SmartCow

an integrated infrastructure for increased research capability and innovation in the European cattle sector

WP5 - Evaluation and standardization of nutrient use efficiency and emission measurement techniques

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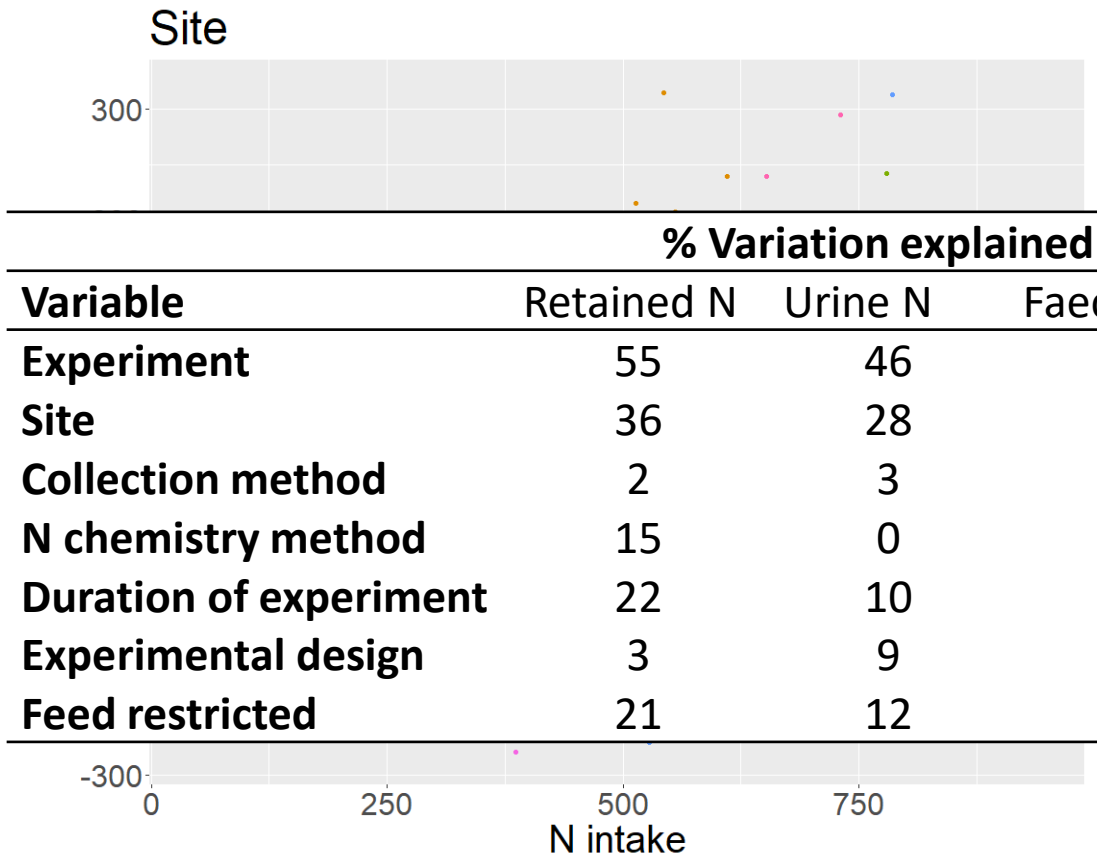
SmartCow – WP5 *Approach and methodology of the WP*

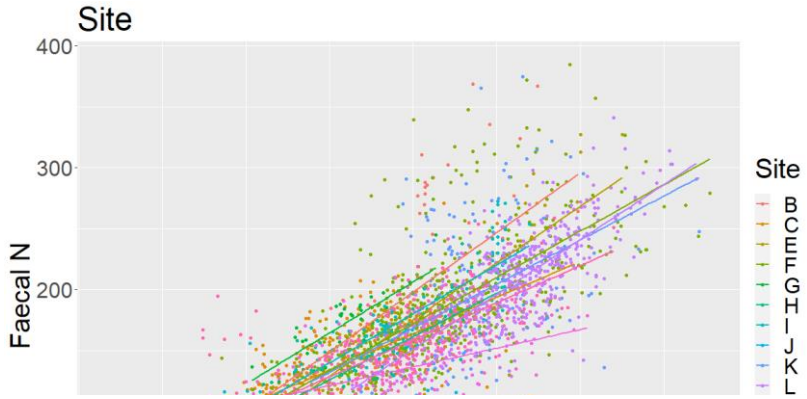
- **AIM:** To identify and address sources of variation in key *in vivo* measurements of dietary nutrient use efficiency and associated emissions of methane and nitrogen by cattle
- **Key Measurements within SmartCow:**
  - Feed digestion and N balance, including excretion in urine and faeces (feed and N efficiency)
  - Methane emissions
- *Key concerns: precision, accuracy, animal welfare*
- **Results will be assembled and analyzed to determine variation due to location, sources of experimental error, and animal variation.**
  - *measurement error addressed to:*
    - *Increase precision (Refine) and Reduce animal use*

