27.08.2012 EAAP – Closing the phenomic gap

Milk metabolites as new traits and their role for genetic evalution of traditional milk traits

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FOR FARM ANIMAL BIOLOGY

General information

genetic variation

phenotypic variation

SNPs (single nucleotide polymorphisms)

milk traits:
fat content
protein content

Genomic selection

50K

classical approach

Genomic selection

50K

classical approach

50K

reduced classical approach

Genomic selection

50K

classical approach

3-6K

reduced classical approach

Our approach

50K

classical approach

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Metabolite profiles

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Experimental data

1,305 Holstein cows (1. lactation):

- **genotypes**
  - Illumina 50k SNP-chip
  - in total: 40,317 SNPs

- **milk metabolite profiles – hydrophilic phase of milk**
  - amino acids
  - sugar
  - carbon acids
  - in total: 190 metabolites (~ 2,000 milk metabolites are expected in the literature)

- **milk traits**
  - fat content
  - protein content
  - pH value

Statistical Model

$$y_{ijkl} = ah_i * stp_j + gld_k + b_1 * ltp + b_2 * ltp^2 + se_l + \varepsilon_{ijkl}$$

with

farm (ah)	i = 1,...,18,
test day (stp)	j = 1,...,39,
GC-MS batch (gld)	k = 1,...,47,
day of lactation (ltp)	ltp ϵ {21,...,120},
sire effect (se)	l = 1,...,214

Workflow - Methods

Metabolome

metabolite concentrations

	1	2	3	4	5	6	7	8
cow 1	0.1	0.2	0.1	0.02	1.2	5.2	0.7	0.03
cow 2	0.1	0.4	0.2	0.01	1.0	4.8	0.5	0.12
cow 3	0.1	0.4	0.1	0.06	1.4	5.3	0.6	0.06
cow 4	0.1	0.3	0.4	0.01	1.1	4.9	0.4	0.09

step 1.

Milk trait

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Determining important SNPs for an investigated milk trait.

- random forest (Breiman 2001)
- partial least squares (Wold 1966)

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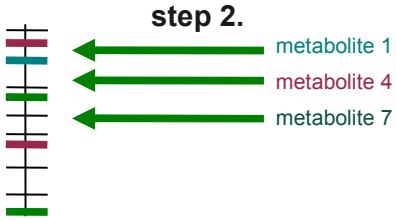
Illumina SNP chip

metabolite approach



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metabolite approach

step 2.

metabolite 1
metabolite 4
metabolite 7

step 1.

reduced classical approach

step 2.

