



Genotype *plus* Environment
Integration for a more sustainable dairy production system



Prediction of energy status of dairy cows using milk MIR spectra

C. Grelet¹, A. Vanlierde¹, M. Salavati, M². Hostens³, L. Foldager⁴, F. Dehareng¹ & Gpluse Consortium⁵

¹ Walloon Agricultural Research Center (CRA-W), Belgium,

² Royal Veterinary College (RVC), United Kingdom;

³ Ghent University, Belgium;

⁴ Aarhus University, Denmark;

⁵ <http://www.gpluse.eu/index.php/project/partners/>



c.grelet@cra.wallonie.be

Global objective of the project

Optimise production efficiency, fertility, health and environmental footprint of dairy cows:

First step:

Develop models predicting **phenotypes of interest** based on easily-measured and large scale **biomarkers in milk:**

- Milk glycan
- Milk metabolites
- Milk MIR spectra**

Final steps:

Genomic study : Genome-Wide Association Study (GWAS) to relate genotype to phenotypes of interest
~15 000 cows

→ **Breeding**

Management study: new management strategies at herd and cow level to improve phenotypes of interest
~600 cows

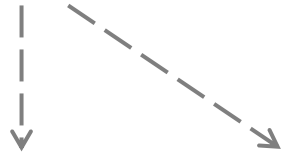
→ **Advisory tools**



Objective

Develop models predicting **phenotypes of interest**:

MIR —————> Energy status:



Fertility:

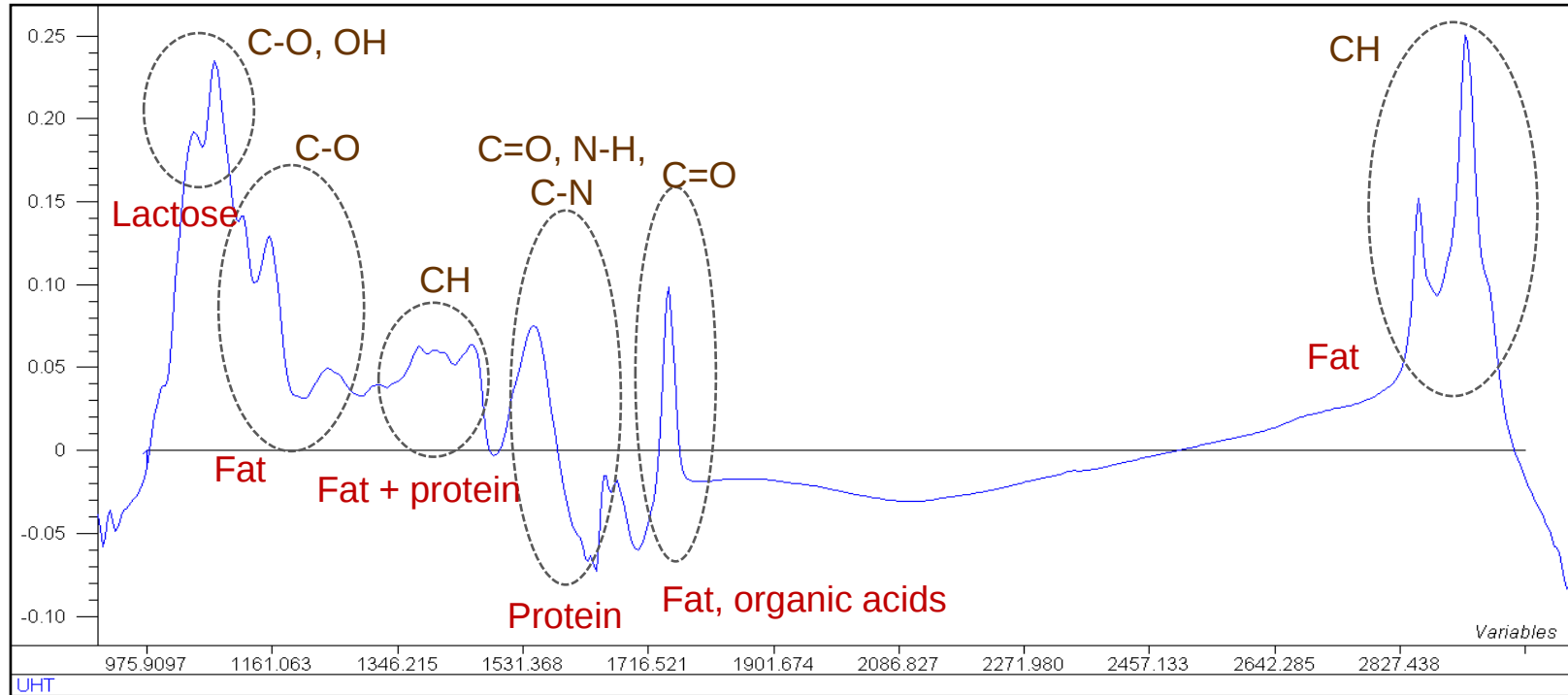
- IGF-I level and LH inhibition (Butler, 2000)
- Uterine health (Hammon, 2006)

Health:

- Inflammation (Esposito 2013)
- Immune response (Hammon, 2006)
- Digestive issues: ketosis, DA... (Collard, 2000)
- Locomotive issues (Collard, 2000)



Why MIR?

