



DEPARTMENT OF
**ANIMAL
BIOSCIENCES**



Identifying genomic regions affecting milk BHB in North America Holstein dairy cattle

Shadi Nayeri¹, V. Kroezen¹, M. Sargolzaei¹², A. Fleming¹, F. Schenkel¹, C. Baes¹,
A. Cánovas¹, J. Squires¹, and F. Miglior¹³

¹CGIL - University of Guelph; ²The Semex Alliance, Guelph, Ontario, Canada; ³Canadian Dairy Network, Guelph, Ontario, Canada

Metabolic diseases

- Increase in milk production accompanied by higher incidence of metabolic diseases
 - Ketosis, fatty liver, displaced abomasum
- Reasons:
 - Metabolic changes and challenges early in lactation
 - Mobilizing more fat reserves, production of non-esterified fatty acid (**NEFA**) and **ketone bodies** (including **Beta-hydroxybutyrate, BHB**)
- Failure to maintain energy balance
 - Detrimental effects on immune function, milk production and overall health
 - **Increased incidence of displaced abomasum, ketosis, milk fever, metritis, cystic ovaries and lameness**

Targeting a new trait for selection

- Target health traits that **impact profitability** for farmers and dairy industry

Metabolic Disease Resistance (MDR)

Official evaluation implemented by Canadian Dairy Network in 2016

MDR index = 50% **BHB**, 25% **Ketosis**, 25% **DA**

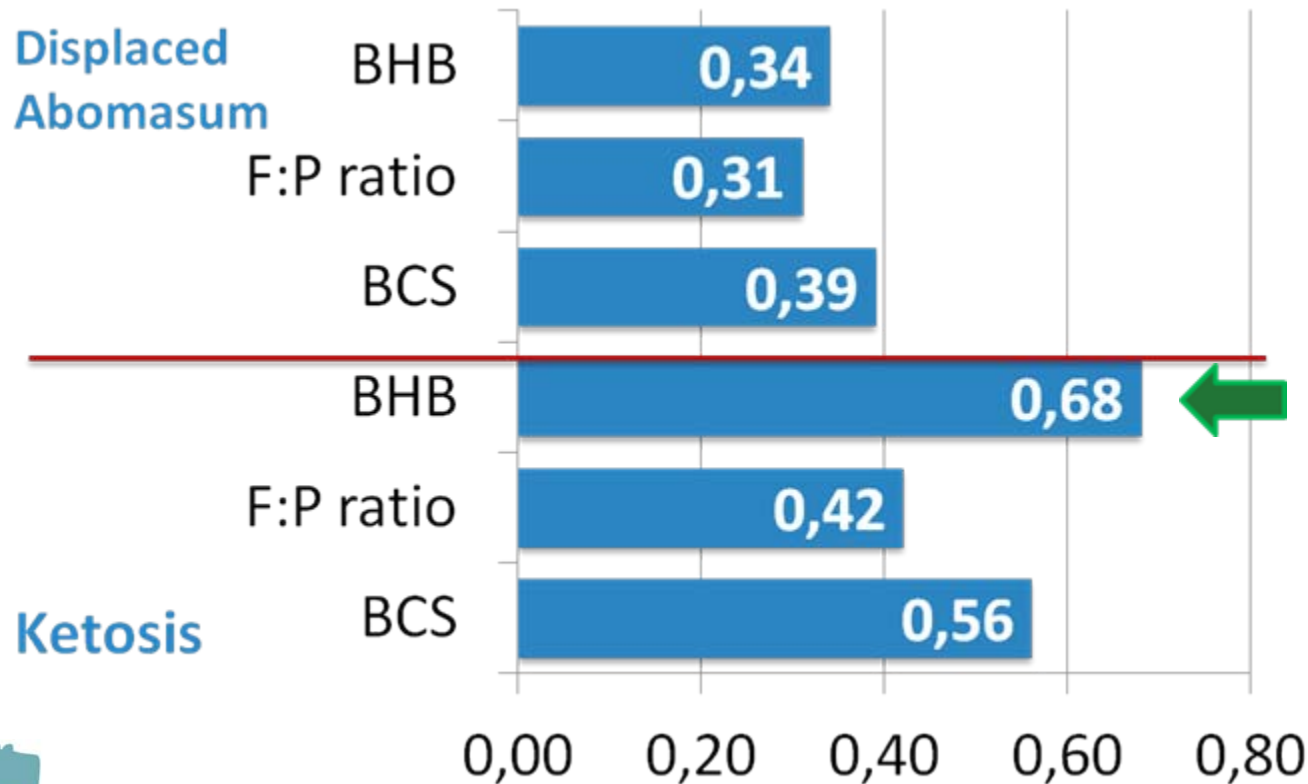
Subclinical ketosis
(BHB data from DHI)

Clinical ketosis and
displaced abomasum (DA)
(Producer recorded data)

