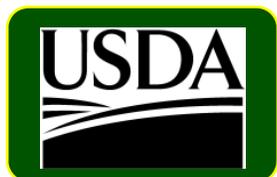


Rumen Microbes: A Cow's Best Friend

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RUMINANT ANIMALS

- Domesticated (cattle, sheep, goats, etc.) and wild (bison, antelope, etc.).
- Originally developed as grazers/browsers, more recently have been adapted to mixed rations.
- Nutrition based on plant material that cannot be digested by most other herbivores, including man.



HOW DO THEY DO IT?
Microbial fermentation!



Domestic Ruminants

Feed and Forage —————> Meat, milk, wool, hides

~ 2 billion cattle/sheep/goats worldwide

~ 100 billion liters of fermentation capacity

**The rumen fermentation is the world's
largest commercial fermentation !**

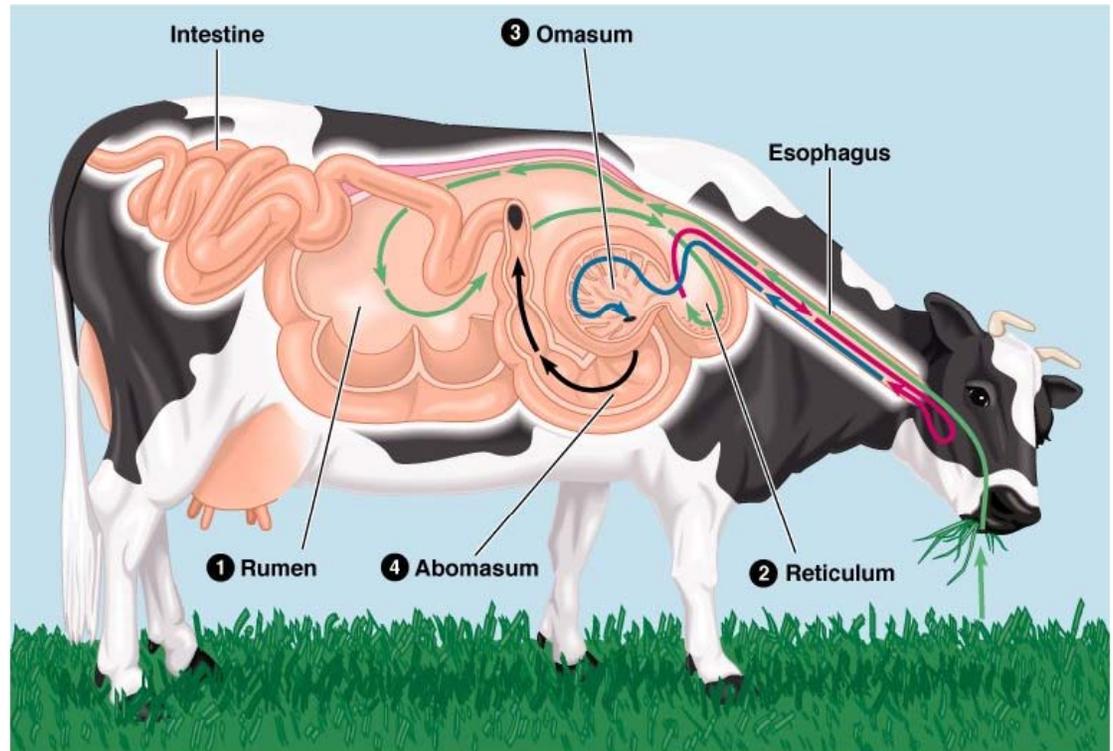
“Four Stomachs”

Rumen - “Fermentation Vat”

Reticulum - Additional fermentation

Omasum - Water and salts absorption; return of large feed particles to reticulorumen

Abomasum - Acid digestion of proteins to amino acids and of carbohydrates



©1999 Addison Wesley Longman, Inc.