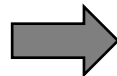


- **Position** of the peaks → **Qualitative** analysis
- **Intensity** of the peaks → **Quantitative** analysis

Why MIR?

- Already available
- Fast
- Cheap

Milk control



MIR



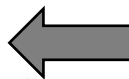
© Bentley



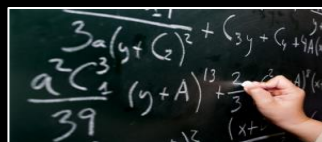
Milk composition

Fat
Proteins
Urea
Lactose

...

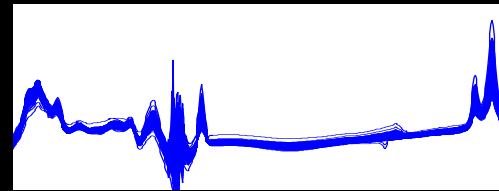


Models



+

MIR spectra of each cow

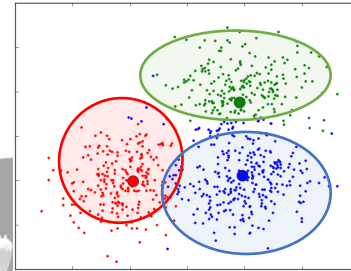


Objective

Develop models predicting **phenotypes of interest**:

MIR $\longrightarrow$ Energy status:

- Feed related phenotypes
- Blood components
- Global metabolic status using the clusters methodology



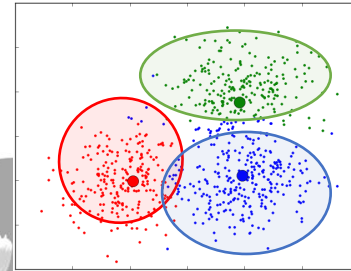
Objective

Develop models predicting **phenotypes of interest**:

MIR

Energy status:

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Materials & Methods



Experiments

- 6 experimental farms
- Common sampling protocol
- 235 cows
- Holsteins
- Calving to DIM 50

	# Cows	PP	MP	MY	Feed diet roughage
AFBI (UK)	61	18	43	31.6	Grass
AU (Denmark)	35	11	14	35.5	Corn/grass
CRAI (Italy)	45	8	37	29.3	Triticale
CRA-W (Belgium)	31	13	18	30.5	Corn/grass/sugarbeet
FBN (Germany)	27	3	24	37.5	Corn/grass
UCD (Ireland)	36	2	34	30.5	Corn/grass
TOTAL	235	55	180	32.1	

Phenotypes of interest

Feed related phenotypes

- Energy balance = $NE_{Intake} - NE_{Milk} - NE_{Maintenance}$ ([NRC, 2001](#))
- $RFI = DMI - PDMI$ (with $PDMI = (0.372 * FCM + 0.0968 * BW^{0.75}) * (1 - e^{(-0.192 * (WeekOfLactation + 3.67))})$) ([NRC, 2001](#))
- $DMI_{Observed}$

Measured daily

Daily DMI measurement in 3 farms (132 cows)



Phenotypes of interest

Blood plasma metabolites and hormones

- Glucose ~ energy
- IGF-I ~ liver status
- NEFA ~ mobilization
- BHB ~ (sub)clinical ketosis

DIM14 & 35

Sampling in the 6 farms (235 cows)