Importance of subclinical ketosis

- Subclinical signs of ketosis are difficult to detect by farmers
- Subclinical ketosis more common than clinical ketosis
 - 10-fold higher frequency at the herd level
 - Median incidence of 11.2%-36.6%
- There is a high correlation between the amount of BHB in milk and ketosis

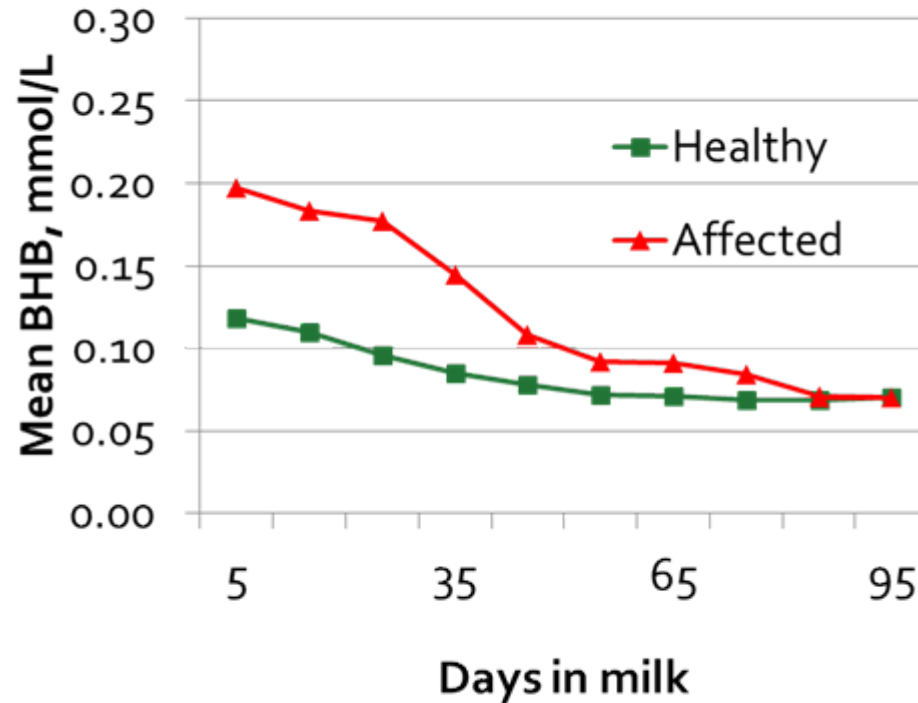
Correlation estimates – First parity



(Jamrozik et al., 2016)

Phenotypic associations – Diseases and BHB

Ketosis and BHB



(Koeck et al., 2016)



Objective

Identify genomic regions and candidate genes that may affect BHB concentrations in milk associated with subclinical ketosis



Genome-wide association analysis (GWAS)

Phenotypes	Genotypes
Canadian Dairy Network (CDN)	
<ul style="list-style-type: none">- Subclinical ketosis first lactation (BHB1)	n= 24,657 cows and bulls (Bovine 50K genotypes)
<ul style="list-style-type: none">- Subclinical ketosis 2nd and later lactations (BHB2+)	Imputed to HD FImpute V2.2.
<ul style="list-style-type: none">- De-regressed EBVs	n= 2,507 reference animals

(Garrick et al., 2009)

(Sargolzaei et al., 2014)

Functional analysis

- **Annotation**

R package BioMart software used for gene annotation

(Durinck et al., 2009)



- **Functional analysis**

Ingenuity Pathway Analysis (**IPA**)

(Kramer et al., 2014)