SmartCow – WP5 *Meta-Analysis of N balance data*

Aim: To explain sources of variation

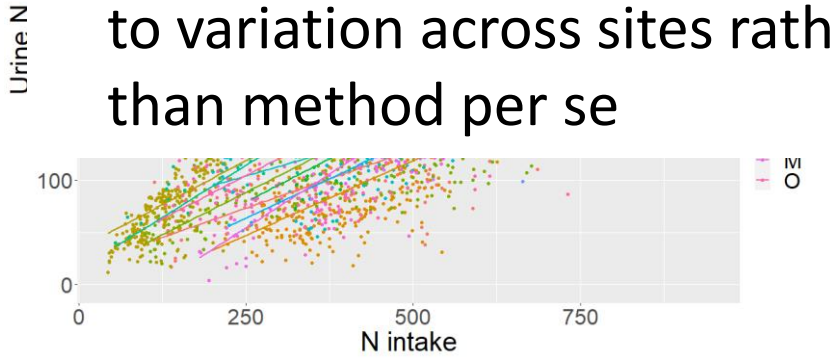


	% Variation explained		
Variable	Retained N	Urine N	Faecal N
Experiment	55	46	60
Site	36	28	38
Collection method	2	3	9
N chemistry method	15	0	7
Duration of experiment	22	10	20
Experimental design	3	9	22
Feed restricted	21	12	4



Sources of variation

- Experiment and Site explain a high proportion of the variation in the data
- Suggests how methods are used (human behaviour) contributes to variation across sites rather than method per se



SmartCow – WP5 : DIGESTIBILITY & N BALANCE EQUIPMENT

ls

New urine and faecal collection equipment

- New steer equipment developed
  Vacuum system, stir plates for acid
- Skin ‘safe’ glues for dairy cow urine collection cups
  New designs for ‘modern’ cows – one size does not fit all!

Local Assessments of Sources of Variance

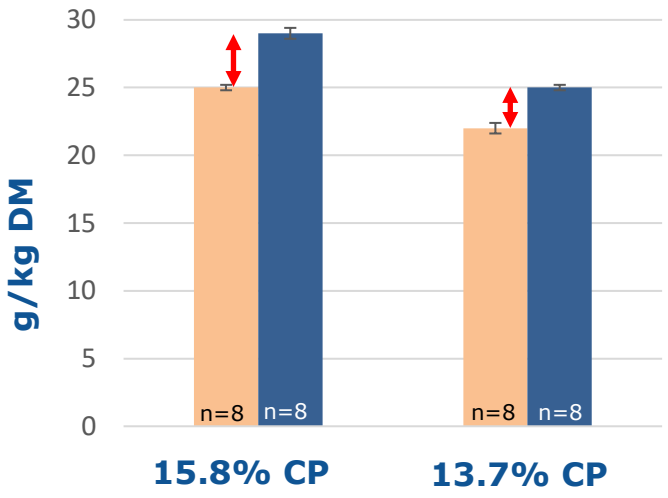
Sources of variation for digestion and N balance measurements evaluated at each SmartCow Installation

- Method evaluation as appropriate for local methods, e.g.:
- Days of sampling (INRAE and Uread)
- Volatile N losses and N analysis method (WU and FBN)
  - N intake higher with Dumas N analysis of feed (~4.5%)
  - Measuring volatile N loss reduces N balance
- Spot sampling for faecal markers (AU)
  - 3 samplings (morning, early afternoon, late afternoon) over 2 consecutive days .
- Results reviewed and discussed (January Workshop)
  - Agreed key factors – especially attention to detail!

FBN for WP5

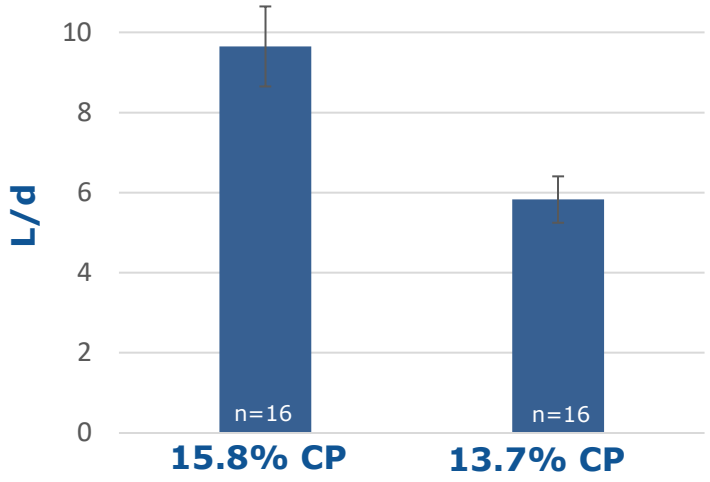
Deviations in N-intake due to different sample processing

