



UNIVERSITÀ
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DAFNAE
Department of Agronomy Food
Natural resources Animals Environment

World Congress on Genetics Applied to Livestock Production
Aotea Centre Auckland, New Zealand,
11–16 February 2018

Exploiting the network-based association weight matrix approach for the genetic dissection of milk nitrogen fractions in dairy cattle

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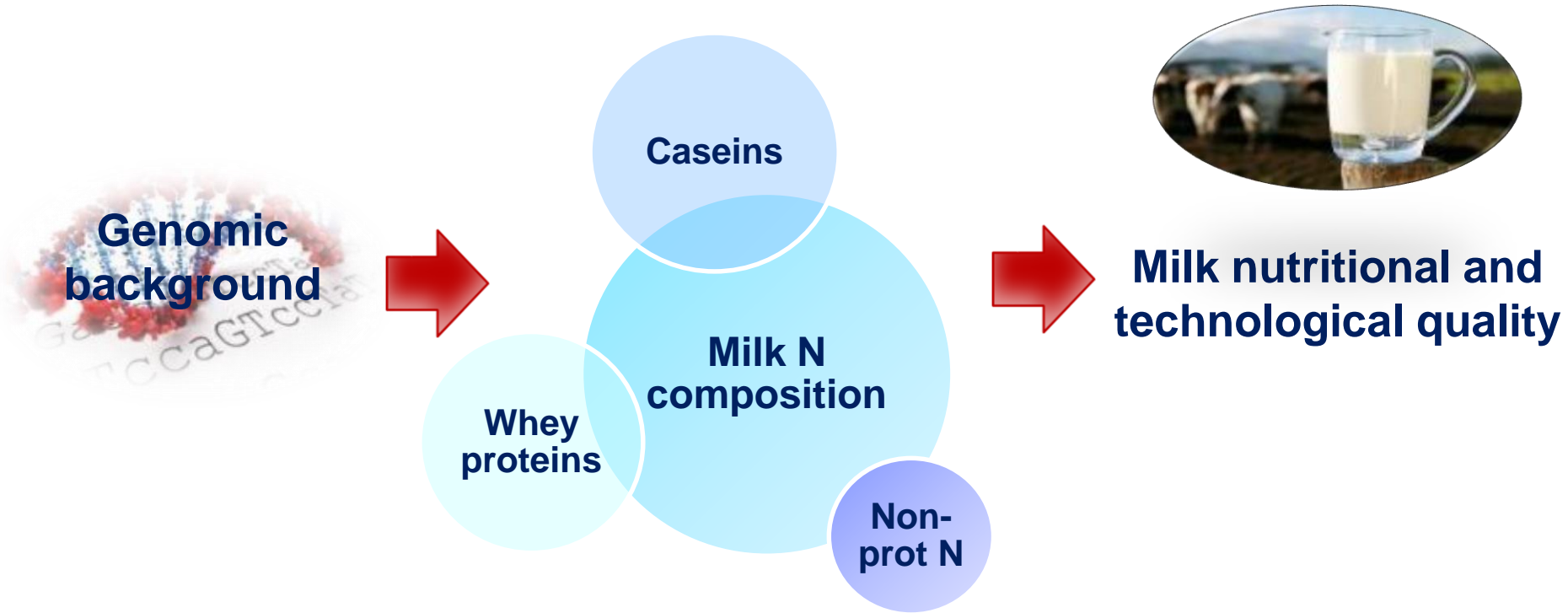
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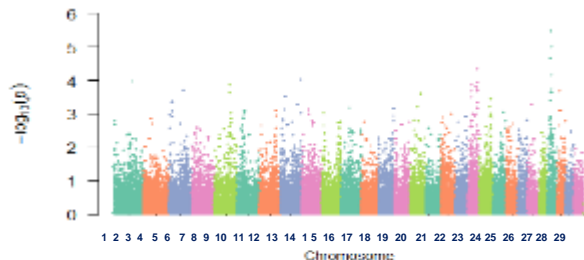


Background



Aim of the study

GWAS results (P-val & additive effect)