Rumen



Reticulum



Omasum



Abomasum



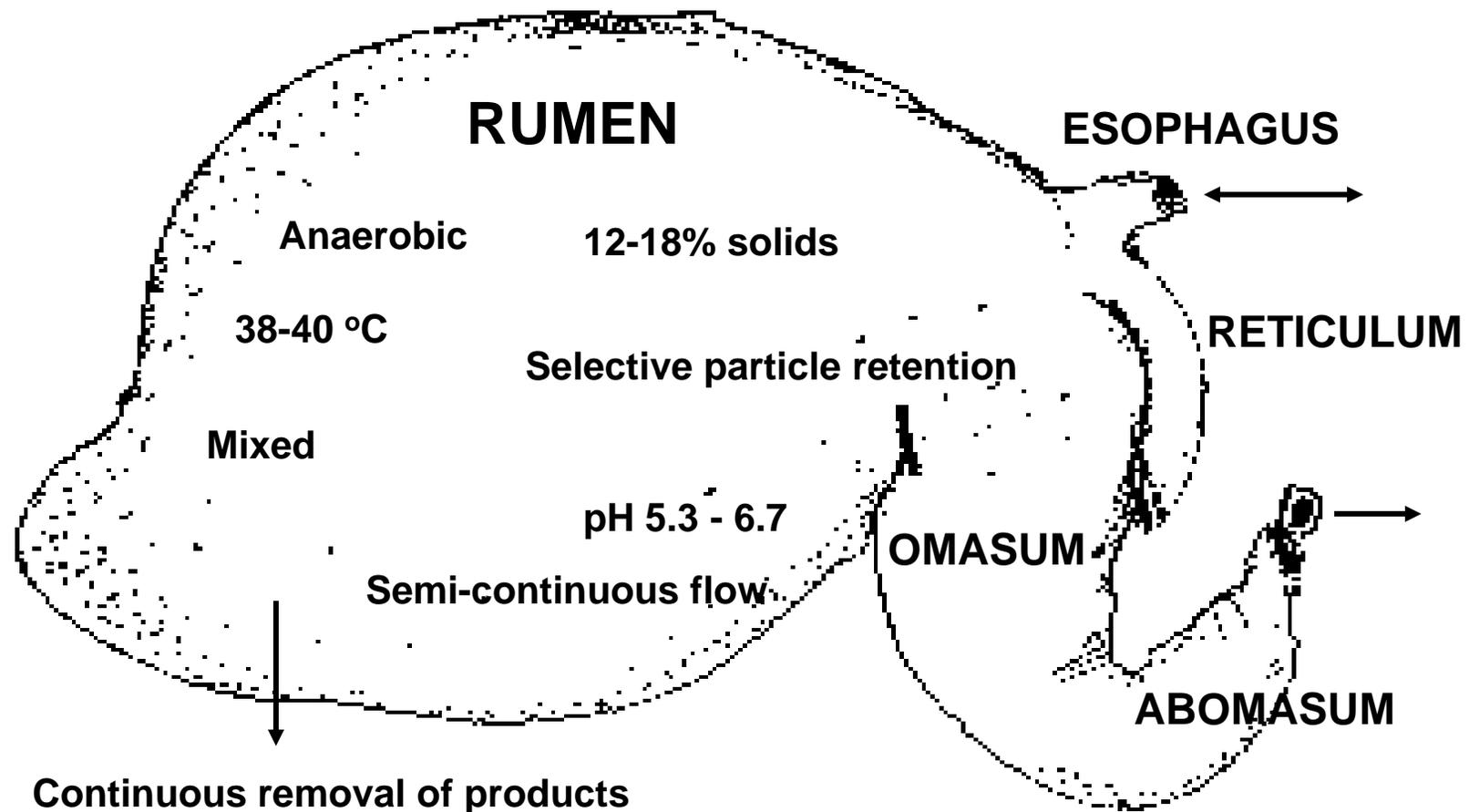
FEED PROCESSING

- Bites swallowed with minimal chewing initially.
- Feed fermentation begins in the rumen.
- Partially-digested feed is regurgitated from the rumen and re-chewed (rumination, or “chewing the cud”).
- Unique chewing motion maximizes substrate surface area while maintaining long fibers that stimulate rumination).
- Rumination also stimulates saliva production, which helps buffer ruminal pH.

A grazing cow takes about 35,000 bites per day!

KEY POINT: Effective physical pretreatment combined with long residence time in the rumen permits more complete degradation of plant biomass.

The rumen is a highly efficient natural bioreactor for processing plant biomass



Rumen fistula provides access to the rumen habitat



In vitro experiments



Removal of rumen inocula for degradation experiments in lab, and for characterization of microbial populations

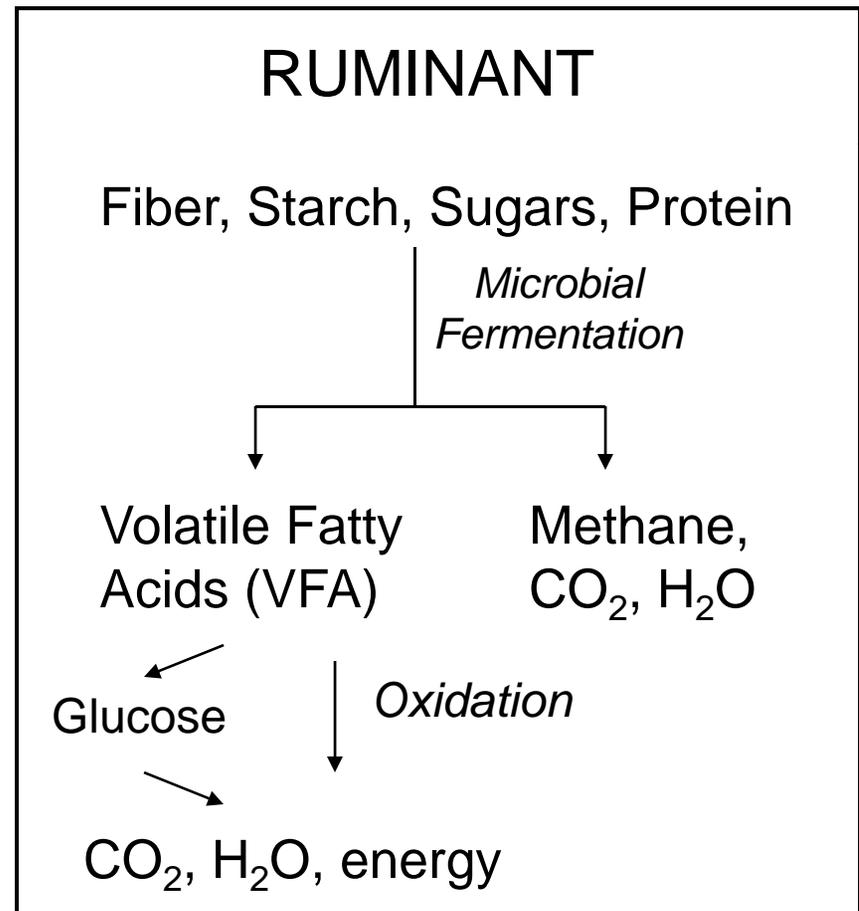
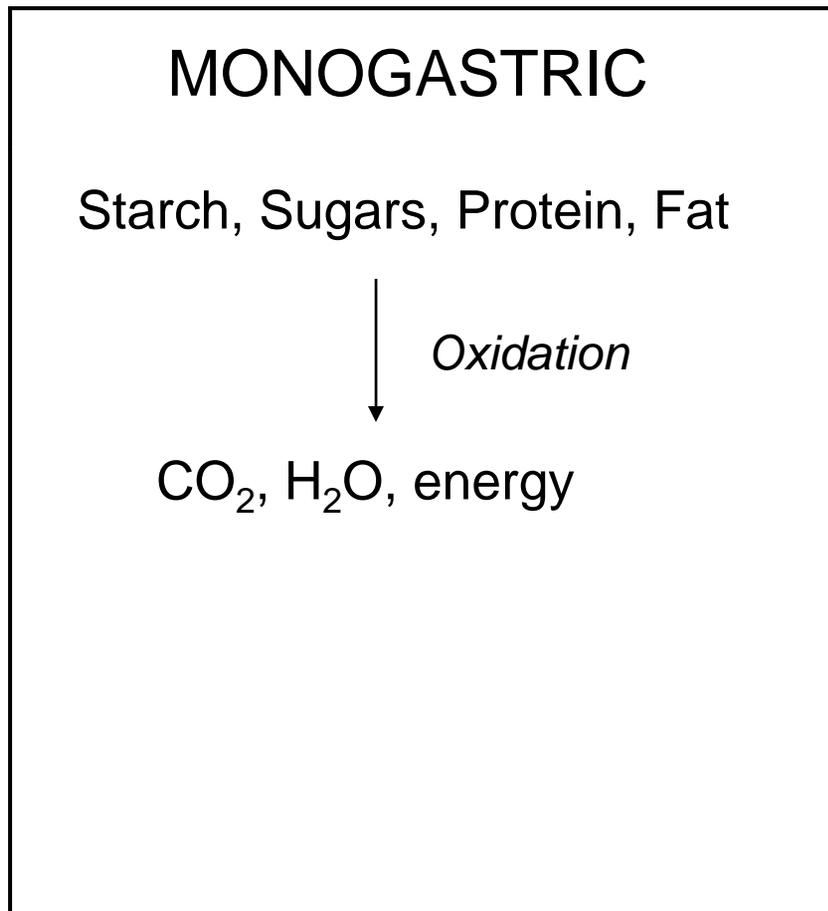
In sacco experiments