Dried feed: 60°C, 2d
Frozen fresh feed: ground in CO_2



11- 14% Difference

Quantifying volatile N-losses while not collecting urine/faeces



4- 7 g excreta N loss /d

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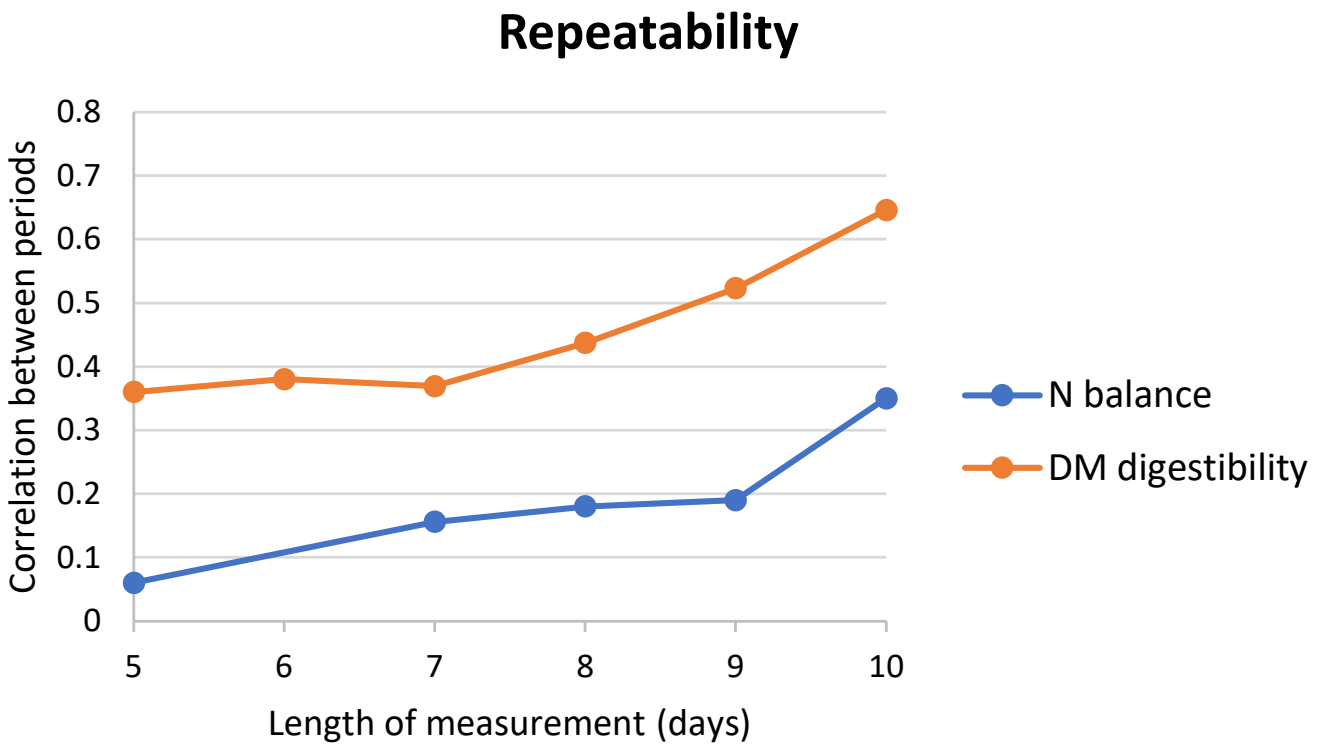
INRAE - Repeatability according to the length of measurement

Digestion and N Balance over 2 measurement periods with 16 growing bulls receiving 2 diets



Repeatability increases with the length of collection period with values around 65% and 35% for DM digestibility and N balance, respectively, for 10 days