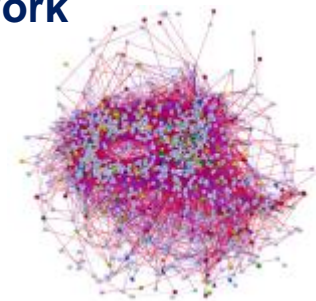
Gene-set enrichment
analysis



Associated weight matrix (AWM)
and network framework



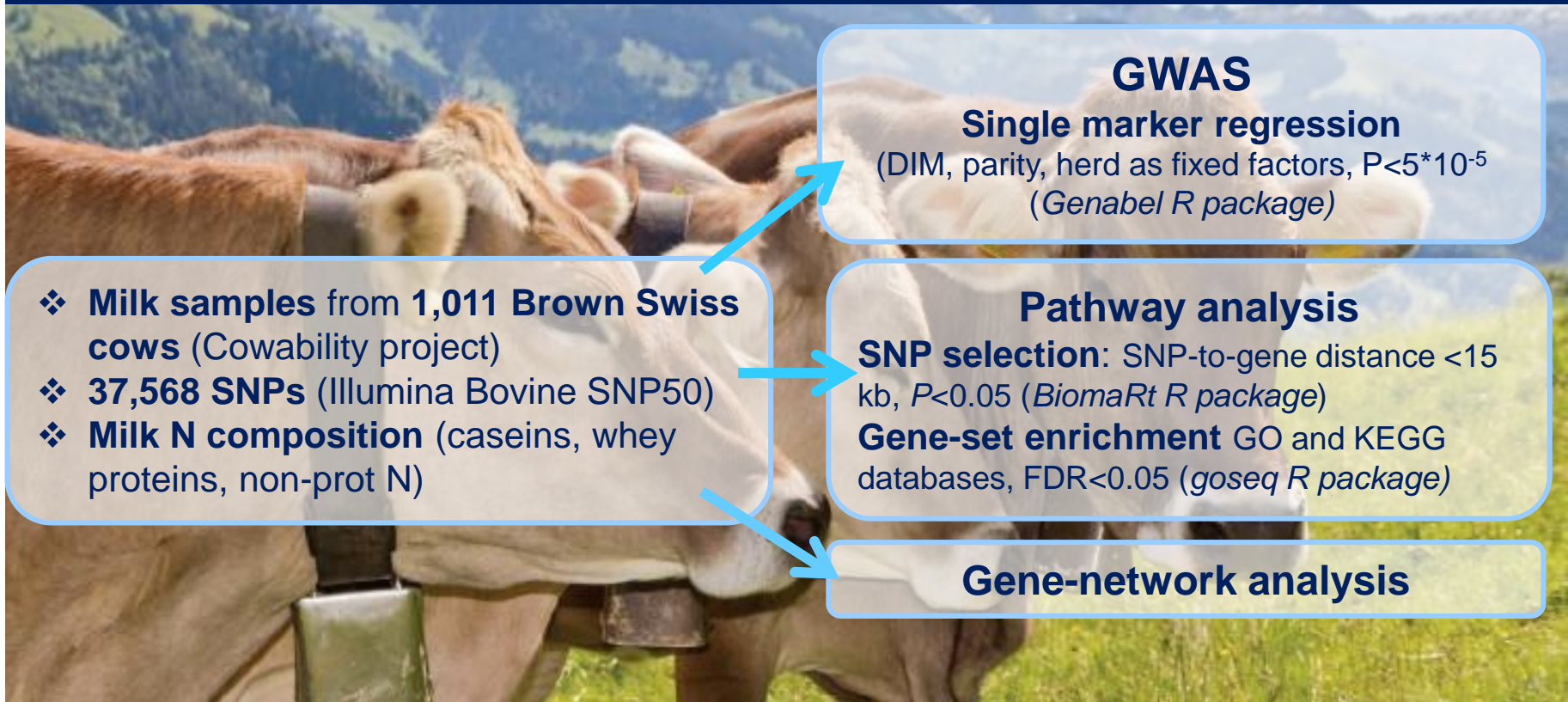
	κ -CN	β -LG	MUN
<i>SUSD1</i>	-0.925	-2.066	...
<i>SSPN</i>	1.177	-1.108	...
<i>TERT</i>	0.118	0.159	...
...