Insertion of feed materials into rumen to determine their rate and extent of degradation

Bovine energy metabolism

Cows and other ruminant animals have a different digestion strategy than humans and other “monogastric” animals.



The major groups of rumen microorganisms and their primary roles

BACTERIA

Ferment fiber, starches and sugars in feeds to VFA, H₂ and CO₂

Produce most of microbial cell protein, but also ferment feed proteins to VFA + NH₃

Alter toxicity of plant metabolites

PROTOZOA

Consume and ferment bacteria to VFA + NH₃

Sequester and ferment starch

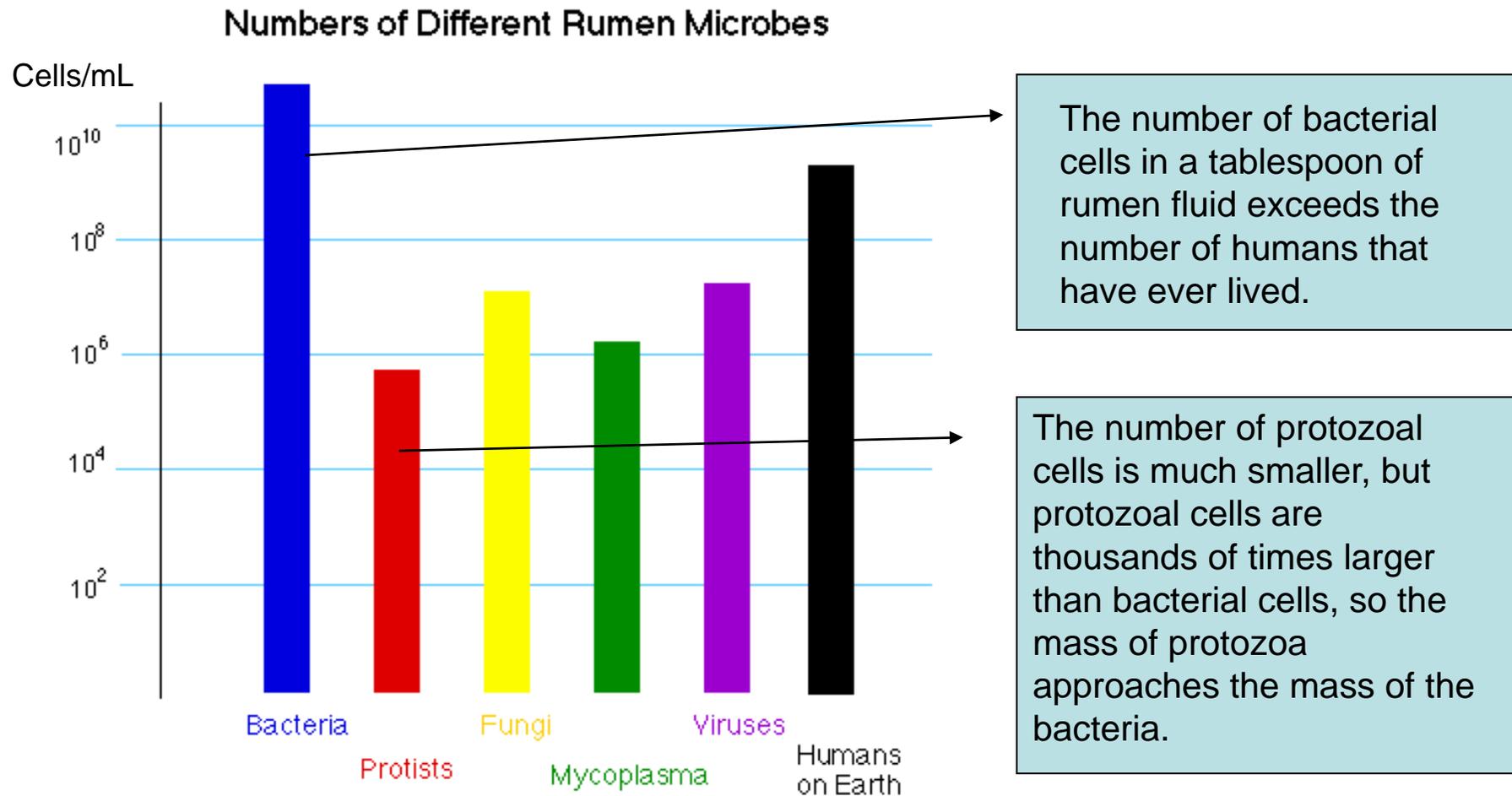
Recycle N

ARCHAEA

Thermodynamic displacement, producing methane

FUNGI Assist in fiber degradation

Microbial populations in the rumen



Data courtesy of Mel Yokoyama, Michigan State University

“Good bugs” and “bad bugs” ?

Few rumen microbes are all good or all bad.

Protozoa - Consume bacteria, reducing efficiency of microbial protein synthesis, *but* engulf and slowly ferment starch, limiting rapid starch conversion to lactic acid.

Methanogens - Produce methane (GHG), *but* allows for more complete feed utilization (--> higher cell yield --> greater microbial protein synthesis).

Microbes work as a team, and it is difficult to rate individual species in isolation.