Bellagi et al., EAAP 2021, Animal in preparation

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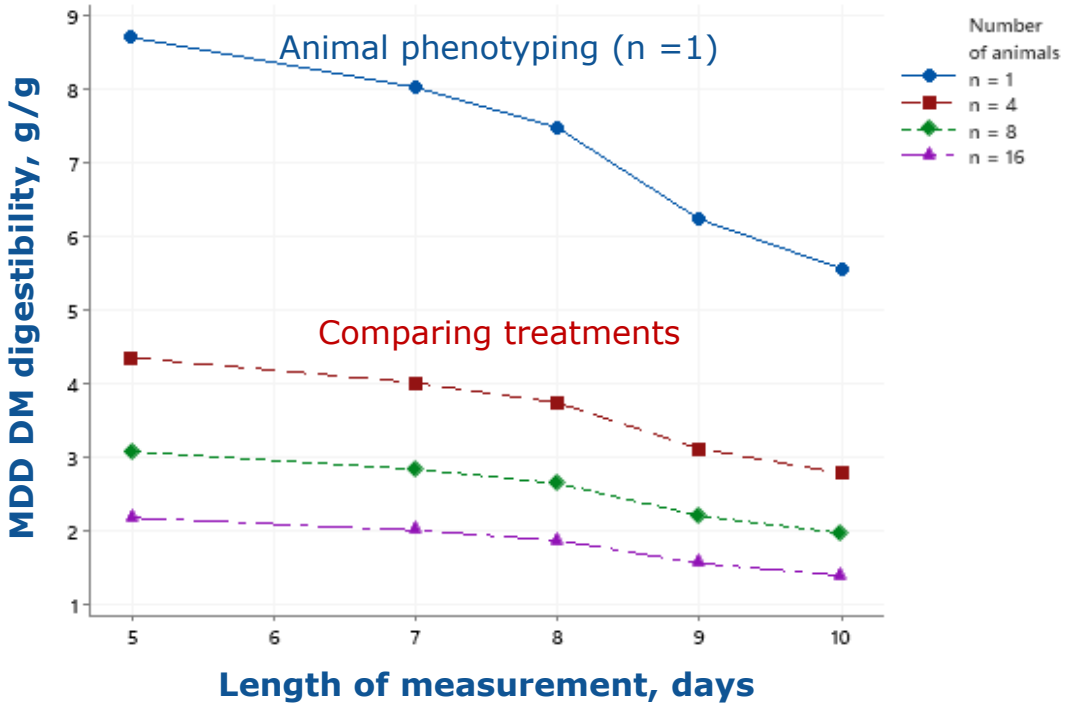
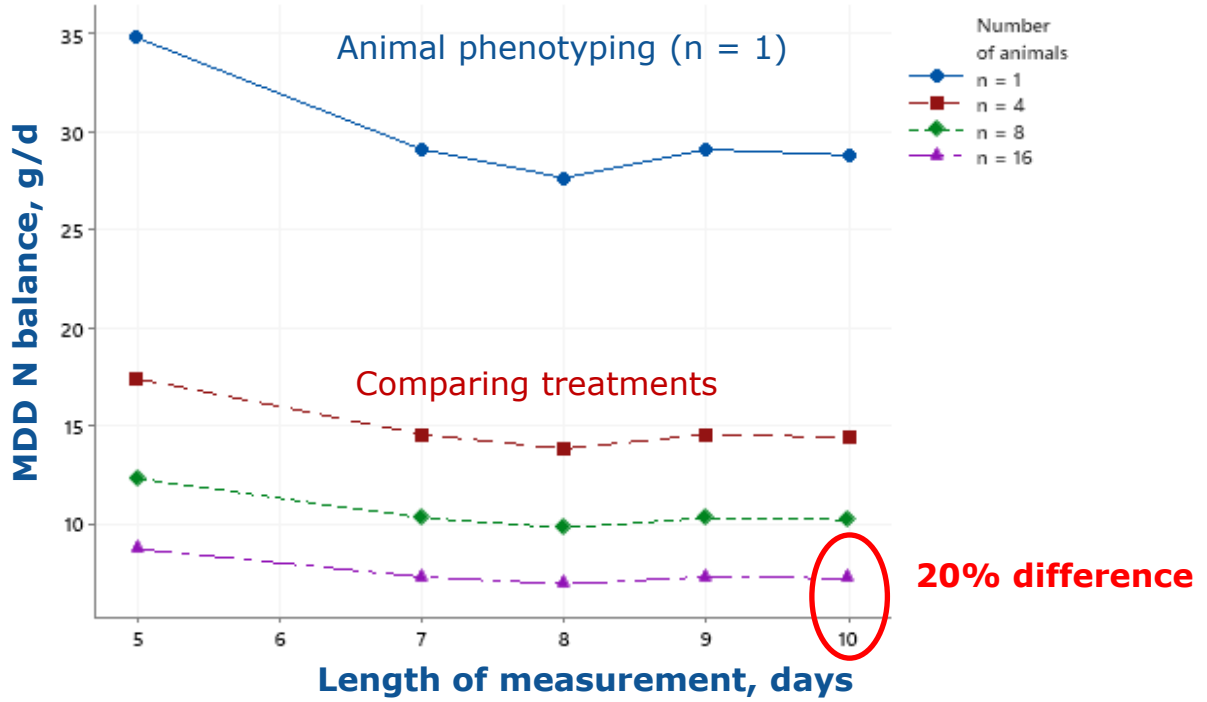
INRAE – Minimum detectable difference (MDD) according to the length of measurement

Bellagi et al., EAAP 2022, Animal in preparation

DM DIGESTIBILITY, g/g



N BALANCE, g/d



The minimum detectable difference decreased with the length of measurements and number of animals:

