β-lactoglobulin genotype prediction based on milk Fourier Transform InfraRed (FTIR) spectra

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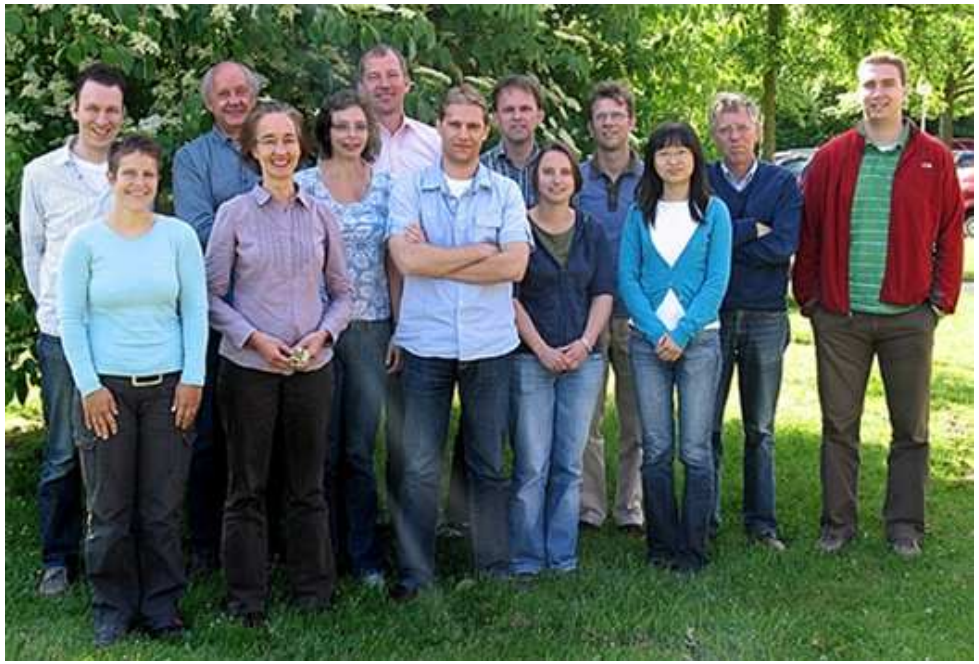
Milk Genomics project team



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Introduction

- β-Lactoglobulin (β-LG) genotypes are quantitatively associated with milk protein composition
- β-LG BB milk has (Heck et al., 2008):
  - Increased casein content

    ~3% higher cheese yield for BB as compared to AA (v/d Berg et al. 1992)
  - Increased α-lactalbumin content
  - Decreased β-LG content

} Infant nutrition

Introduction

β-LG A and B variants differ qualitatively – physicochemical properties

- Denaturation temperatures differ – β-LG AA milk results in stronger fouling of heaters (Hill et al. 1997)

β-LG genotypes are associated with milk characteristics and therefore might be subject of selection.

→ This requires information on β-LG genotypes

Introduction

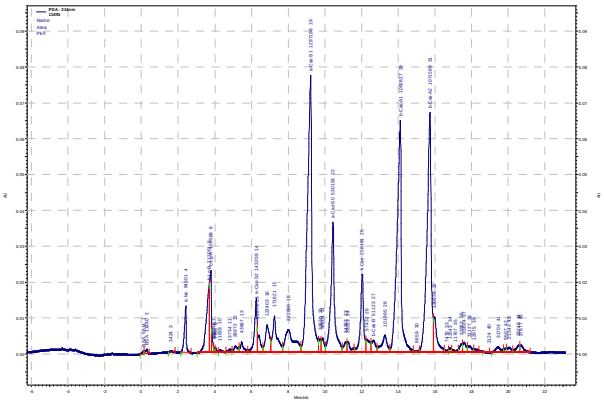
Genotyping

- at the protein level

  Iso-electric focussing

  Capillary Zone Electrophoresis

  .....

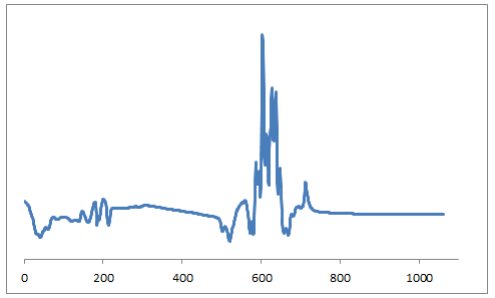


- at the DNA level

  β- LG genotypes of males might be available, not of most females.

Introduction

- FTIR is used to determine fat% and protein% in routinely collected milk samples
- FTIR also can be used to predict fat (e.g. Soyeurt et al., 2006) and protein composition (Rutten et al., in press)



Aim of the present study

Can FTIR be used to predict β-LG genotypes?