Can we “direct” microbial community composition?

- Demonstrated successes in alleviating toxicoses (mimosine, oxalate, fluoroacetate)
- Inconsistent benefit of probiotics
- No success with “mainstream” rumen bacteria (fibrolytics, etc.)

Suggests that indigenous microbial population is stable and does not welcome invaders, unless they fill an unoccupied niche.

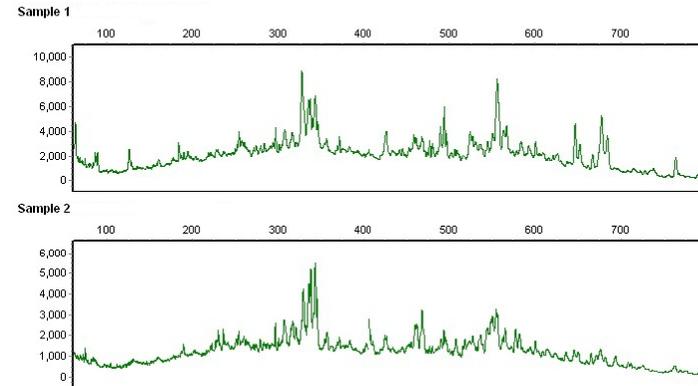
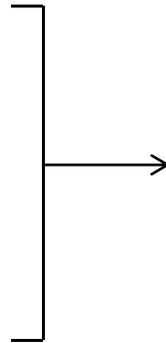
Inconsistent effects suggest that rumen microbial communities differ among cows.

Analyzing differences in microbial communities via ARISA

(Automated Ribosomal Intergenic Spacer Analysis)



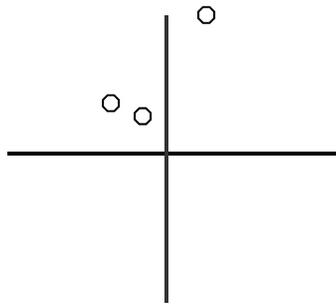
Extract and purify DNA
PCR-amplify ITS region
between 16S and 23S
rRNA genes



Separate amplicons by
capillary electrophoresis



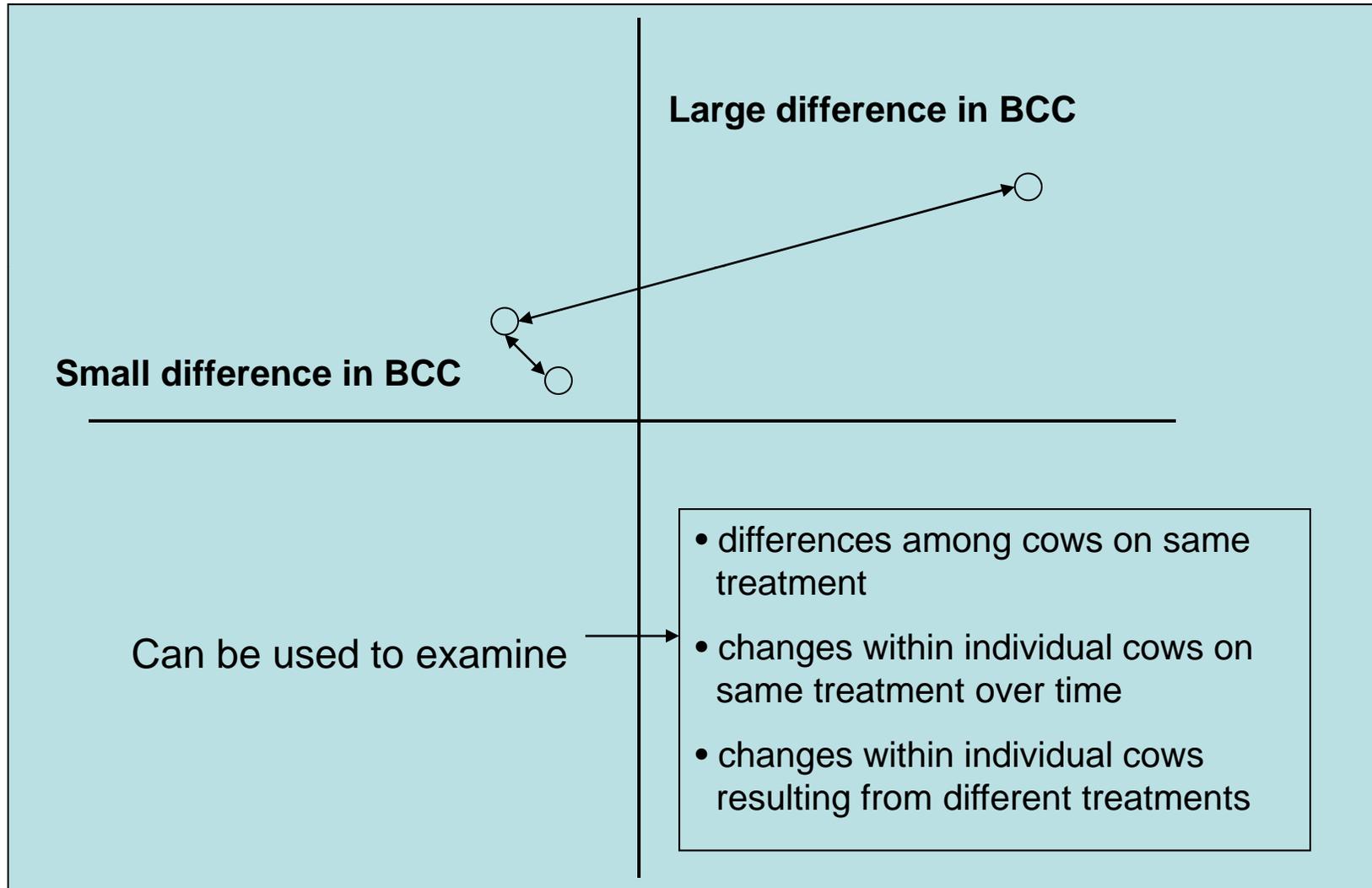
| ITS length | samp 1 | samp 2 | samp 3 | samp 4 |
|------------|--------|--------|--------|--------|
| 461 | 0 | 0.6960 | 1.1559 | 1.3742 |
| 462 | 0 | 0 | 0 | 2.2501 |
| 468 | 0 | 0 | 2.0226 | 2.336 |
| 472 | 0 | 0 | 0 | 0 |
| 474 | 0.6882 | 0.5221 | 1.0031 | 1.2493 |
| 477 | 0.9635 | 1.0141 | 1.4755 | 1.8457 |
| 480 | 0.6669 | 1.4532 | 2.1095 | 2.6771 |
| 482 | 0 | 0 | 0 | 0 |
| 484 | 0.572 | 0.5045 | 0.9139 | 1.0761 |