With 16 animals/treatment and 10d of measurement a difference of 20% in N balance between diets can be significantly detected

Some Conclusions Digestion and N Balance

- Strict attention to detail and protocols critical to success
- Assessment of impact of specific procedures
  - Number of days of collection
    - conclusions similar for N balance at URead (lactating cows)
    - Variation in intake a key factor - cow comfort very important
    - 7 to 10 days improved accuracy at INRAE (growing bulls)
  - Feed and faecal handling
    - Immediate analysis the 'gold standard' but frozen bulk samples acceptable
    - Faecal preservatives – acid ethanol decreased N concentration
    - Losses of ammonia minimized with attention to detail
  - Urine handling – acidification and mixing critical
    - Lower urine N concentrations overall for frozen bulk samples
      - 7.17 vs 6.75 and 10.39 vs 9.97 g/kg for low and high CP diets, respectively
  - Method of sample drying and grinding for N analysis – avoid drying!
    - Chop if needed using dry ice

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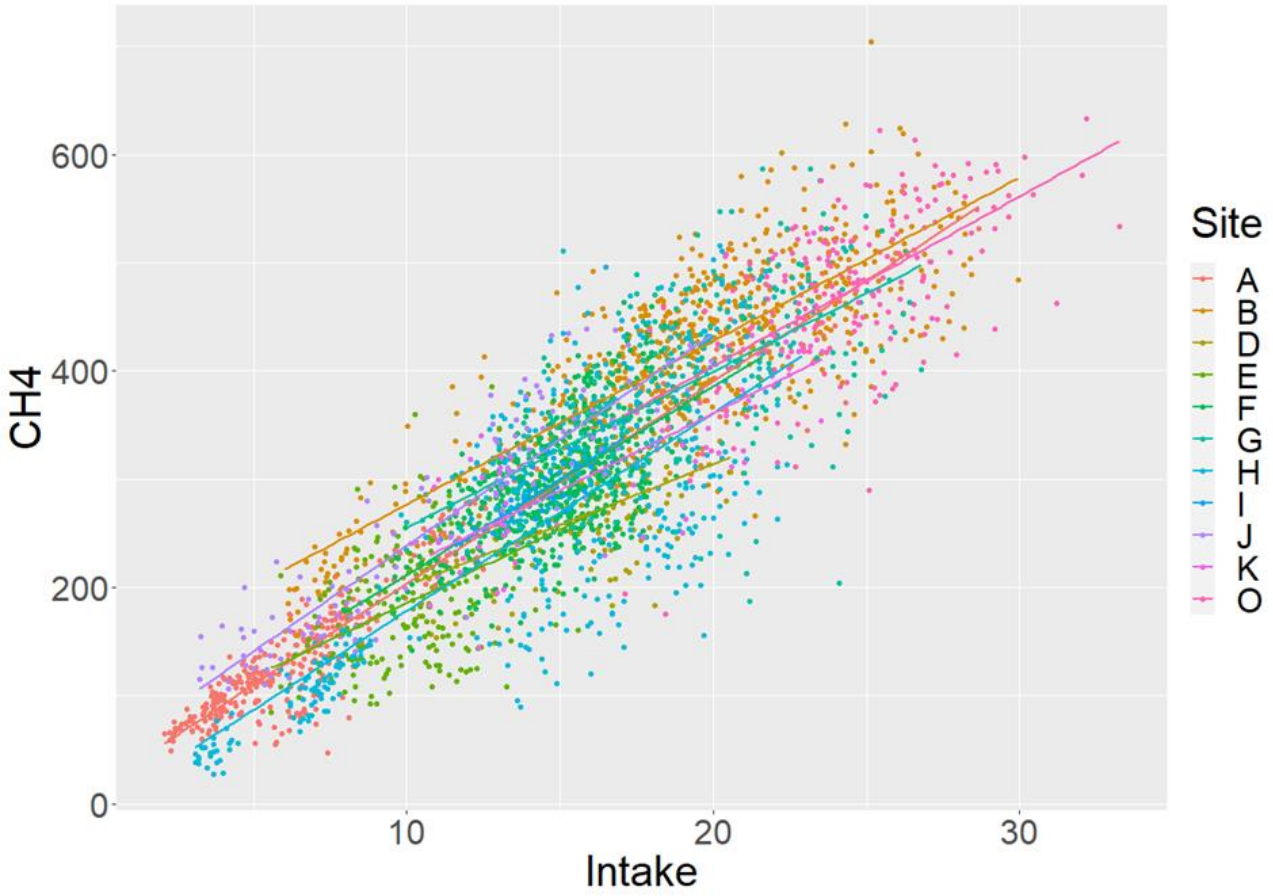
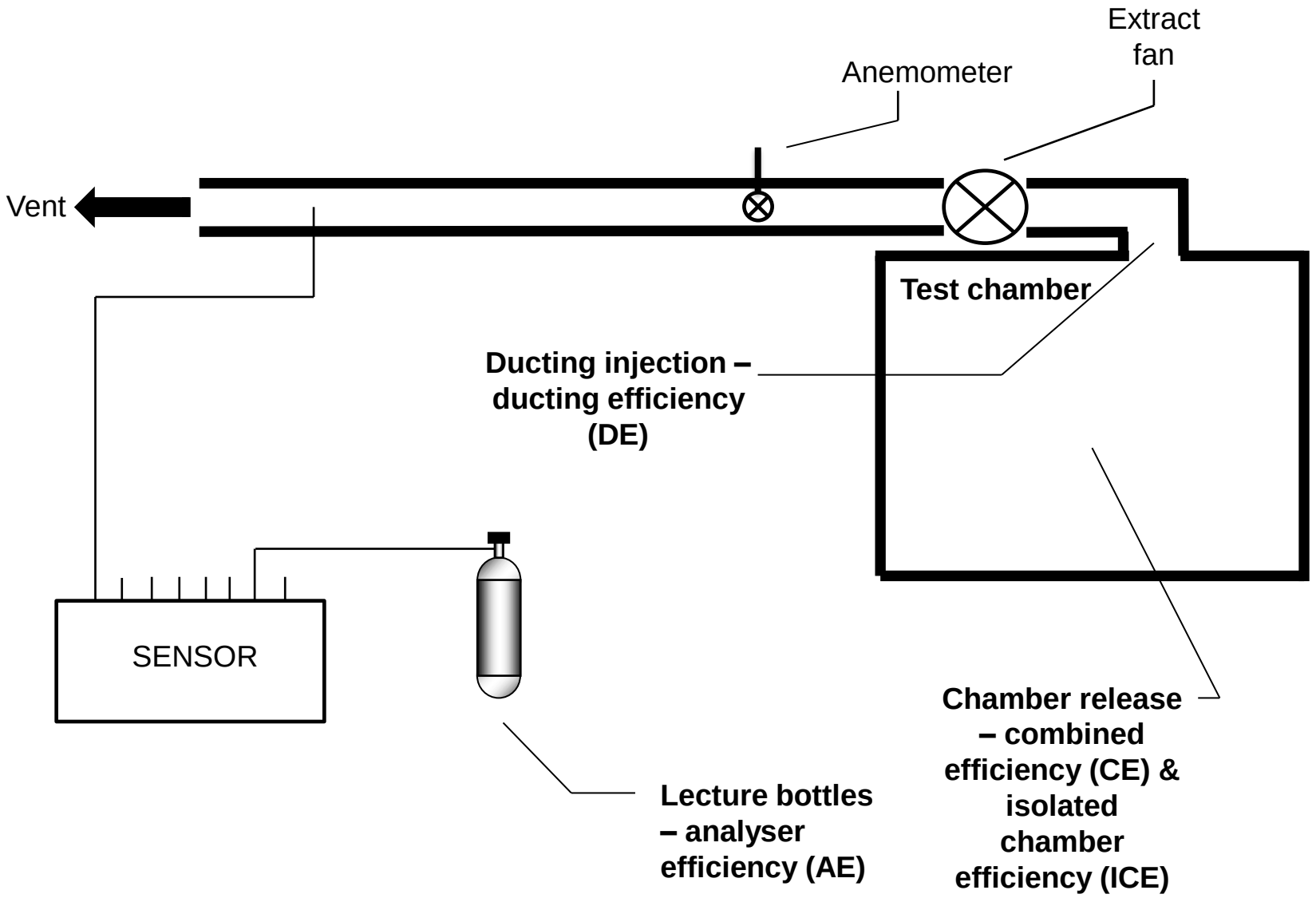
Meta-Analysis of Methane Emission Measurements