Analyzed at UCD & Aarhus university



Phenotypes of interest

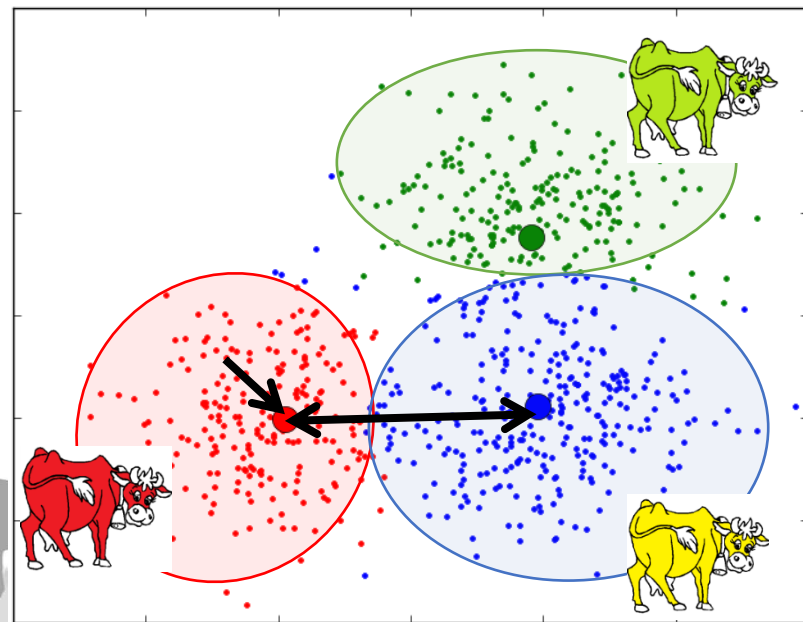
Blood metabolites and hormones are usefull when used togheter

→ Clusters:

- Combination of the blood metabolites and hormones into 3 groups of cow status

K-means nearest neighbour clustering

K=3



Phenotypes of interest

- Metabolic clusters

3 : healthy cows



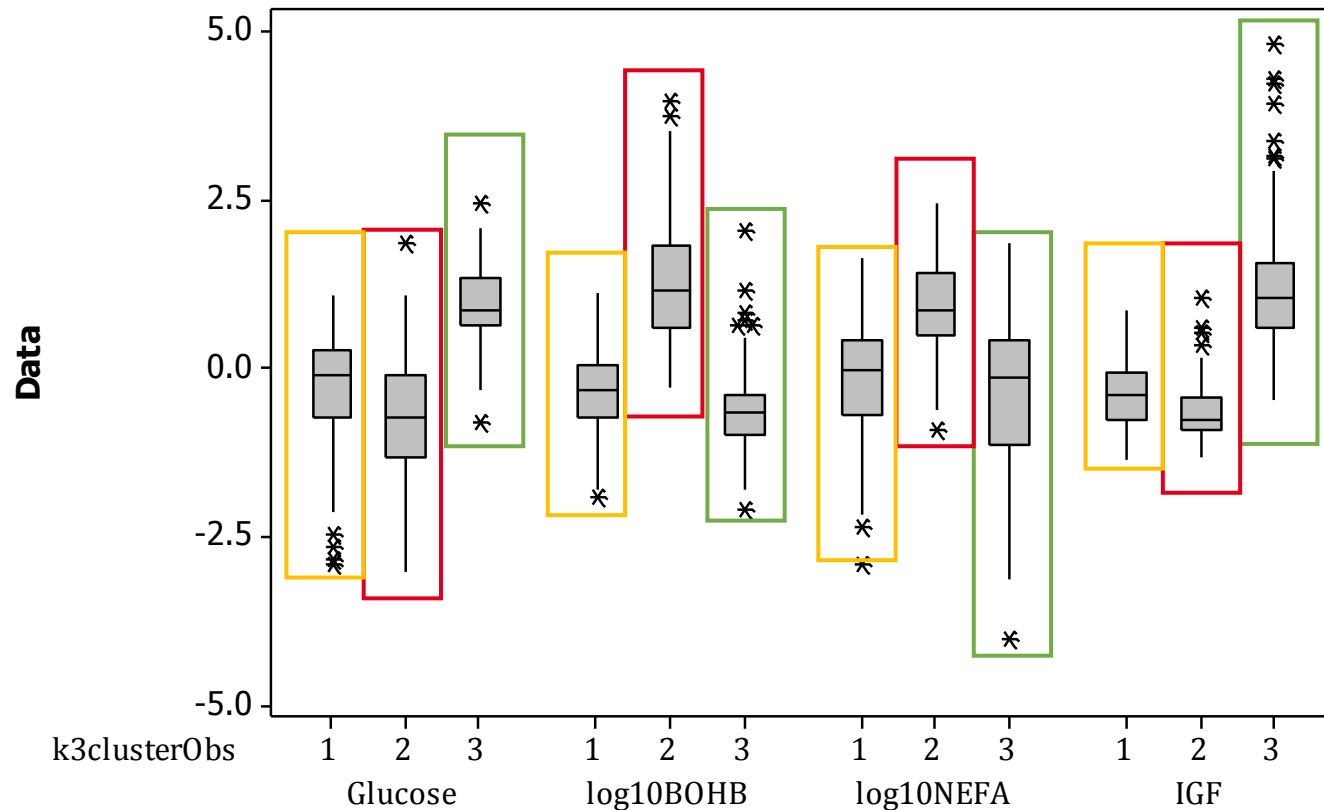
1 : intermediate cows



2 : imbalanced cows



Boxplots of metabolites and hormones following clusters



MIR analysis of milk

- Analyzed locally or at CRA-W
- Twice per week
- AM & PM (weighted average)
- Foss and Delta