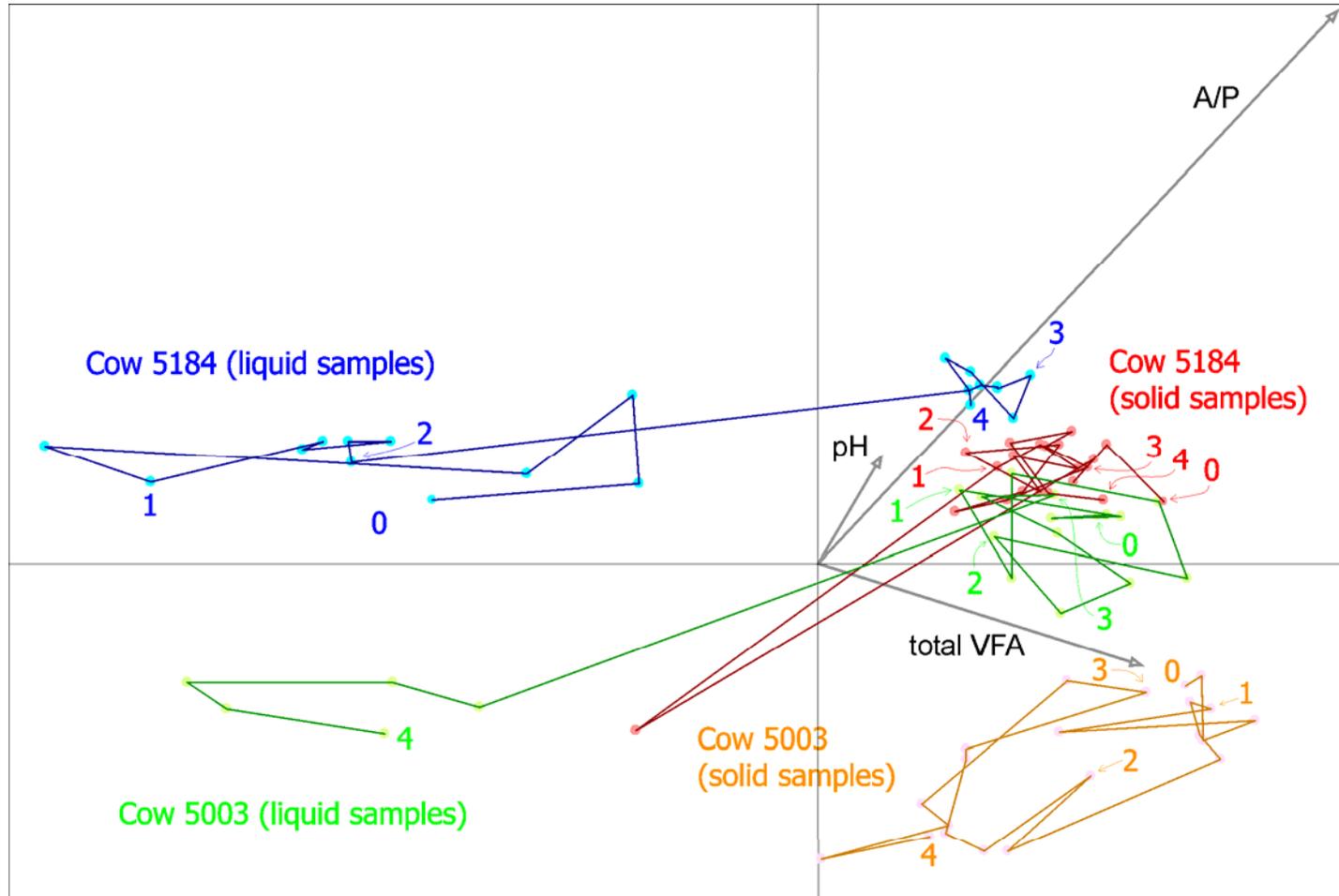
Multivariate statistical
analysis (CA)

Construct data matrix (typically 100 to 300 rows ALs)

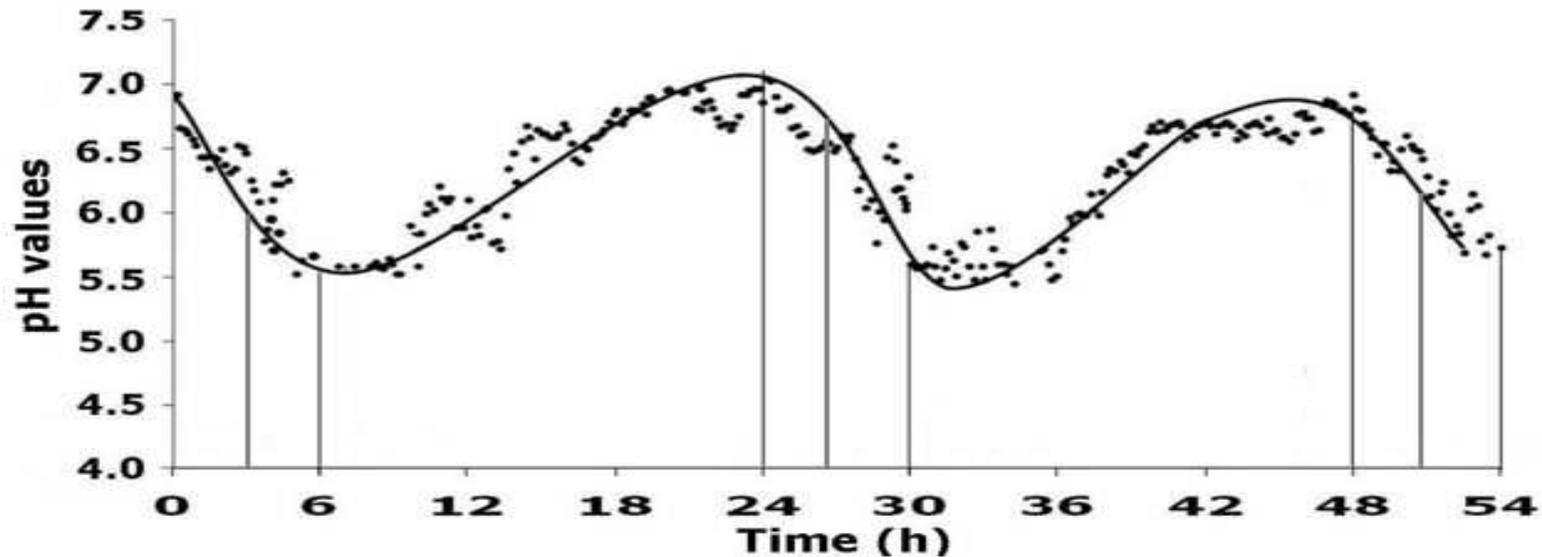
Ordination Bi-Plots from Correspondence Analysis