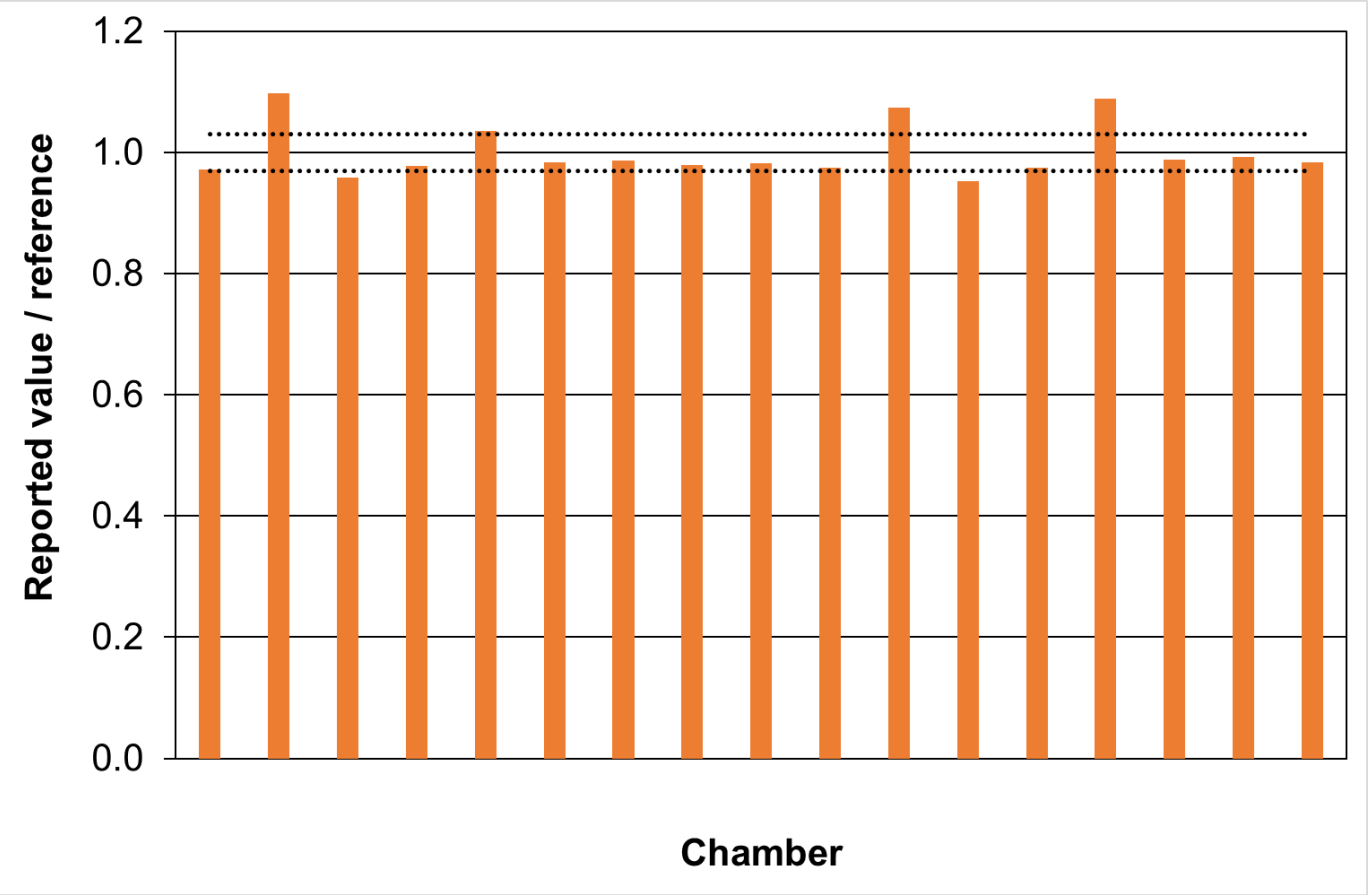
% Variation		
Experiment	Site	Residual
49.5	8.9	41.6