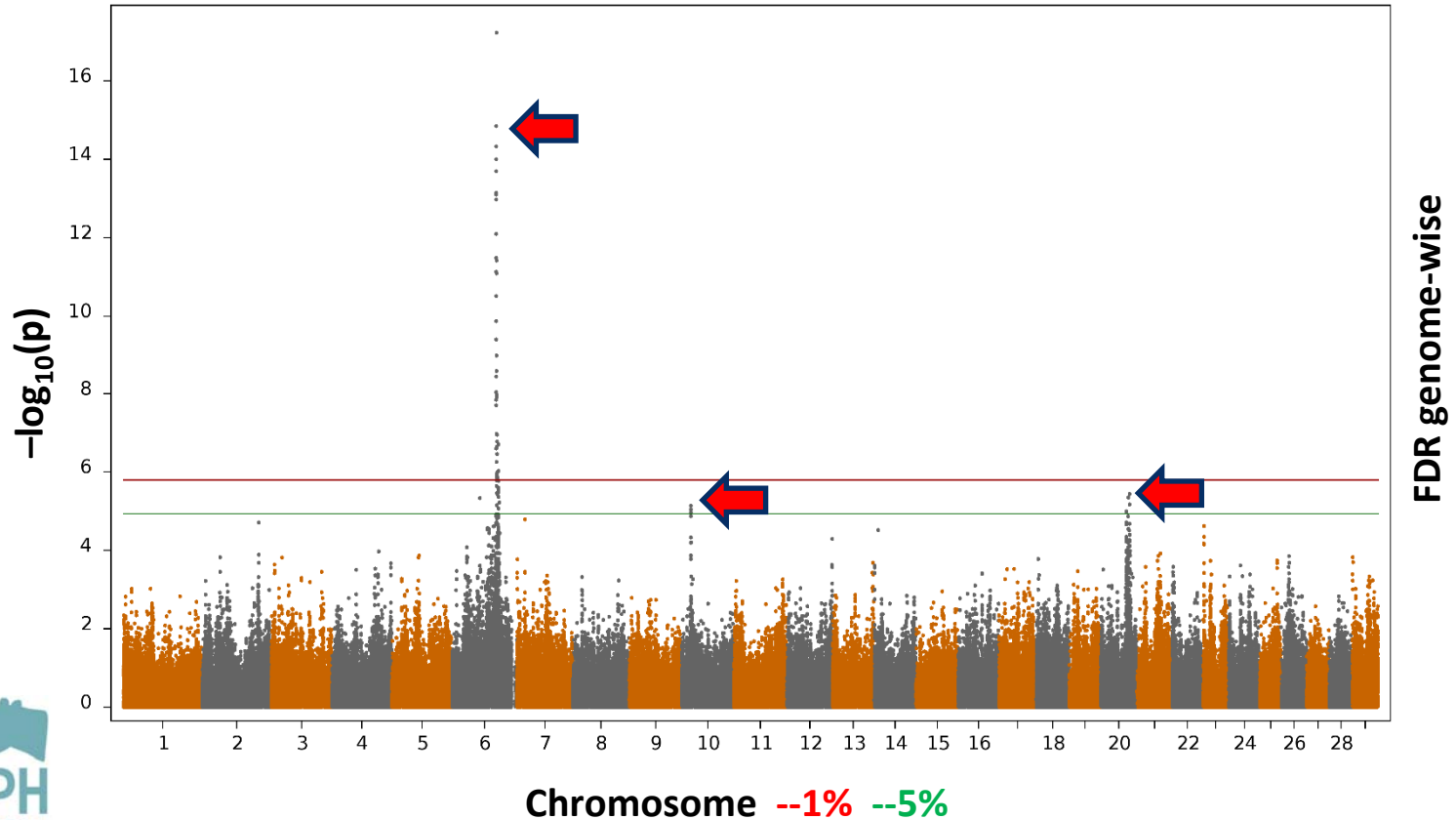
PANTHER classification system

(Thomas et al., 2003)