Bacterial populations change during the feeding cycle



Rumen pH changes during the feeding cycle



Palmonari et al., JDS 93:279 (2010)

Do cows having different rumen pH profiles differ in their microbial communities?

Experiment with 8 cows with continuous pH monitors, fed the same TMR

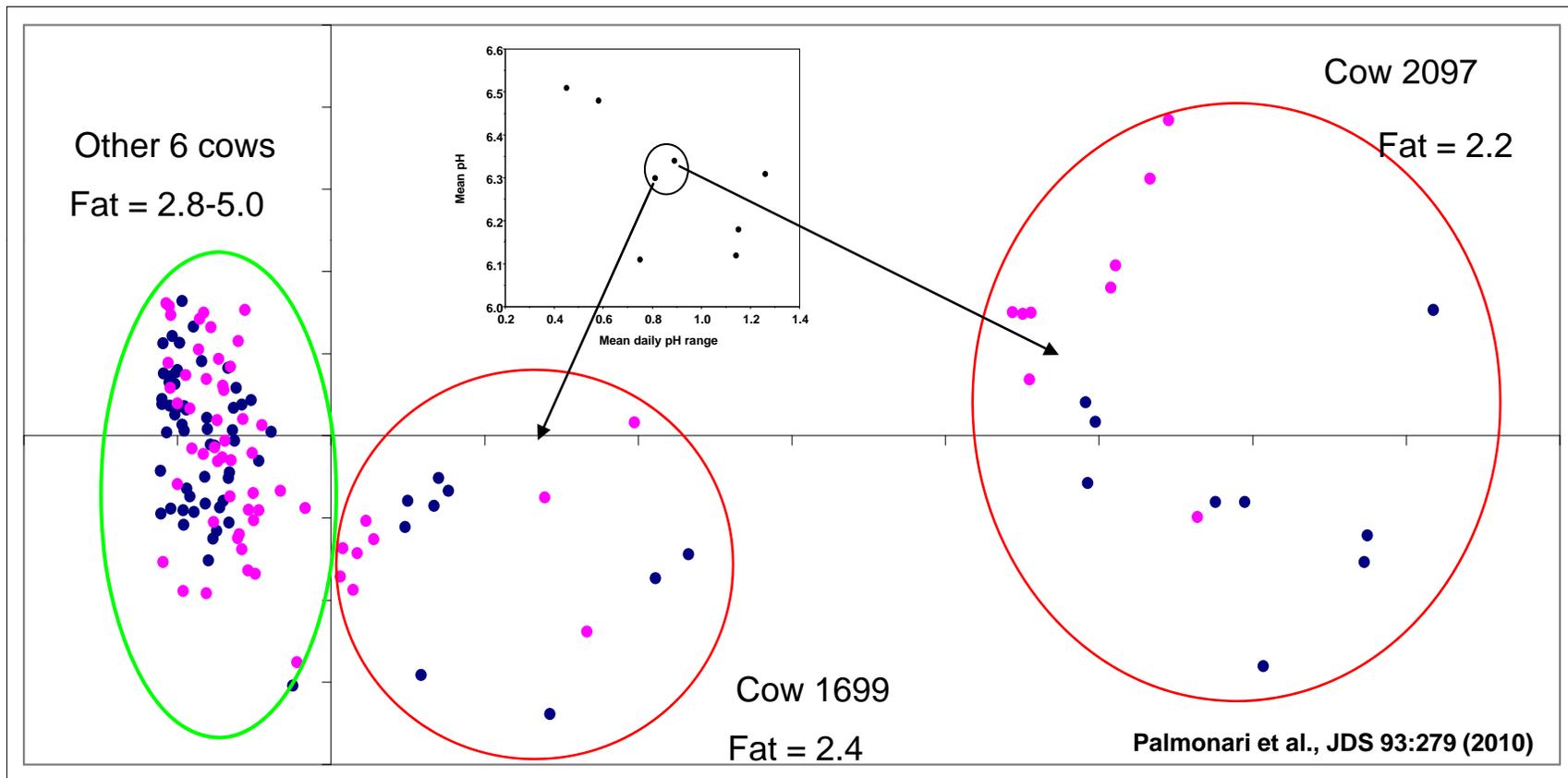
Profiles were used to calculate daily mean pH and daily pH range

6.11-6.51

0.42-1.28

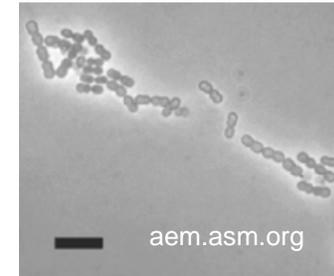
pH Dynamics Study

BCC in ruminal solid and liquid phases examined by ARISA.



BCC was independent of pH dynamics, but the most unusual BCCs were observed in cows that exhibited milk fat depression.