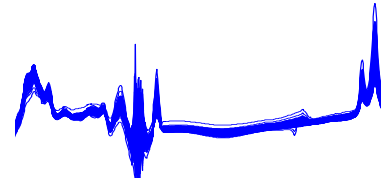
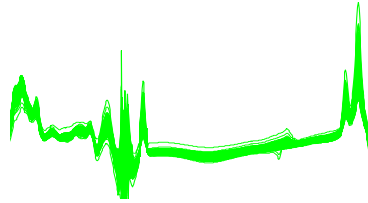
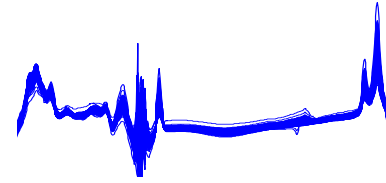
MIR standardisation

- Different instruments
- Standardisation step needed



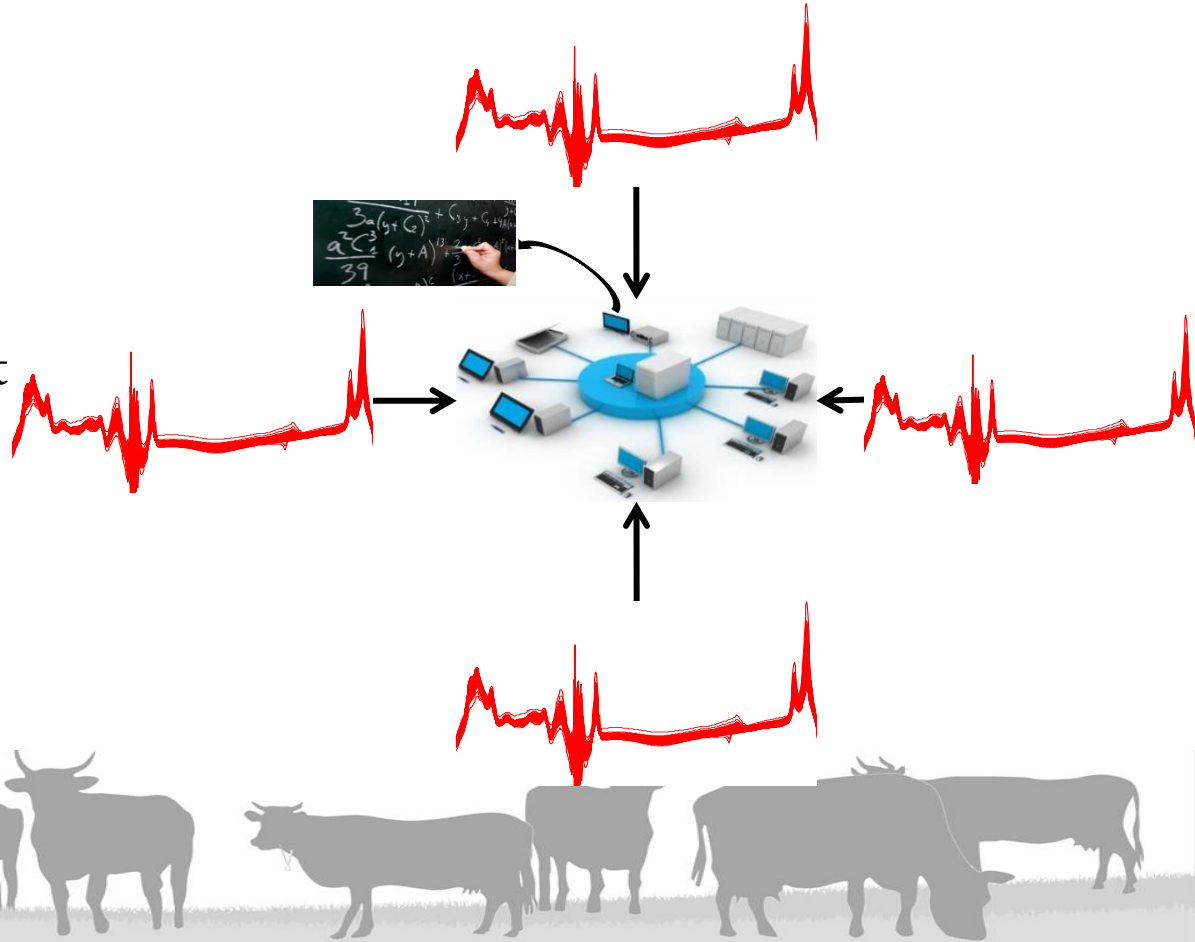
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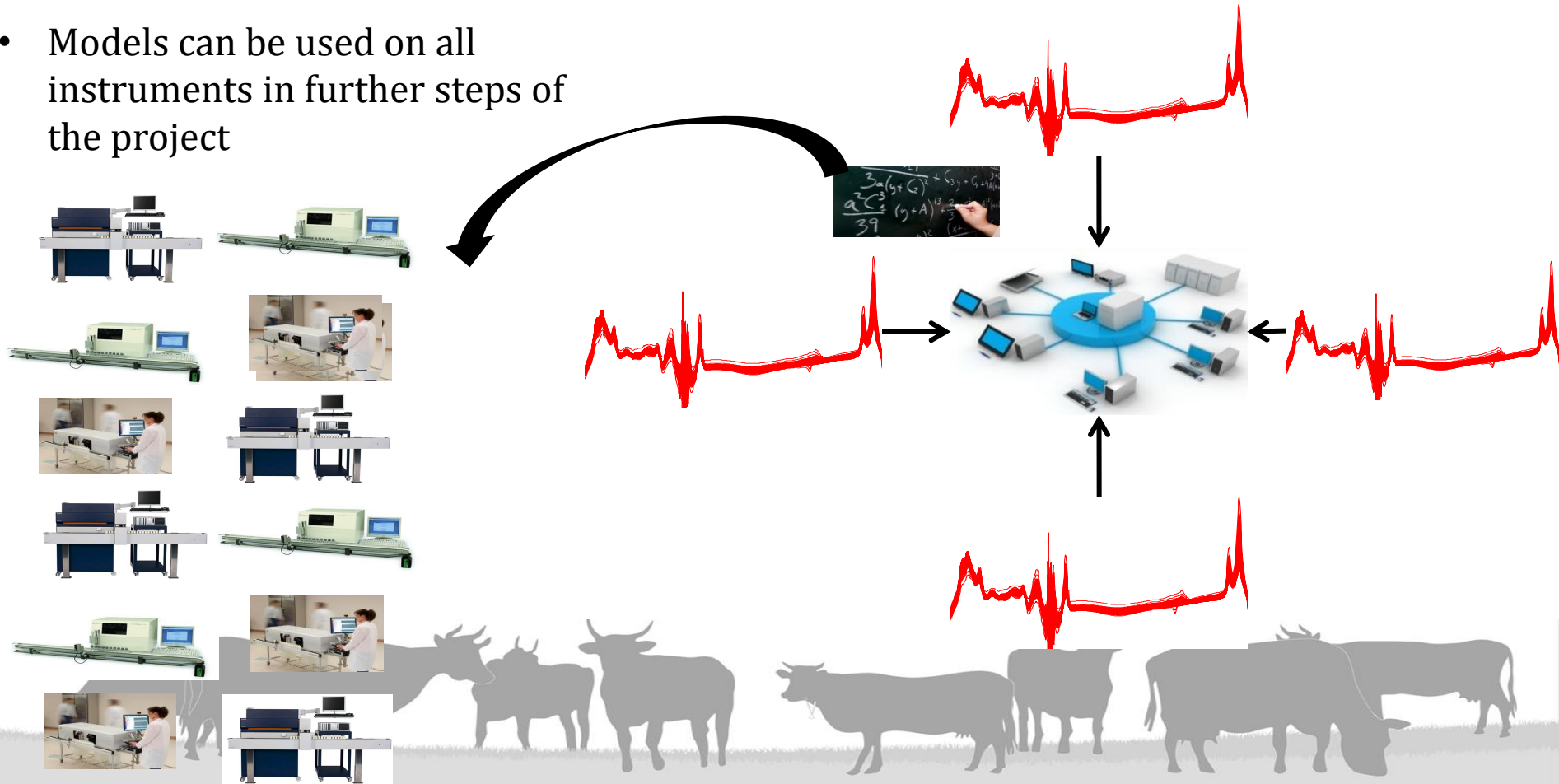
MIR standardisation