Results

BHB1 – GWAS result

Distribution of $-\log_{10}(p)$ for BHB1



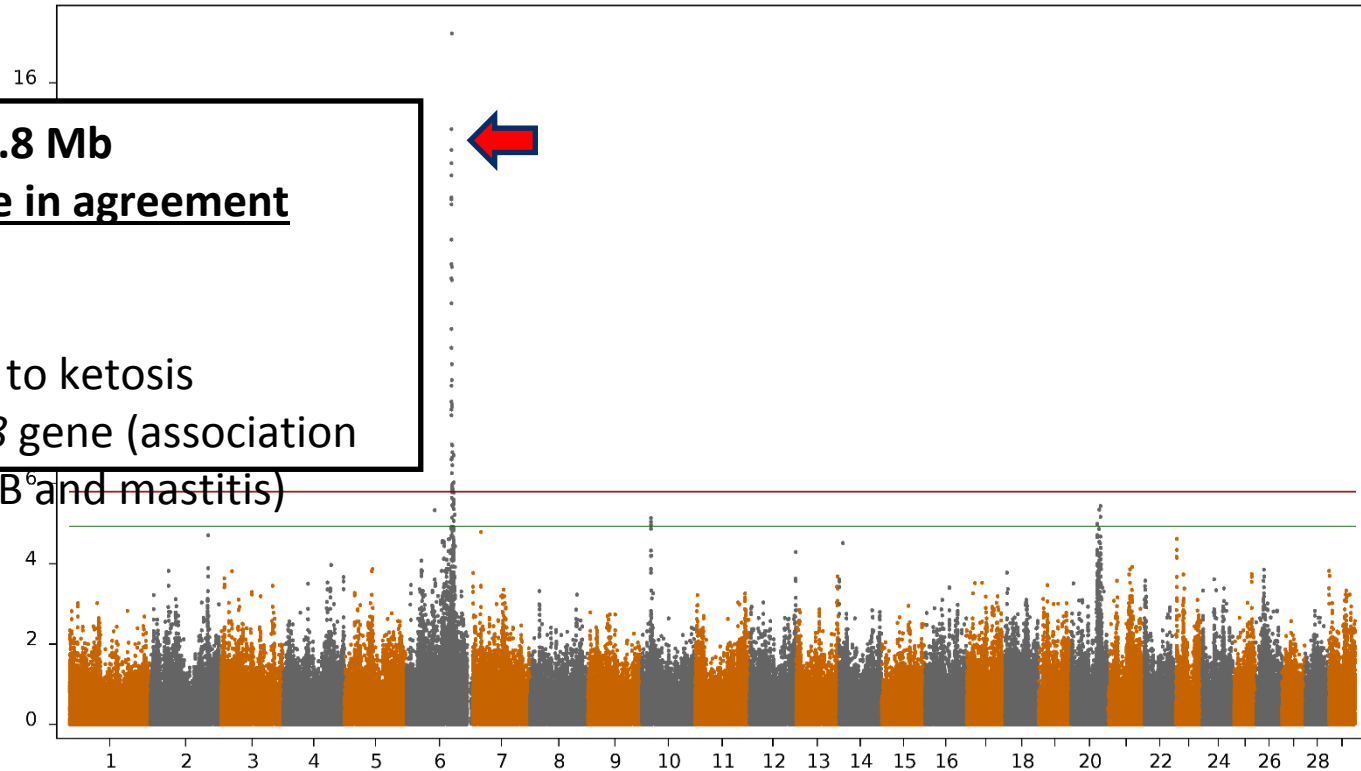
BHB1 – GWAS result

Distribution of $-\log_{10}(p)$ for BHB1

BTA6: 88.4-94.8 Mb

The results are in agreement with:

- Mastitis
- Susceptibility to ketosis
- *GC* and *CXCL8* gene (association with blood BHB^o and mastitis)