Megasphaera elsdenii populations are elevated in MFD cows



| Fat test | Cow | Proportion of <i>M.elsdenii</i> (% of bacterial 16S rRNA gene copies) |
|-----------------|------------|---|
| Depressed | 1699 | 0.320 ^b |
| | 2097 | 1.946 ^a |
| Not depressed | 1272 | 0.018 ^b |
| | 2088 | 0.032 ^b |
| SED | | 0.27 |

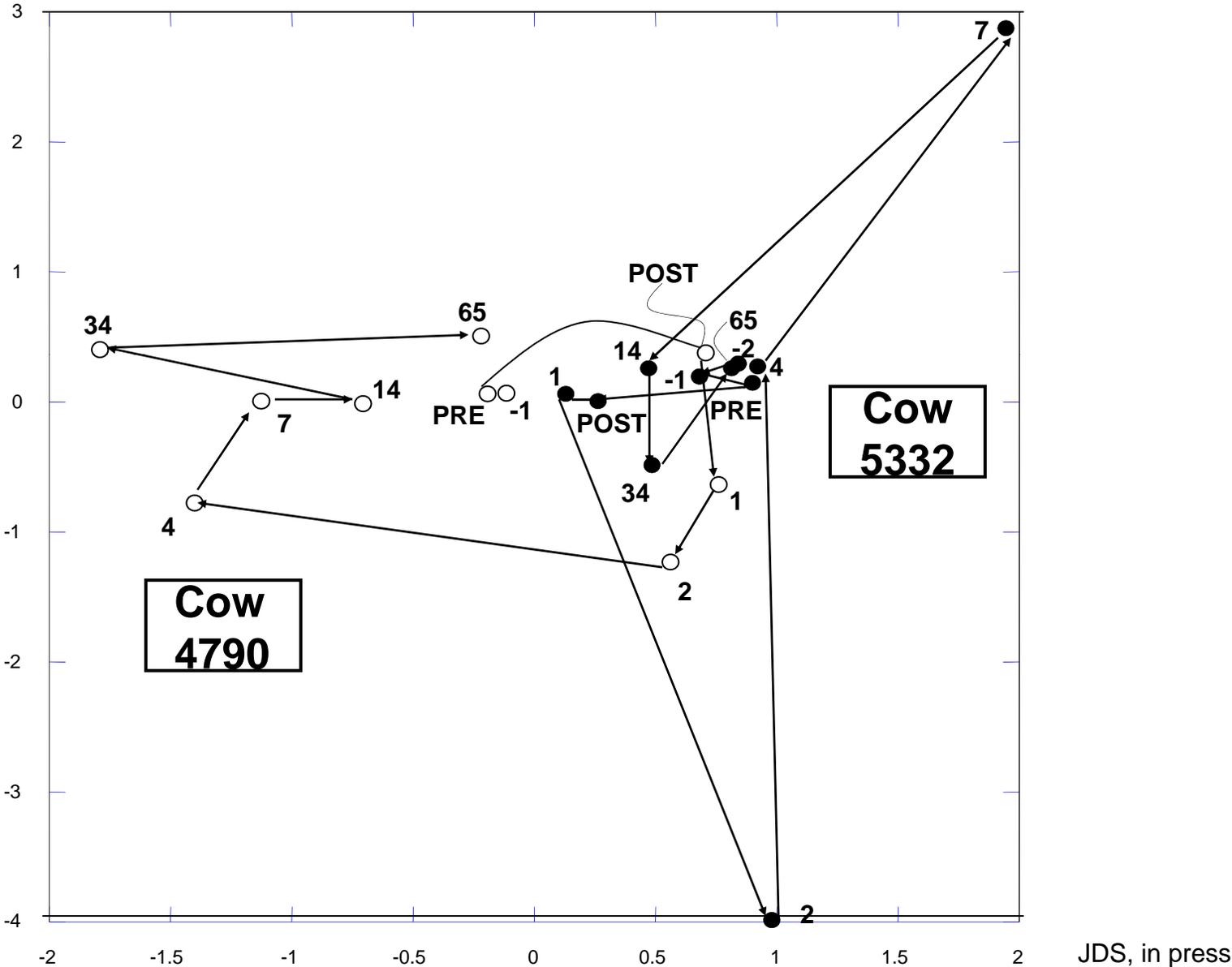
^{a,b} Means with different superscripts differ (P<0.05)

How stable and “individualized” is the rumen microbial community ?

“Rumen exchange” experiments

- Adapt rumen-fistulated cows to same TMR.
- Use ARISA to identify cows having largest difference in rumen bacterial composition.
- Exchange ~95% of rumen contents.
- Examine changes in BCC and rumen chemistry (pH, VFA composition over time.

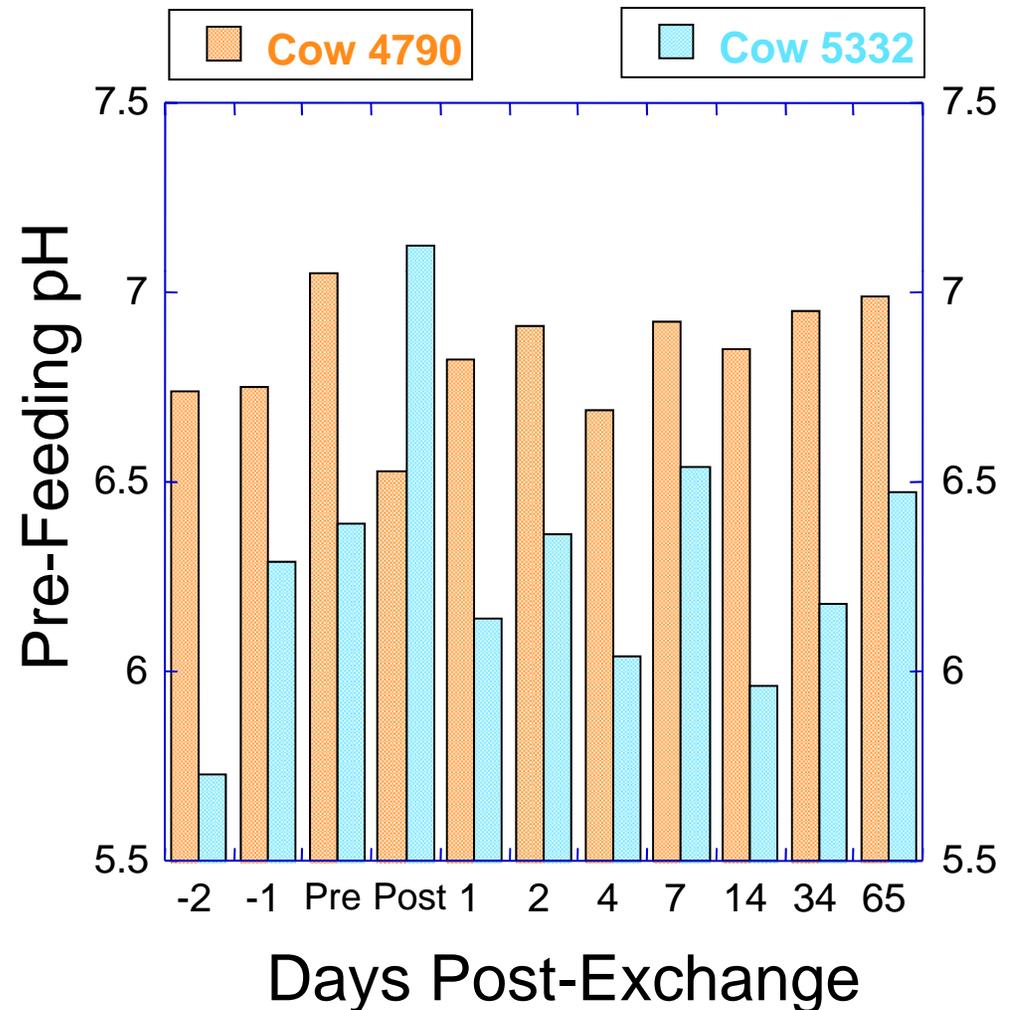
BCC changes following near-total exchange of rumen contents.