- 
- Harmonize the spectral format
- Creation of common models



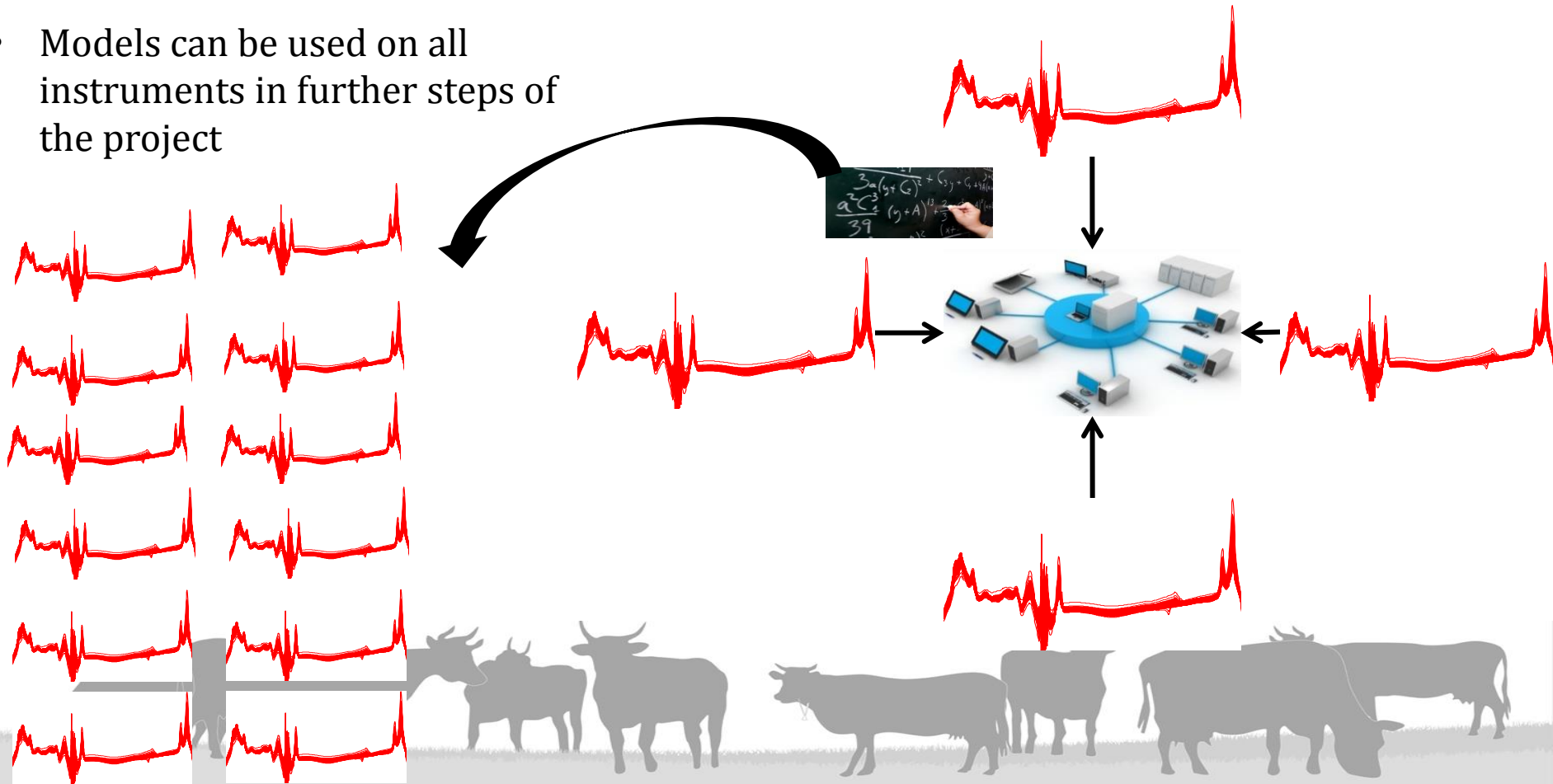
MIR standardisation

- Models can be used on all instruments in further steps of the project



MIR standardisation