FDR genome-wise

Chromosome --1% --5%

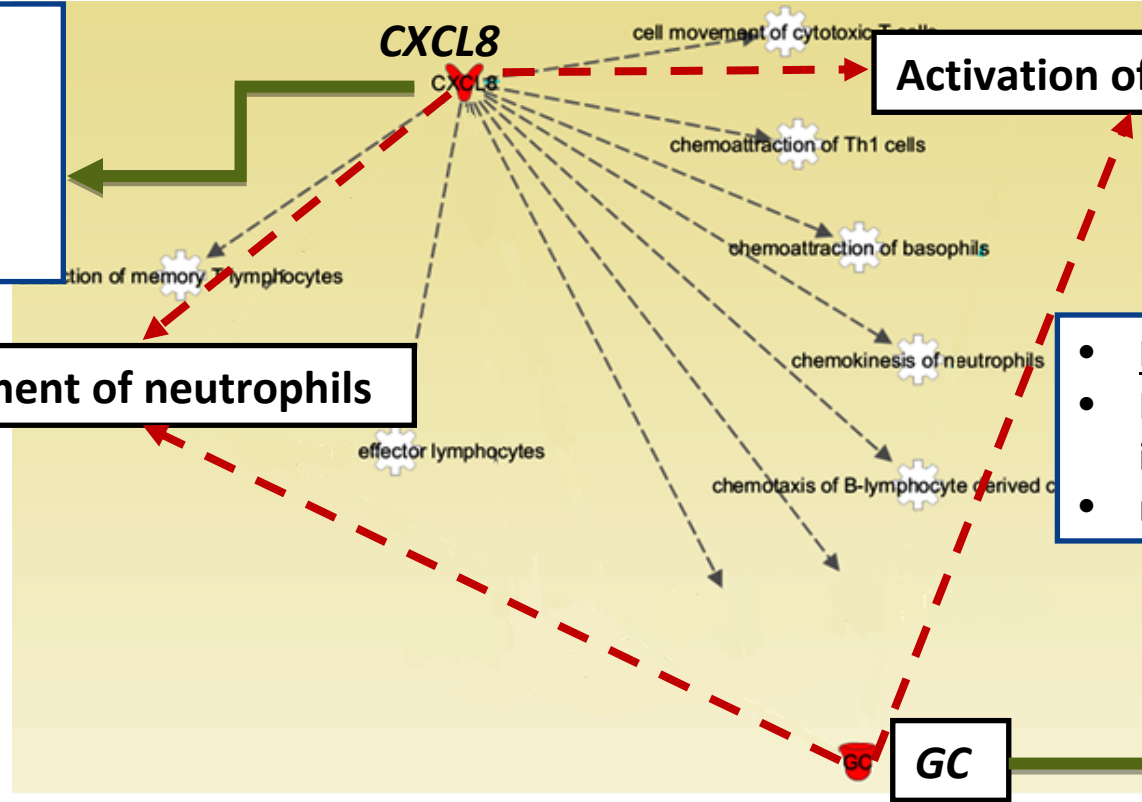
Functional analysis-Ingenuity Pathways

- Role in:
- Autophagy
- Immunity to extracellular pathogens

Recruitment of neutrophils

Activation of macrophages

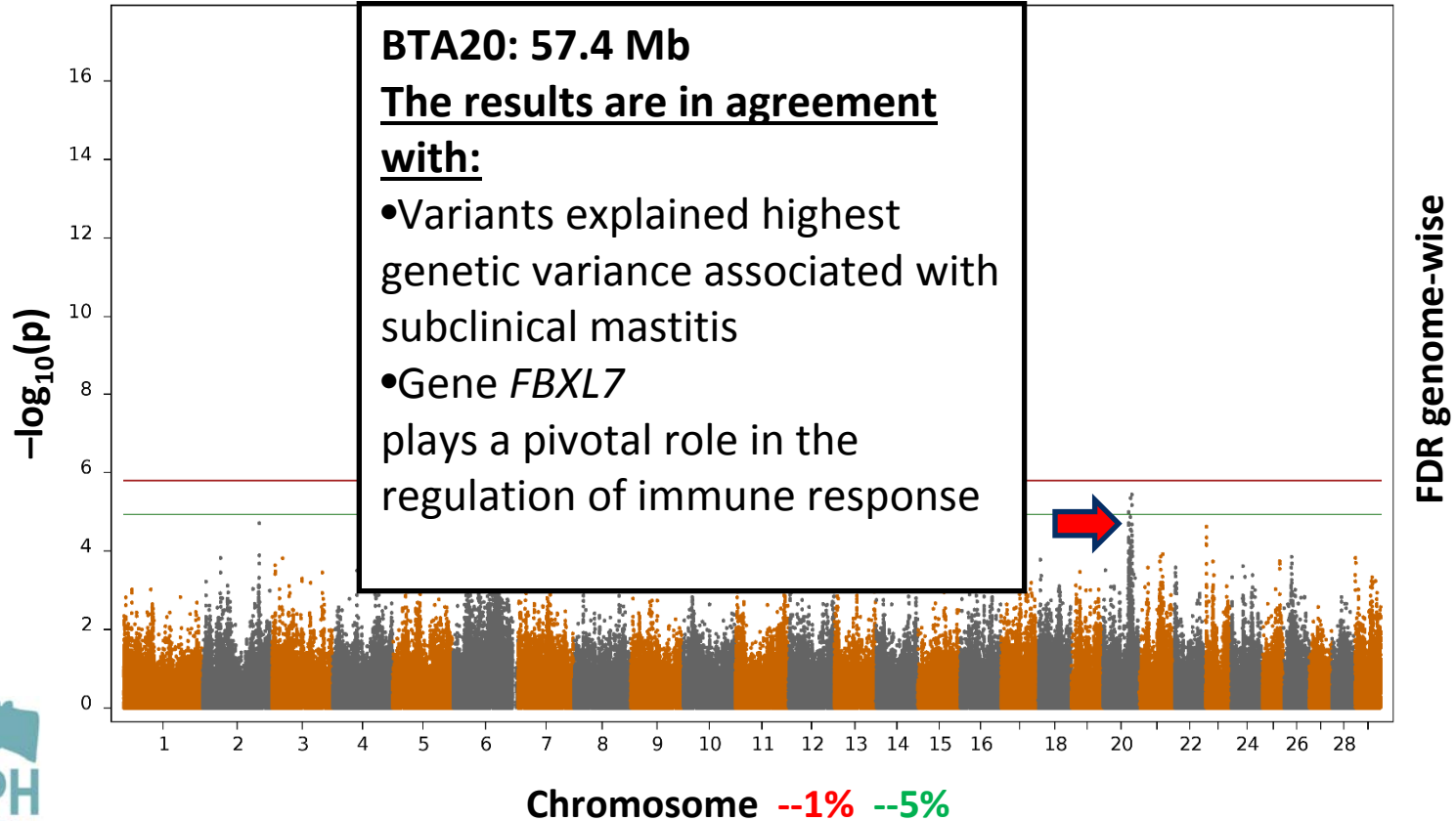
- Role in:
- Host defense and immune system
- milk production



(McLeod et al., 1989; Mount et al., 2009; Goddard et al., 2014; Ryter et al., 2014)

