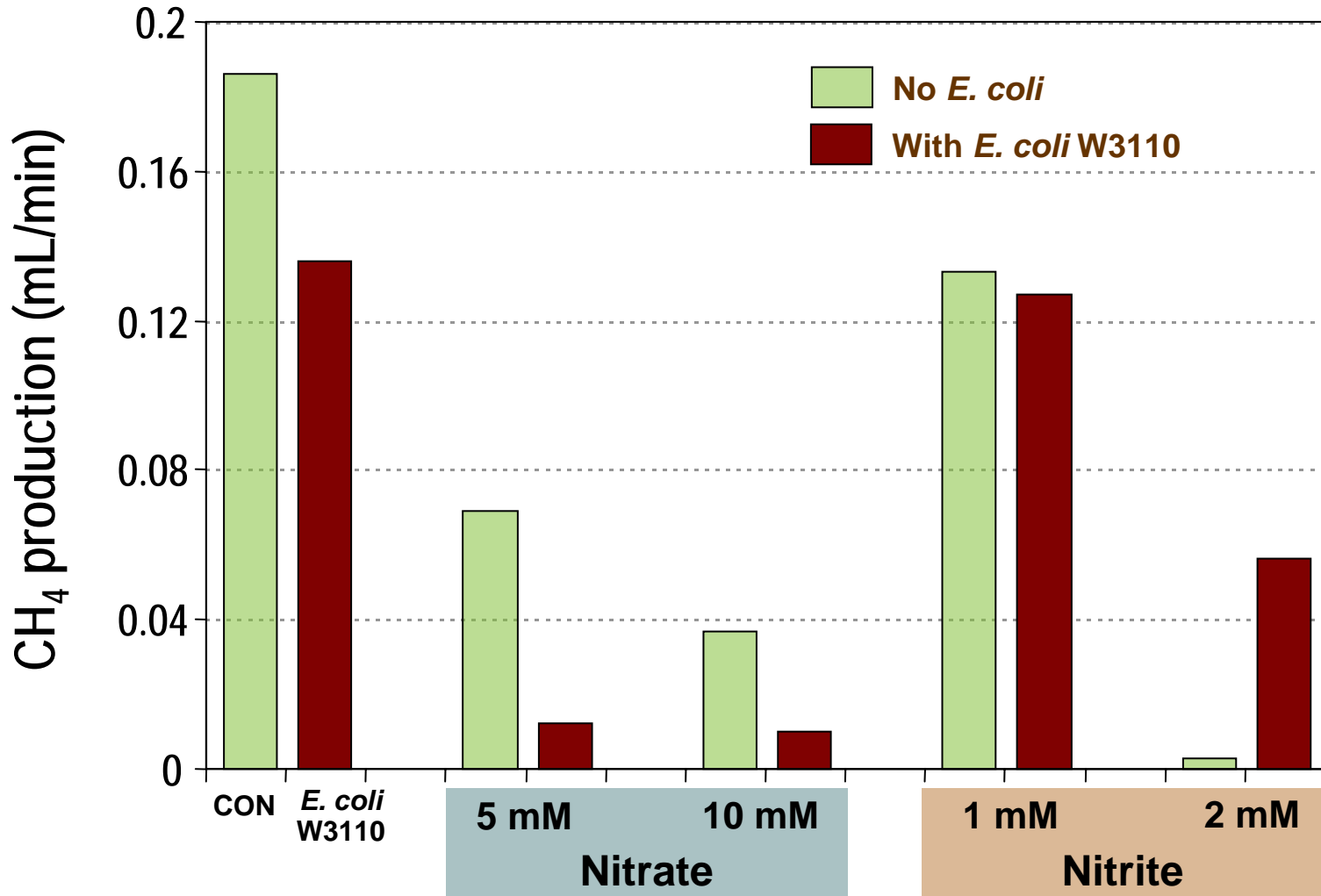
Methane production by lambs supplemented with fumaric acid



Redirecting H⁺

- Nitrate as an alternative electron sink
 - Problem with nitrite toxicity
- *Escherichia coli* W3110
 - Nitrite reduction activity
- *In vitro* continuous culture incubation
 - Buffered (1:1) ruminal fluid inoculum
 - NaNO₃ (5 or 10 mM); NaNO₂ (1 or 2 mM)
 - *E. coli* W3110 (approx. 1×10^9 CFU/mL)
 - Rates of production of CH₄ and CO₂

Rate of CH₄ production





Questions

Acknowledgements

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