Deeper knowledge about genetic regulation of the physiological and cellular mechanisms required for milk protein synthesis and secretion



Methods



GWAS

Single marker regression
(DIM, parity, herd as fixed factors, $P < 5 \cdot 10^{-5}$)
(*Genabel R package*)

- ❖ **Milk samples** from **1,011 Brown Swiss cows** (Cowability project)
- ❖ **37,568 SNPs** (Illumina Bovine SNP50)
- ❖ **Milk N composition** (caseins, whey proteins, non-prot N)

Pathway analysis

SNP selection: SNP-to-gene distance < 15 kb, $P < 0.05$ (*BiomaRt R package*)
Gene-set enrichment GO and KEGG databases, $FDR < 0.05$ (*goseq R package*)

Gene-network analysis

Methods - AWM construction

SNP selection criteria from GWAS:

- ✓ Selection of key phenotype (κ -CN)
- ✓ Primary SNP selection: $P \leq 0.05$ for κ -CN
- ✓ Secondary SNP selection: SNPs with $P \leq 0.05$ in ≥ 3 non- key phenotypes
- ✓ SNP-to-gene distance: < 10 kb
- ✓ One SNP-One Gene: 1) $> n^\circ$ phenotypes; 2) lowest P -val



Pearson's r

	κ -CN	β -LG	...MUN
$SUSD1$	-0.925	-2.066	...
$SSPN$	1.177	-1.108	...
$TERT$	0.118	0.159	...
...

❖ Rows: 1,917 SNP/genes
❖ Columns: milk N composition
❖ Cell: normalized (z-scores) additive effect

PCIT algorithm

(Partial correlations - Information Theory)