Rumen pH rapidly re-stabilizes after exchange of rumen contents

- Pre-feeding pH returned to previous level within 1 day of switch

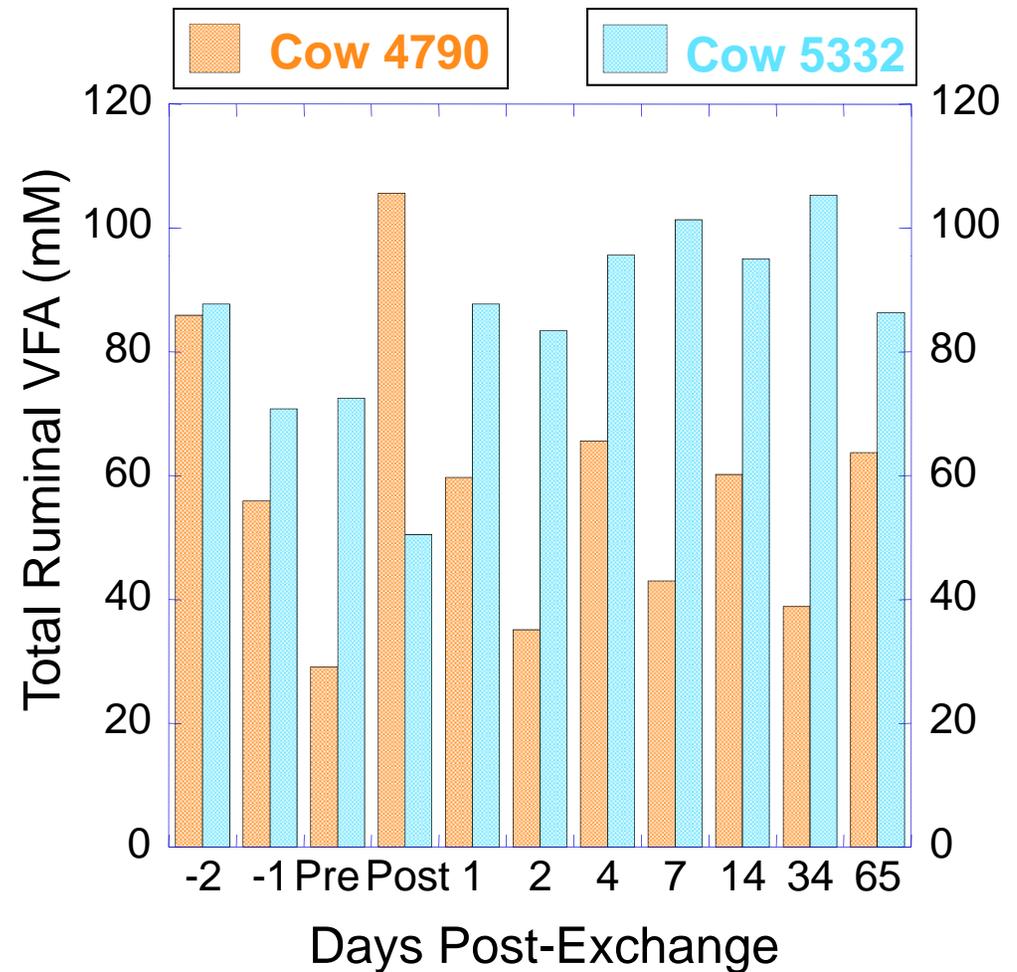
| Mean pH (pre-feed) | Cow 4790 | Cow 5332 |
|--------------------|----------|----------|
| Pre-exchange | 6.85 | 6.14 |
| Post-exchange | 6.88 | 6.24 |



Total ruminal VFA rapidly re-stabilizes after exchange of rumen contents

- Total VFA returned to pre-exchange pattern within 1 day of exchange

| Mean mM VFA (pre-feed) | Cow 4790 | Cow 5332 |
|---------------------------|-------------|-------------|
| Pre-exchange | 57.0 | 77.0 |
| Post-exchange | 52.3 | 93.5 |



TAKE-HOME

The cow and her ruminal microflora are co-adapted.

Though the ruminal microbes produce the VFA, the cow has a large say in dictating her own rumen chemistry.

To keep your cows happy, keep their microflora happy:

- Avoid sudden dietary changes
- Moderate rumen environmental conditions (temperature, pH, etc.)
- Be alert for exploitable differences among cows

THANK YOU !

This presentation will be posted on the
U.S. Dairy Forage Research Center
web site next week.

www.ars.usda.gov/mwa/madison/dfrc

or “Google” dairy forage research