Material

- 1,669 Holstein Friesian heifers
- Cows were located on 395 different farms
- 4,326 FTIR spectra were available, cows with
  - 1 IR spectrum 0.9%
  - 2 IR spectra 15.4%
  - 3 IR spectra 83.7%
- Spectra relate to milk samples collected in winter, spring and summer
- β-LG genotypes were determined based on DNA analyses

Method

Calibration sample
n=500

β-LG AA → Y= -1
β-LG AB → Y = 0
β-LG BB → Y = 1

Prediction equation using PLS

Validation sample
n=3,826

Predicted β-LG genotypes:

$\hat{Y} \leq -0.5$ → β-LG AA
$-0.5 < \hat{Y} < 0.5$ → β-LG AB
$\hat{Y} \geq 0.5$ → β-LG BB

10 replicates

Results – predicting β-LG genotypes

Predicted	True		
	AA n=1,232	AB n=1,993	BB n=601
AA	76%		
AB			
BB			

Results – predicting β-LG genotypes

Predicted	True		
	AA n=1,232	AB n=1,993	BB n=601
AA	76%		
AB		80%	
BB			

Results – predicting β-LG genotypes

Predicted	True		
	AA n=1,232	AB n=1,993	BB n=601
AA	76%		
AB		80%	
BB			66%

Results – predicting β-LG genotypes

Predicted	True		
	AA n=1,232	AB n=1,993	BB n=601
AA	**76%**		
AB	23%	**80%**	
BB	1%		**66%**

Results – predicting β-LG genotypes

Predicted	True		
	AA n=1,232	AB n=1,993	BB n=601
AA	**76%**	12%	
AB	23%	**80%**	
BB	1%	8%	**66%**

Results – predicting β-LG genotypes

Predicted	True		
	AA n=1,232	AB n=1,993	BB n=601
AA	**76%**	12%	1%
AB	23%	**80%**	33%
BB	1%	8%	**66%**