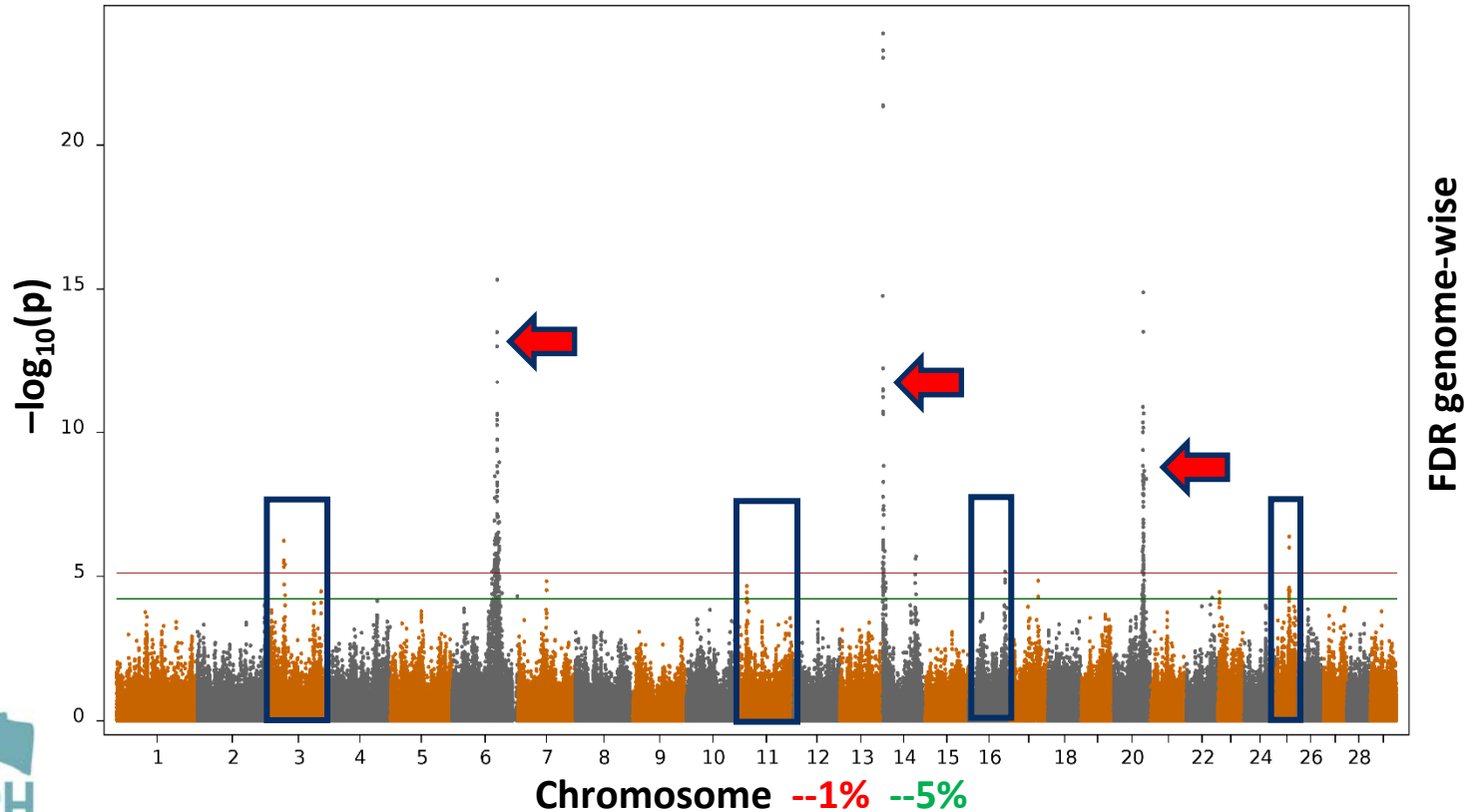
BHB1 – GWAS result

Distribution of $-\log_{10}(p)$ for BHB1



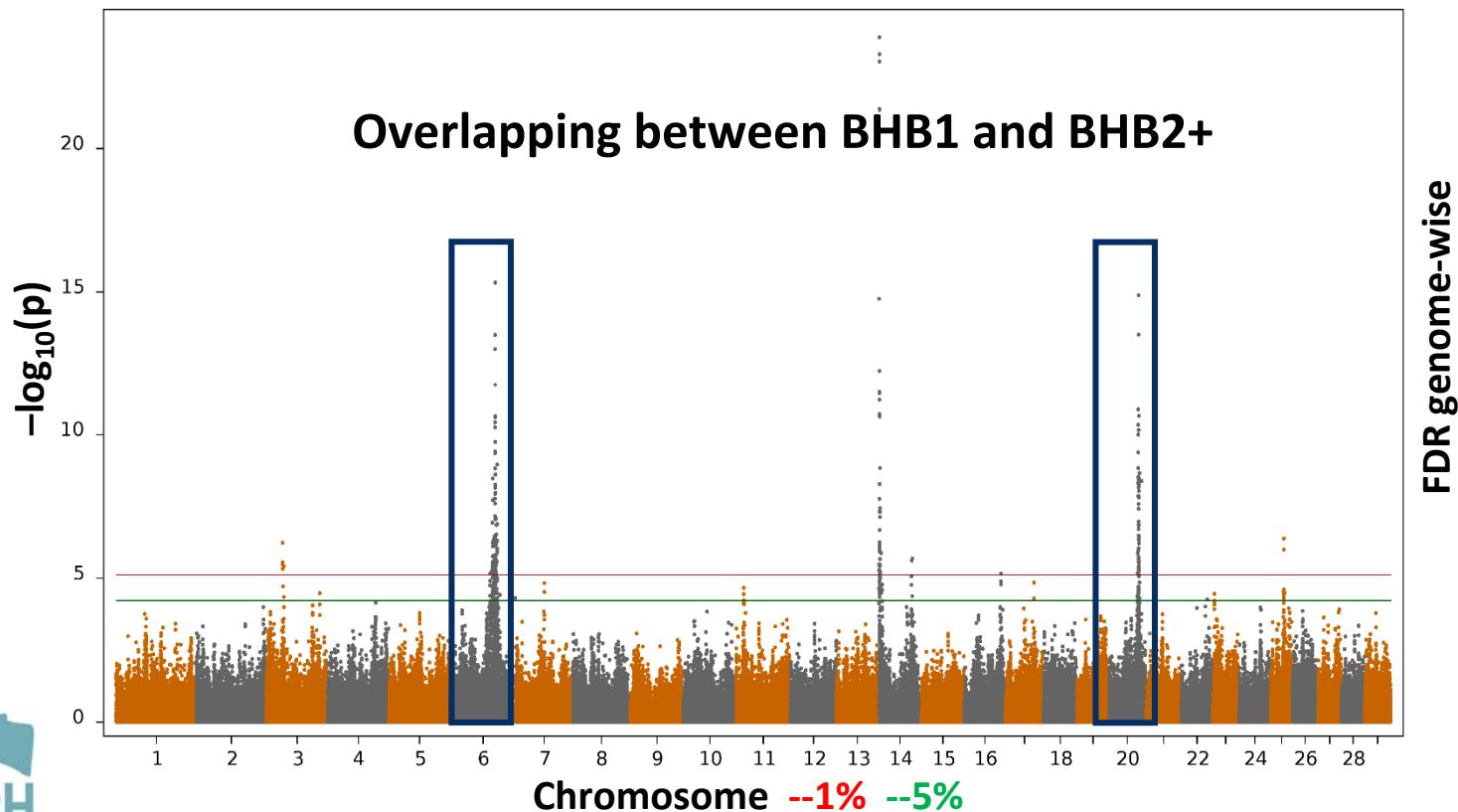