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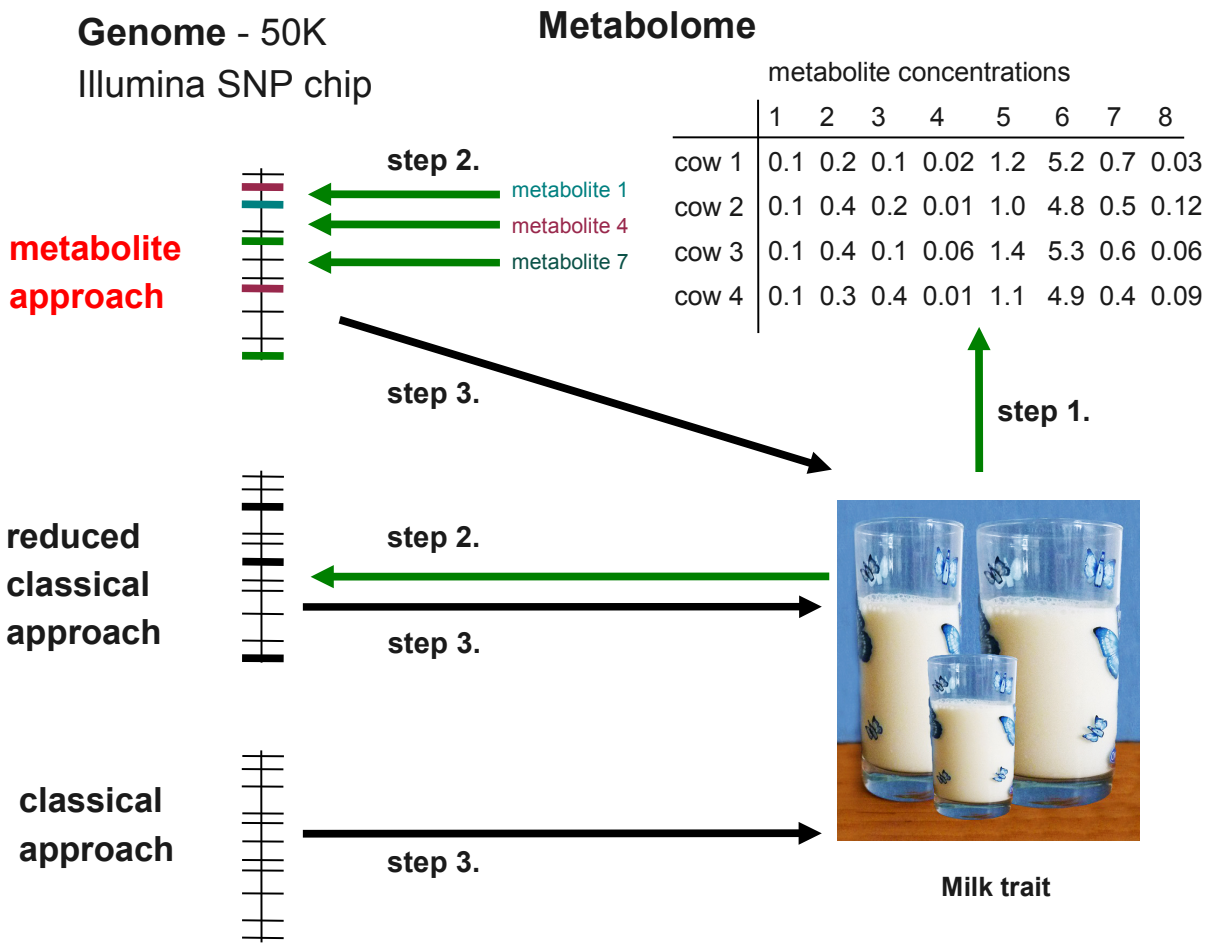

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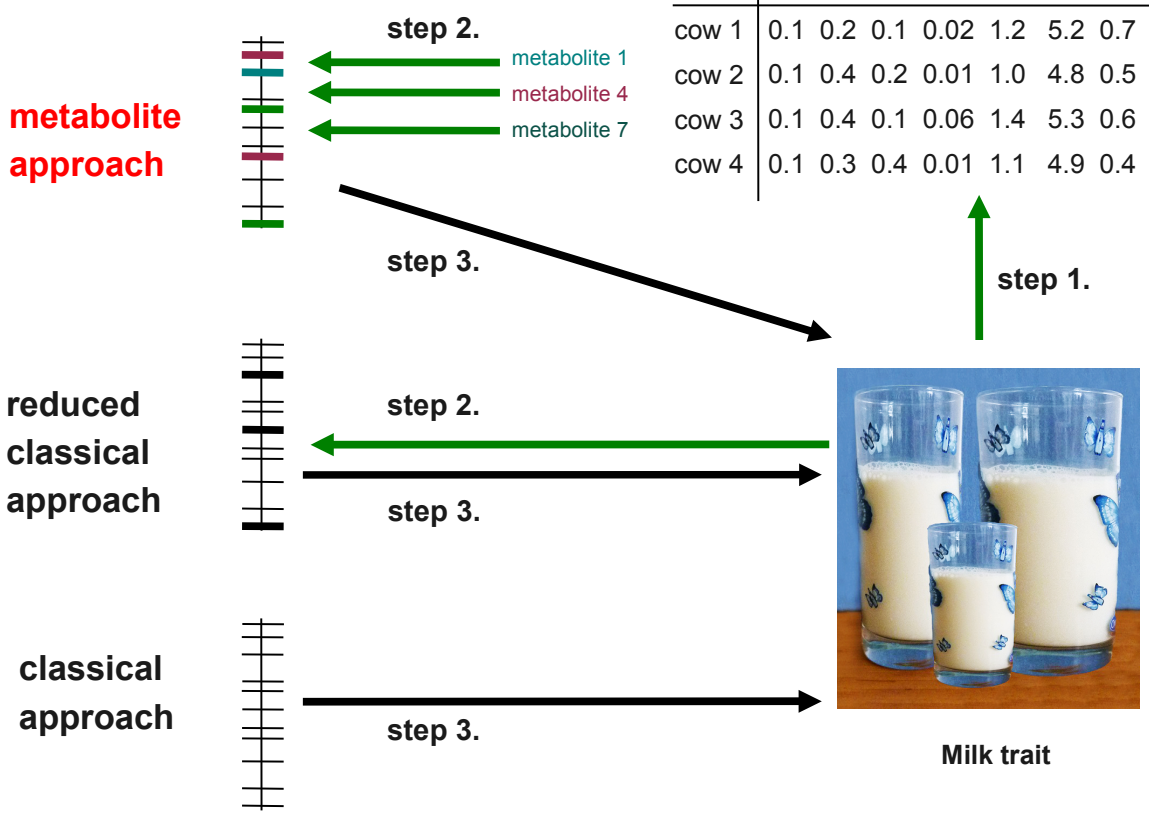