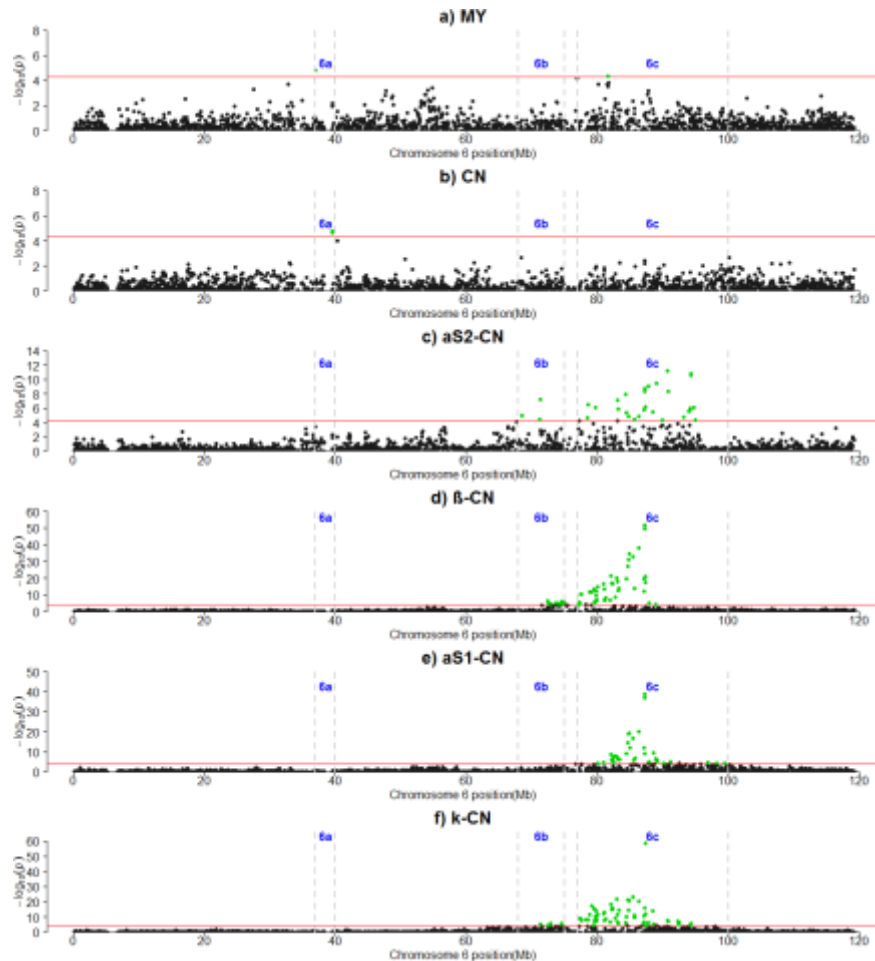
$r \geq |0.80|$

Biologically relevant interactions and key regulators

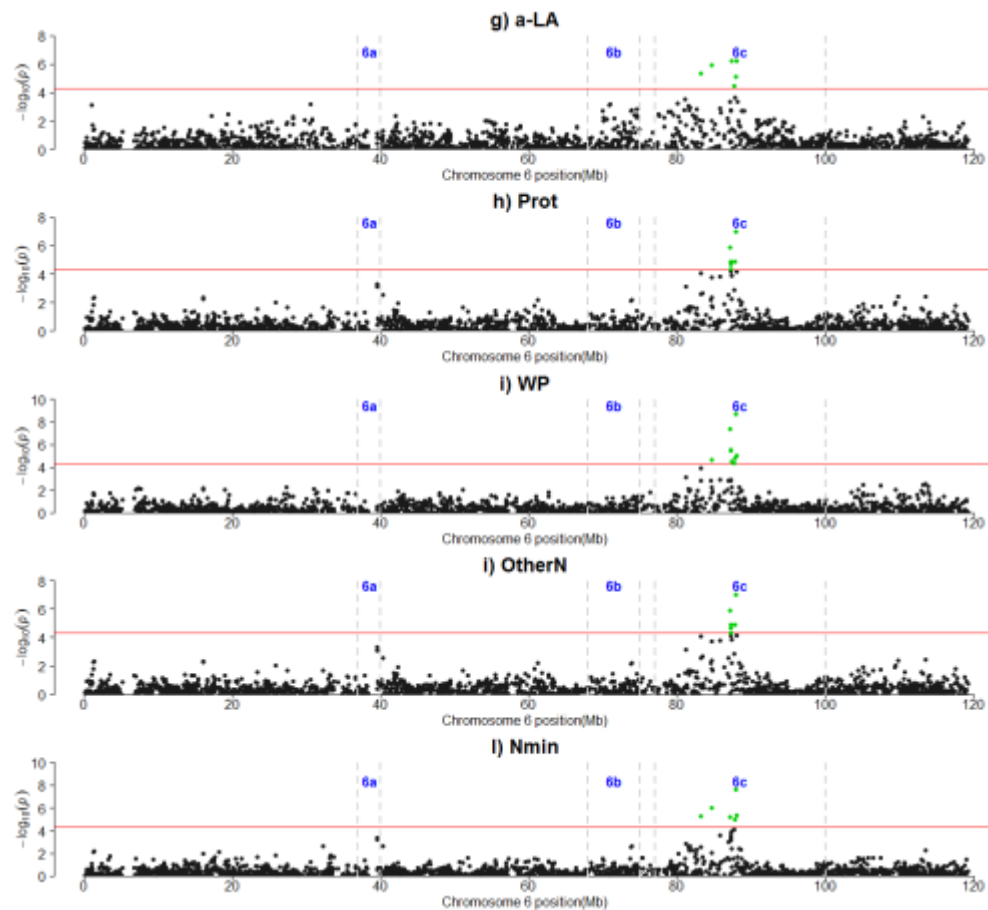
(Cluego, NetworkAnalyzer, IPA)