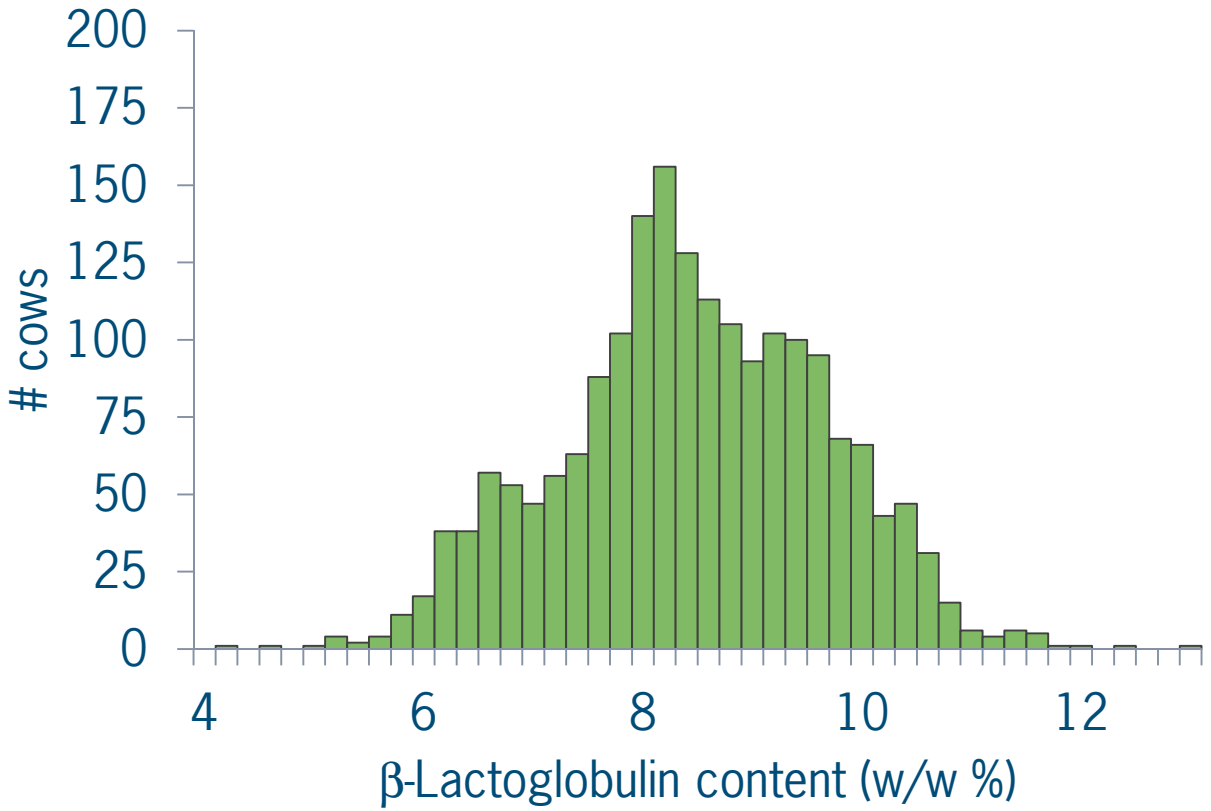
Discussion & Conclusions

- Milk FTIR spectra can be used to predict β-LG genotypes

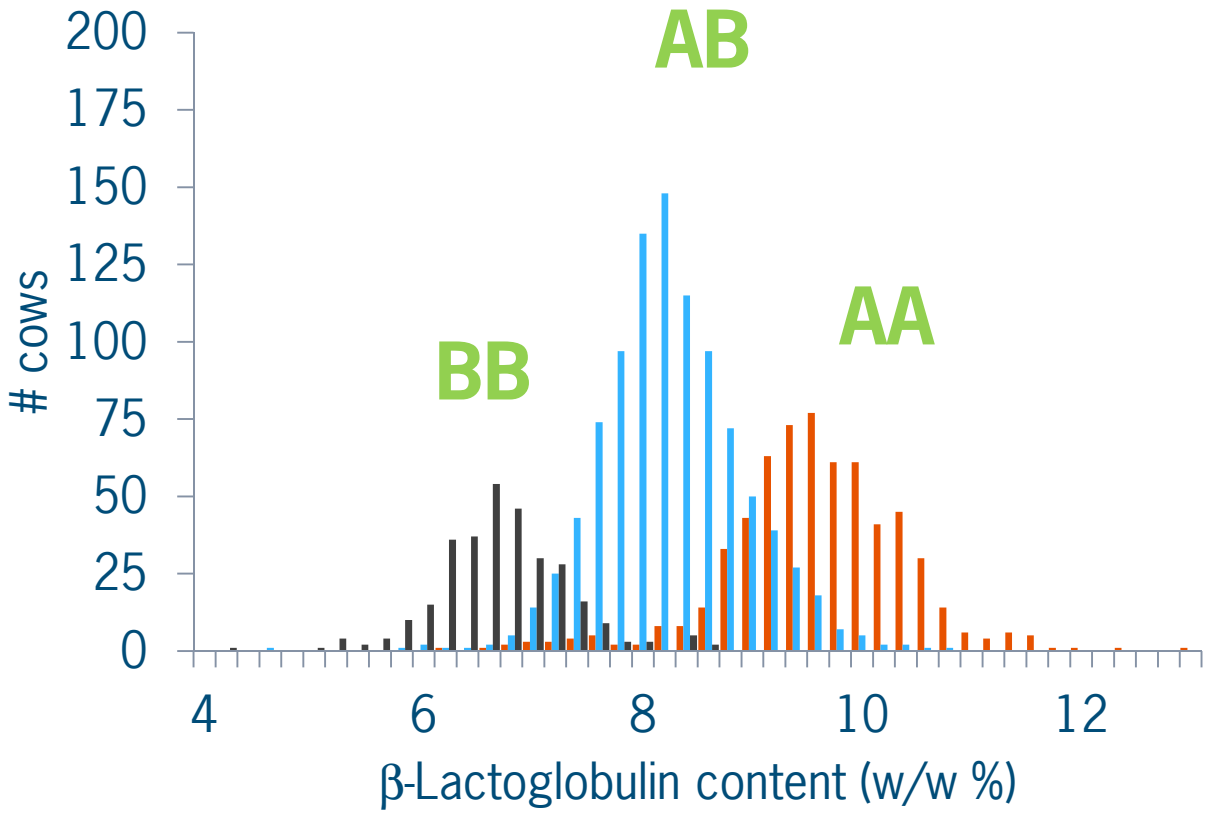
  77% of the samples assigned to the correct genotype

- Why is this possible?
  - β-LG genotypes show a strong association with β-LG content in milk – 80% of the genetic variation!!
  - Prediction of β-LG content based on FTIR: R^2=0.57 (Rutten et al. JDS in press)

Discussion & Conclusions



Discussion & Conclusions



Discussion & Conclusions

Scope for improving the presented procedure:

- In our prediction we did not utilize the fact that cows have repeated observations. Would this help?

  Repeatability of $\hat{Y}$ = 0.85

- We used a simple cut-off (-0.5 and 0.5) to translate $\hat{Y}$ into genotypes. Other approaches might do a better job, e.g. mixture models
- We did not use pedigree information. Improve genotype assignment e.g. by using methods presented by Gengler et al. (2007) – incorporate known sire genotypes

77% of the milk samples can be assigned to the correct β-LG genotypes based on Milk FTIR spectra.

Thanks for your attention