BHB2+ - GWAS result

Distribution of $-\log_{10}(p)$ for BHB2+



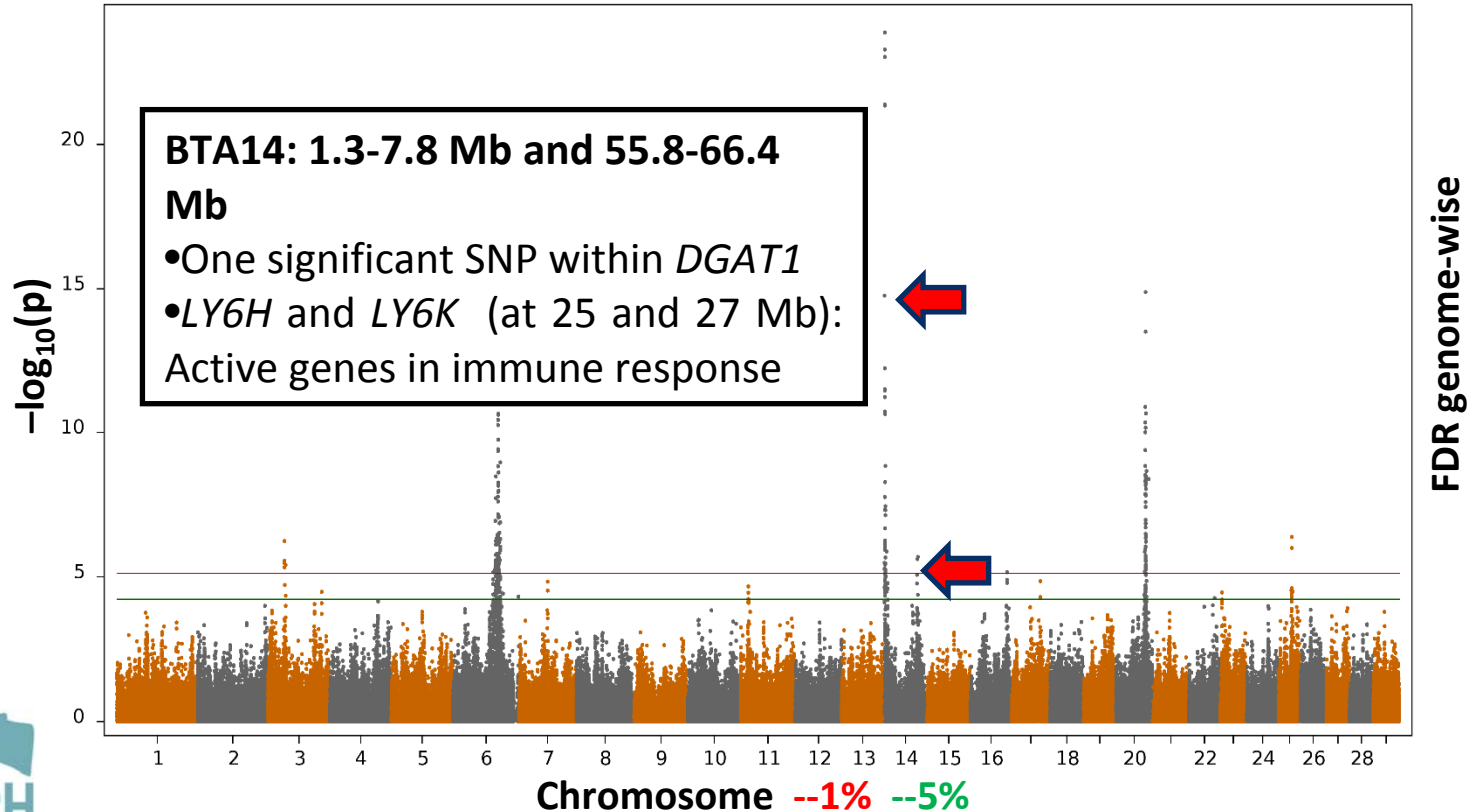
BHB2+ - GWAS result