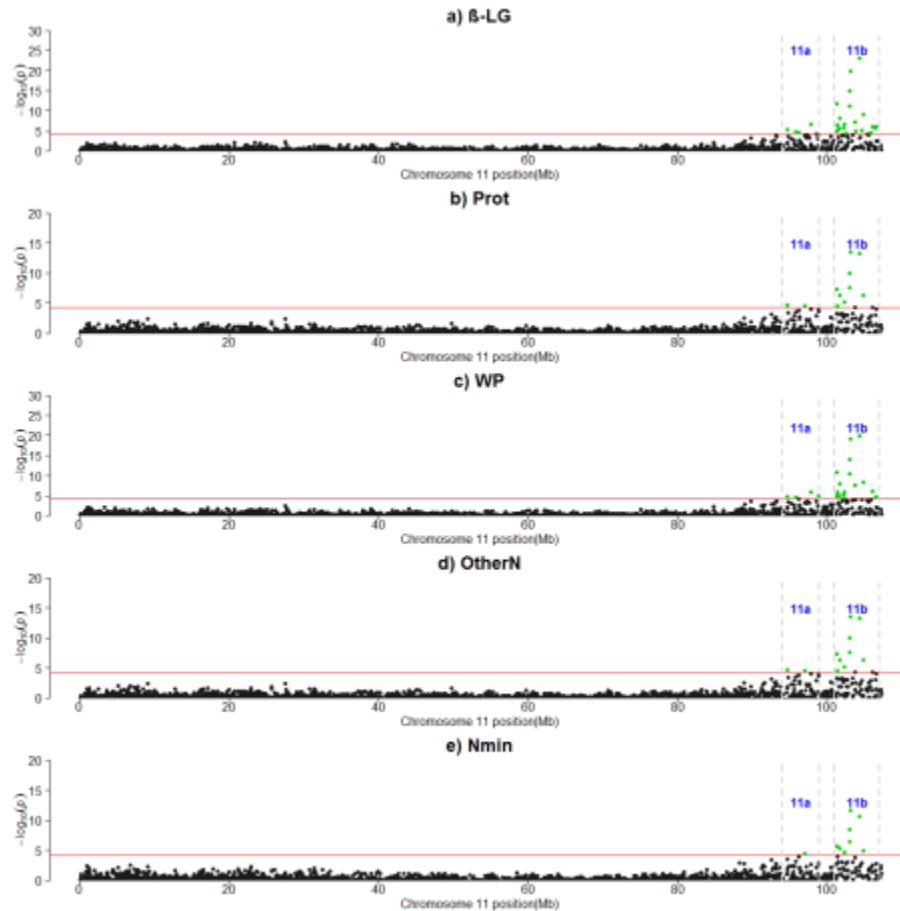
BTA6



BTA6



BTA11



Results – GWAS

GWAS  **170 SNPs:** 103 SNPs on **BTA6** (~77.19-99.45 Mbp, including casein cluster);
22 SNPs on **BTA11** (~101.27 - 106.54 Mbp, including the *PAEP* gene)

Heritability estimations are in general high
Heritability for casein fractions are higher than total casein

Results – GWAS and pathway analysis

37000 SNPs

17000 SNPs btw 15Kb (flanking)

13000 genes

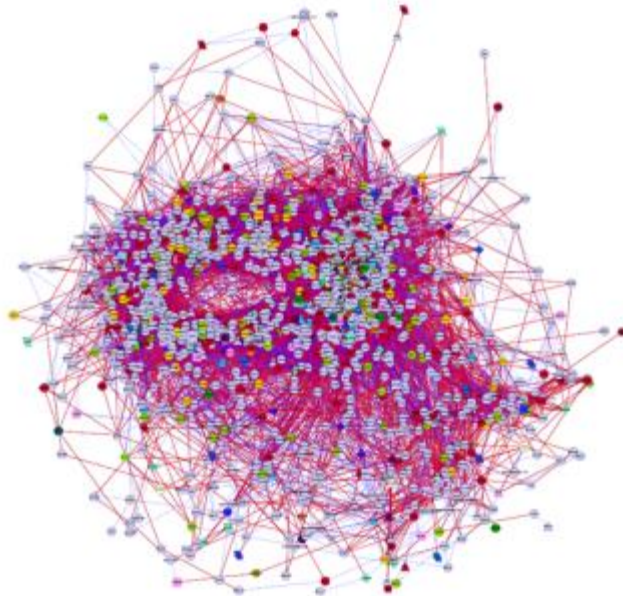
600 genes with significant associations with MY or PROT