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10-fold cross-validation

Results – Step 1 (metabolite ← milk trait)

Average prediction precision (correlation between estimated and observed milk trait values):

	Random forest	Partial least squares
Protein	0.63	0.64
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Average number of detected important metabolites (examples):

Protein	16	(Spermidine, Asparagine, Aspartic acid)
Fat	11	(1,3- Dihydroxyacetone, Arabitol)
pH value	10	(Glycine)

More information can be found in Melzer et al. 2012 (J. Dairy Sci.)

Results – Step 2 (important SNPs ← trait)

Average number of detected SNPs:

Approach	Fat (%)	Protein (%)	pH value
Reduced classical approach	26	83	82
Metabolite approach	129	302	114
Single important metabolites	[4;42]	[5;55]	[4;54]

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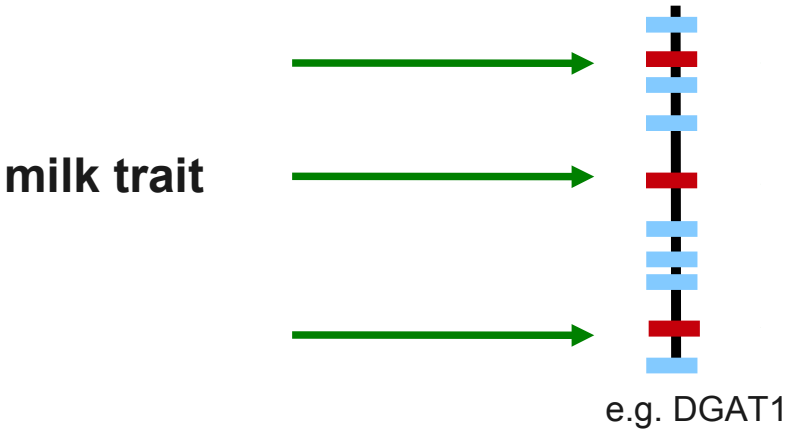