For chamber measurements variation due to site relatively minor after adjustment for other sources of variation (e.g. trial, diet composition)

SmartCow – WP 5 Ring Test of Chamber Methane Recovery by NPL



SmartCow – WP 5 *CH4 Recovery - Chamber Efficiencies*



Ring-test	Comparability / % (*k*=2, 95% confidence)
UK (2012)	25.7
SmartCow	6.2

- 6 instances (out of 17) where variance > +/-3%
- Guidance on identifying sources of variation (e.g. flow meter calibration, leaks)
- Recommeded that ring tests be conducted periodically to validate local recovery tests

SmartCow – WP 5 Implications

Digestion and N balance measurements

- Sources of variation at each installation evaluated and addressed
- Improvements in ‘standard’ methods achieved through self assessment and critical evaluation
- Human behaviour critical – strict attention to detail, training and engagement of research staff, students, etc.
- Cow comfort and acclimation to equipment important
- Accurate measurement of intake and diet composition critical!

Chamber measurements of methane emission

- Recovery tests show good comparability across installations
- Potential sources of variation highlighted

Improvements and standardization achieved at SmartCow installations an exemplar for other researchers

SmartCow – WP 5 Future Prospects

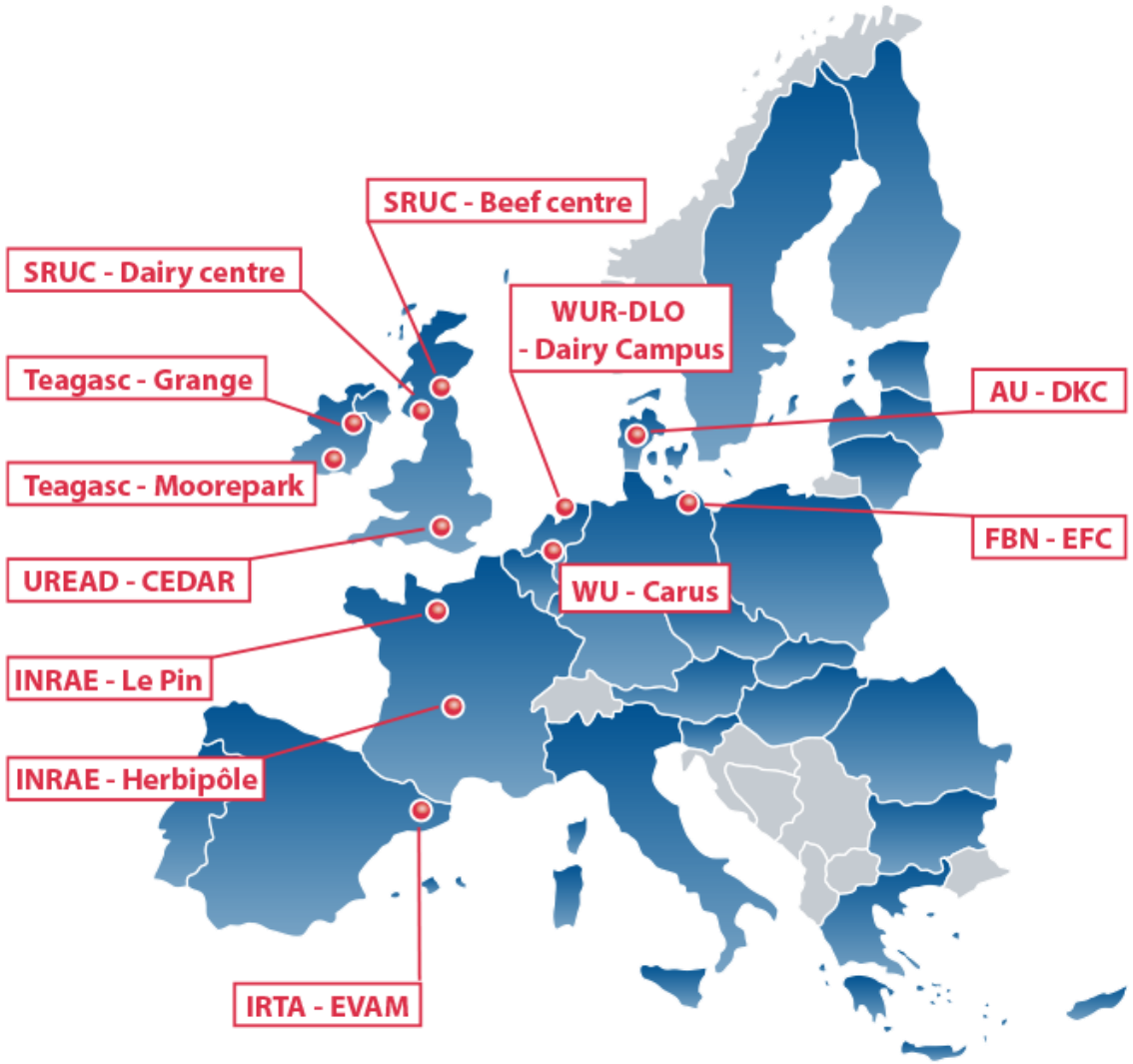
Digestion and N balance measurements

- Still need to address questions raised
  - Effects of freezing on urine N concentration (Uread results)
  - Effects of method of analysis on N concentration of feeds (WU results)
- Sharing of best practice
  - Workshops for training of staff and early career researchers
- Development and adoption of proxy/biomarker estimates

Chamber measurements of methane emission

- Establishment of user group for sharing equipment, training, etc.
  - Future ring-tests for updating validation of facility efficiencies
  - Workshops for training of staff and early career researchers
- Development and adoption of proxy/biomarker estimates

Thank you for your attention



First-class Cattle Research Infrastructures (RIs) across Europe:

- 11 major RIs distributed in 7 EU countries
- 12 locations, which include 18 installations
- 2500 dairy and 1000 beef cows

- **Networking of RIs** to inventorize resources, harmonize procedures, and share data
- **Joint research activities** to improve experimental methods and phenotyping capability
- **Interaction with stakeholders** to stay in line with industry needs and improve dissemination

http://www.smartcow.eu/stakeholders/

TRAINING PROGRAM

For Scientists, Technicians, Stakeholders, PhD students

- Face-to-face training courses
- Free web-conferences
- One-day study tours in 4 different countries

http://www.smartcow.eu/resources/training/

TRANSNATIONAL ACCESS CALLS

Offers external users (academic and industry) free access to SmartCow RIs

- 30 projects during the 4 years of SmartCow
- Access to around 10,000 cow-weeks

http://www.smartcow.eu/calls/

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