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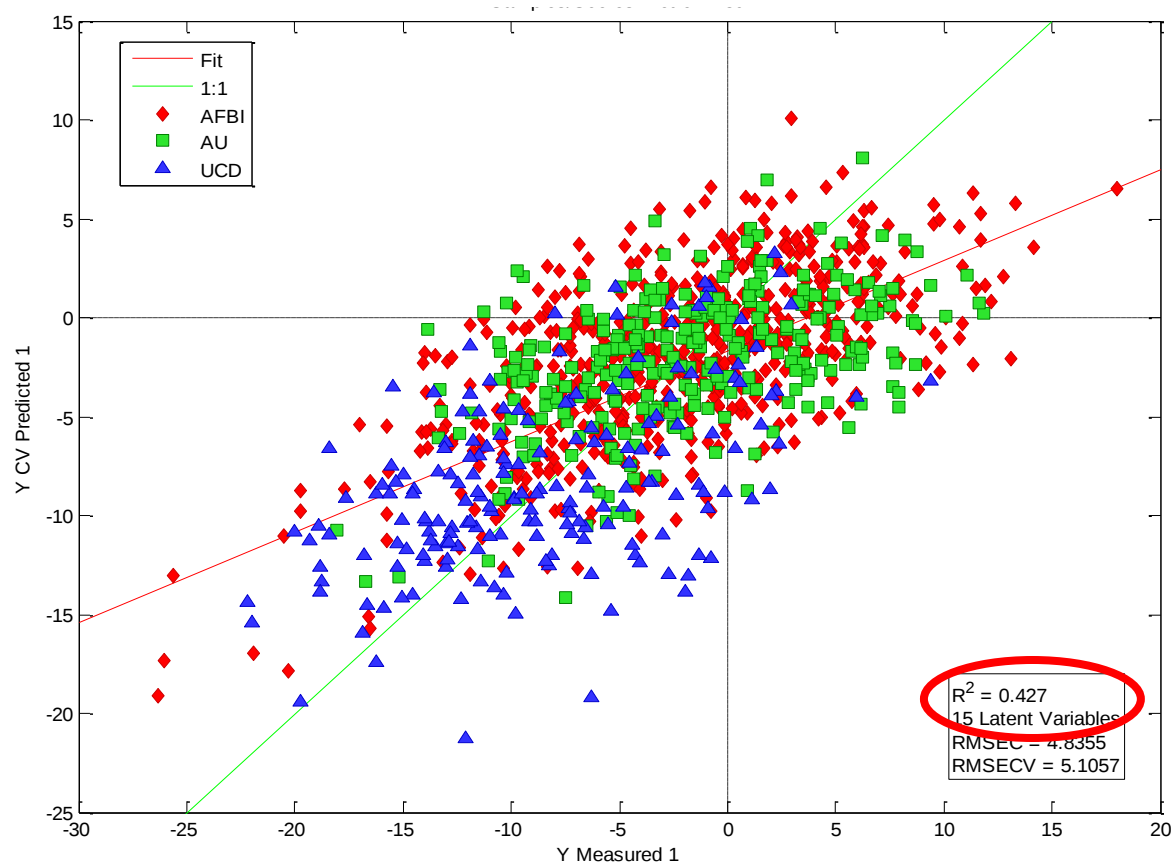


Results



MIR prediction of feed related phenotypes

- Energy balance (Mcal/d)
- PLS
- 132 cows
- 1069 data in the model
- 15 latent variables
- CV 10 subgroups