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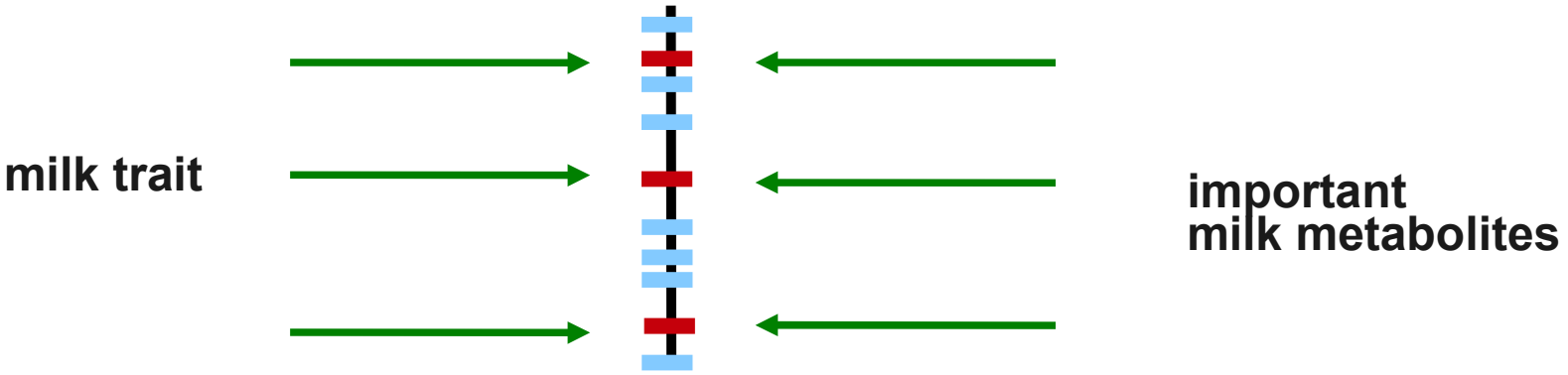
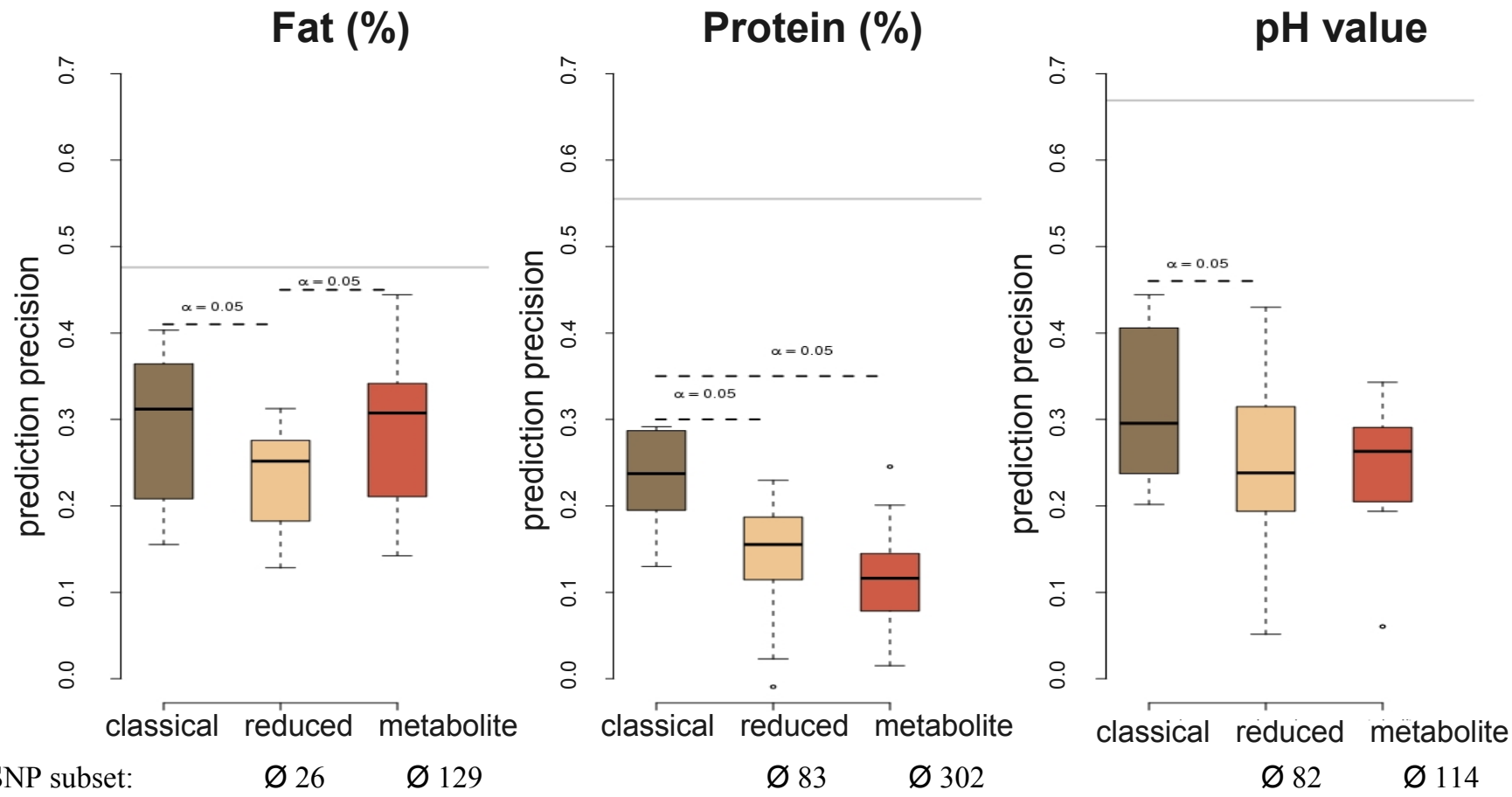