**PATHWAY
ANALYSIS**

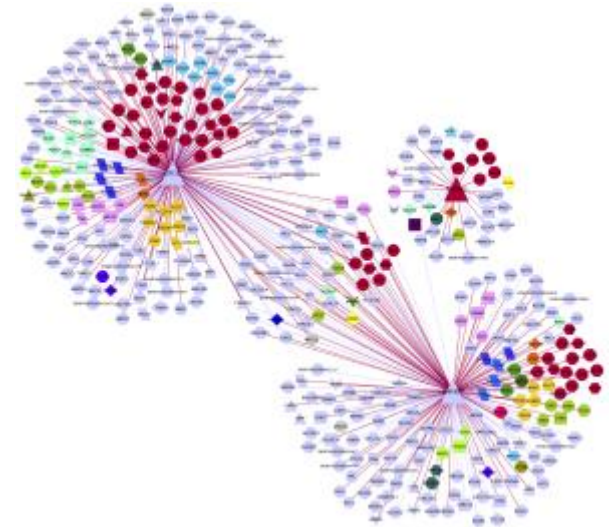


Results – Network analyses

1,904 nodes/genes
101,284 edges/interactions

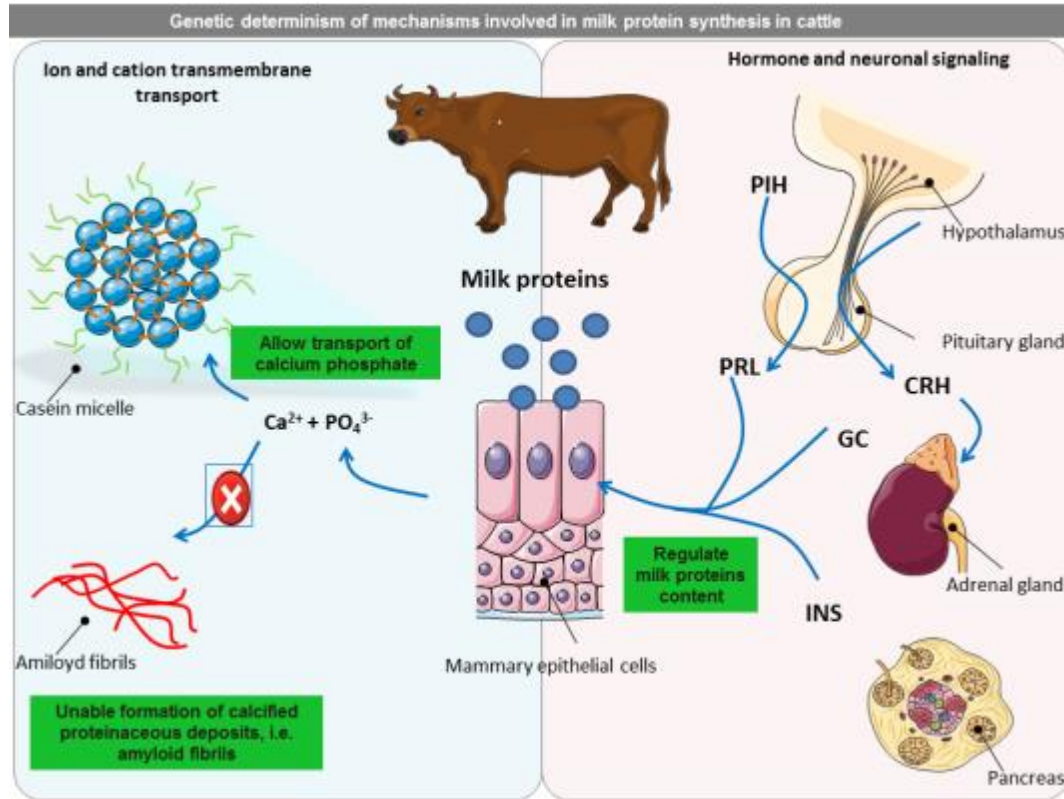


TF-trio
GFI1B, *NR5A1* and *ZNF407*

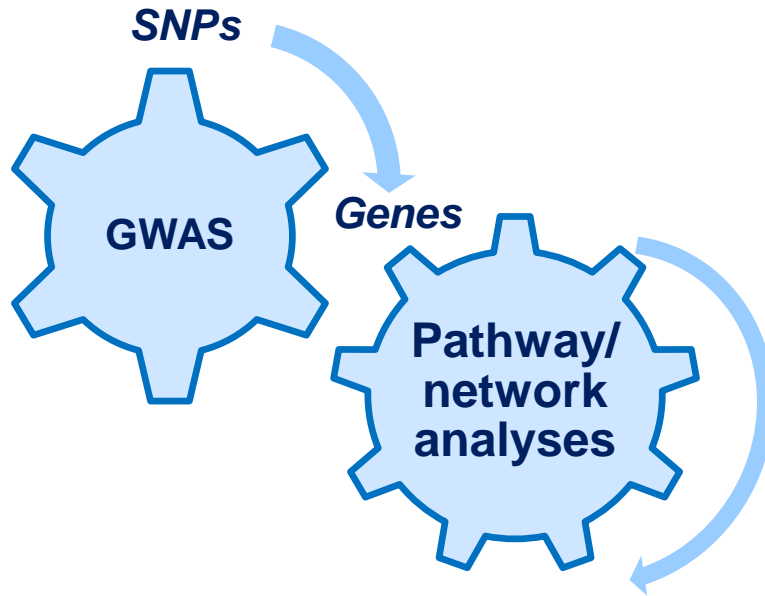


452 genes (24% of genes in
the filtered AWM)

Results - Functional analyses



Conclusions



- ✓ Genetic control of milk protein composition
- ✓ Mammary gland functionality
- ✓ *GFI1B*, *NR5A1* and *ZNF407* as key regulators

↓
Validation studies? **FUTURE**

Selection strategies to improve milk quality and technological characteristics