MIR prediction of feed related phenotypes

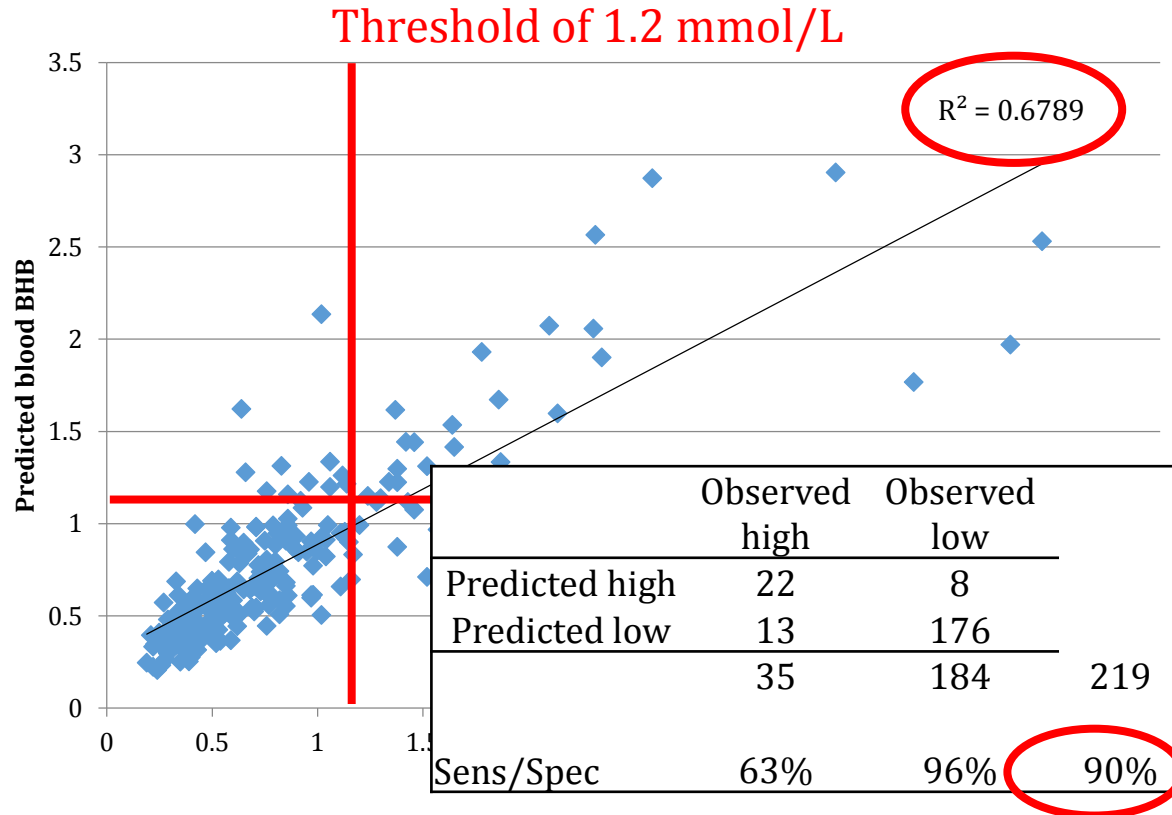
- PLS, LOOCV

	Predictor traits	n	#LV	R ² _{cv}	RMSECV
Energy balance (Mcal/d)	MIR	1069	15	0.43	5.1
Energy balance (Mcal/d)	MIR, DIM, FPCM	1098	12	0.51	4.8
RFI (kg/d)	MIR	1115	14	0.46	2.9
RFI (kg/d)	MIR, DIM, FPCM	1097	14	0.53	2.7
DMI (kg/d)	MIR	1052	14	0.47	3.2
DMI (kg/d)	MIR, DIM, FPCM	1098	8	0.59	2.9



MIR prediction of blood metabolites and hormones

- BHB blood
- PLS
- 235 cows
- 219 samples
- Log transformation
- Random removing of low values
- LOOCV
- 8 LV



MIR prediction of blood metabolites and hormones

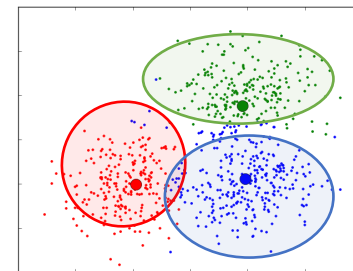
- PLS, LOOCV

Phenotype	n	#LV	R ² _{cv}	RMSECV
Blood NEFA (micro-ekv/L)	392	12	0.37	302
Blood Glucose (mmol/L)	378	8	0.43	0.39
Blood BHB (mmol/L)	219	8	0.68	0.37
Blood IGF-I (mg/L)	378	12	0.59	47.18



MIR prediction of energy status clusters

- MIR, MY, Parity
- PLS-DA, 6LV



Healthy Intermediate Imbalanced



Predicted
Healthy



90

31

8

Predicted
Intermediate



15

110

15

Predicted
Imbalanced



8

33

54

113

174

77

364



MIR prediction of energy status clusters

- MIR, MY, Parity
- PLS-DA, 6LV

		Healthy Intermediate Imbalanced					Glob
					Sens	Spec	acc.
Predicted Healthy		90	31	8	80%	84%	
Predicted Intermediate		15	110	15	63%	84%	70%
Predicted Imbalanced		8	33	54	70%	86%	
		113	174	77	364		

MIR prediction of energy status clusters

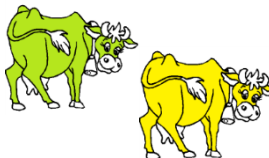
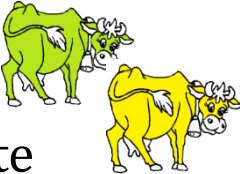
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Predicted Imbalanced		8	33	54	70%	86%	
		113	174	77	364		

Only 4.4% of extreme misclassification

MIR prediction of energy status clusters

- MIR, MY, Parity
- PLS-DA, 6LV

	Healthy-or-Intermediate		Imbalanced			
						
Predicted Healthy or Intermediate		246	23		Sens 70%	Spec 86% Glob acc. 82%
Predicted Imbalanced		41	54			
		287	77	364		

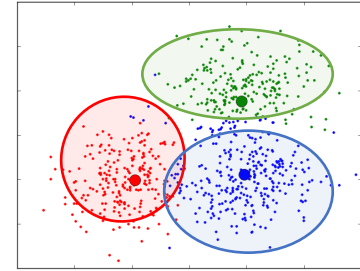


Conclusions

- MIR models on energy status:
 - Direct EB, RFI or DMI
 - Blood BHB and IGF-I
 - Global metabolic status using the clusters methodology



- Variable accuracy following traits
- To be used in genomic studies
- Herd or individual cows management tools



Thank you for your attention!

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