Results – Step 3 (SNPs ⟶ milk trait)



SNP subset: Ø 26 Ø 129 Ø 83 Ø 302 Ø 82 Ø 114

Summary

Step 1:

- both regression methods revealed similar important metabolites
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Step 2:
- mostly fewer important SNPs were detected for single important metabolites
- important SNPs with high genetic effects for milk traits also showed an impact on at least one of the important metabolites

→ considering the heritability of metabolites (Poster Session 12 No. 23: "Milk metabolites and their genetic variability", Wittenburg et al., 2012)

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Step 3:
- observed prediction precisions were mostly higher for the metabolite approach compared to the reduced classical approach

Conclusion

- considering an intermediate level in the classical genotype-phenotype map enables various investigation opportunities

- for two investigated milk traits, fat and pH value, the genotype-phenotype map is mirrored on the metabolome level

Our metabolite approach seems to be promising and in addition provides functional information.

Thank you for you attention!

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- PD Dr. C. Kühn (FBN, WG QTL Regions)
- A. Rief, R. Grahl (FBN)

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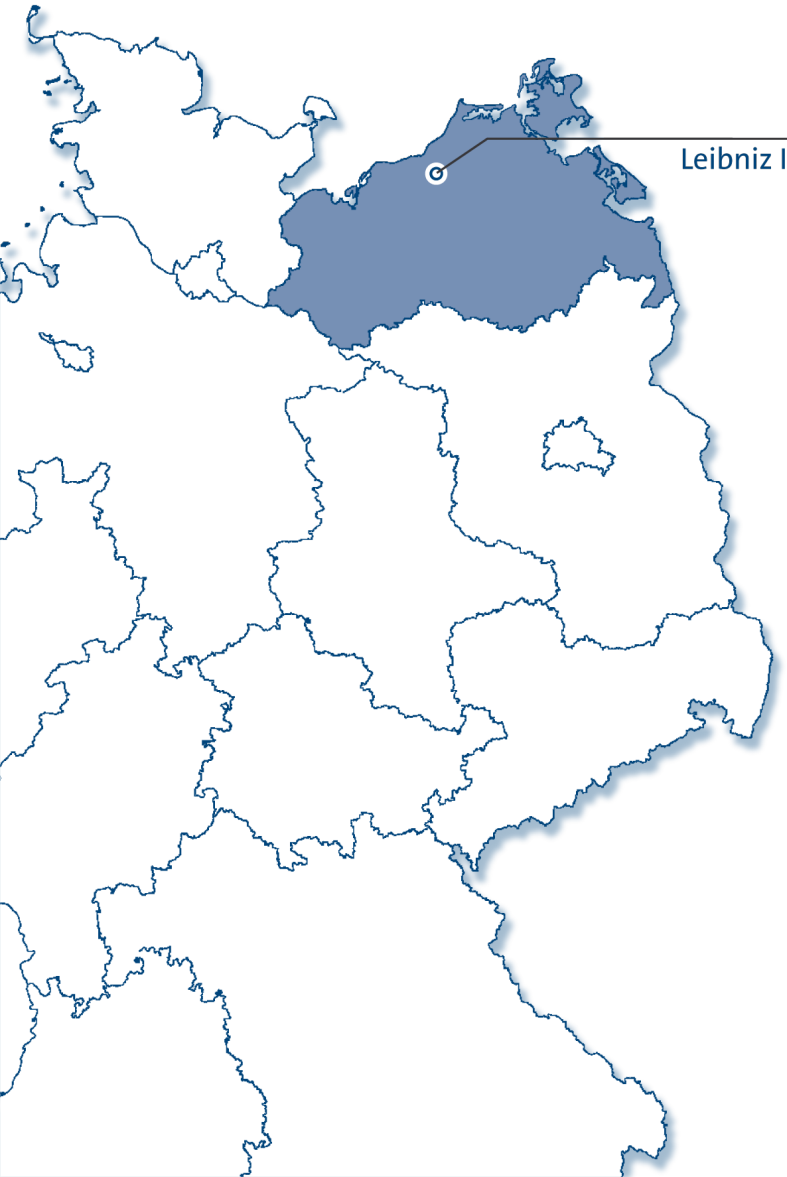

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