Distribution of $-\log_{10}(p)$ for BHB2+



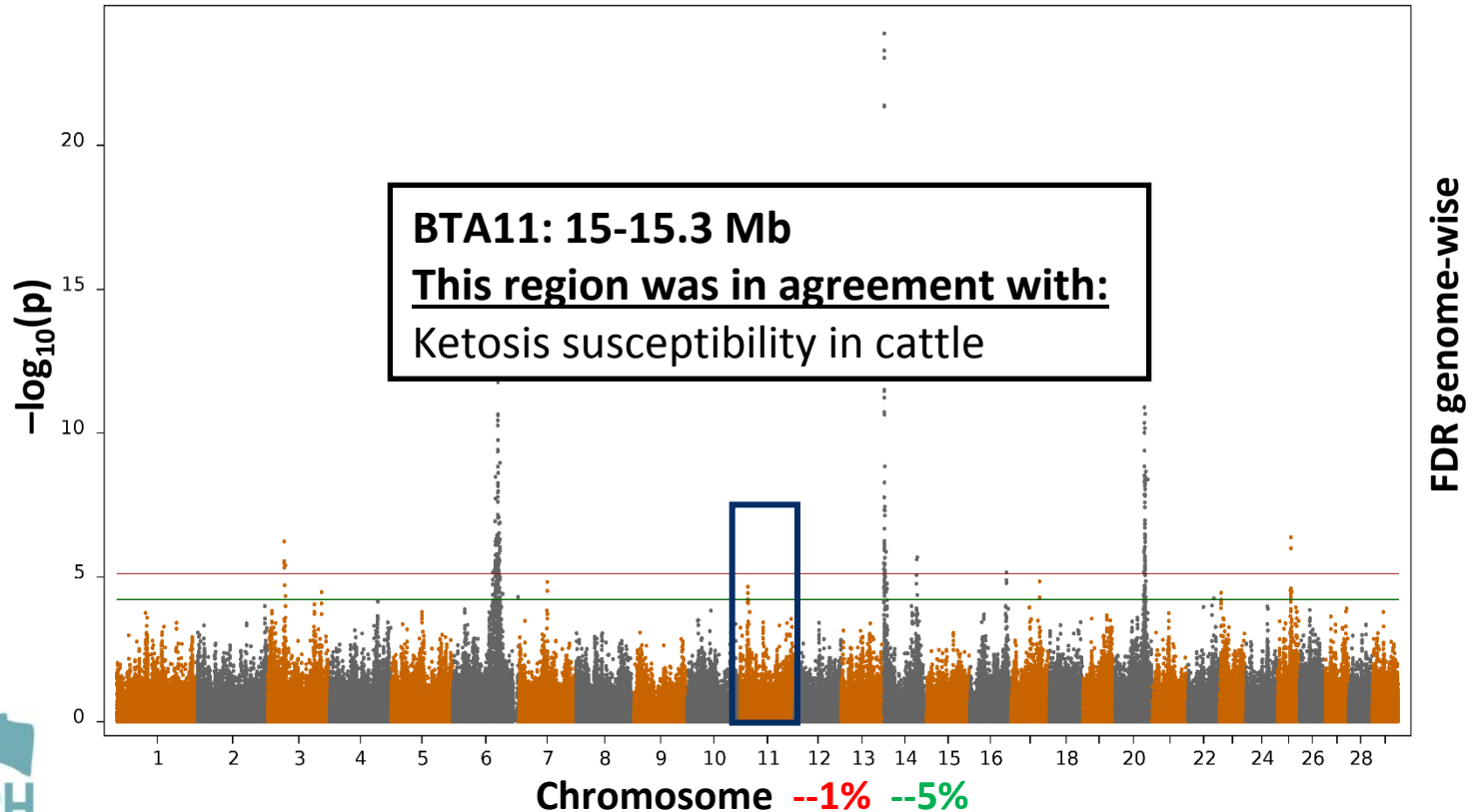
BHB2+ - GWAS result

Distribution of $-\log_{10}(p)$ for BHB2+



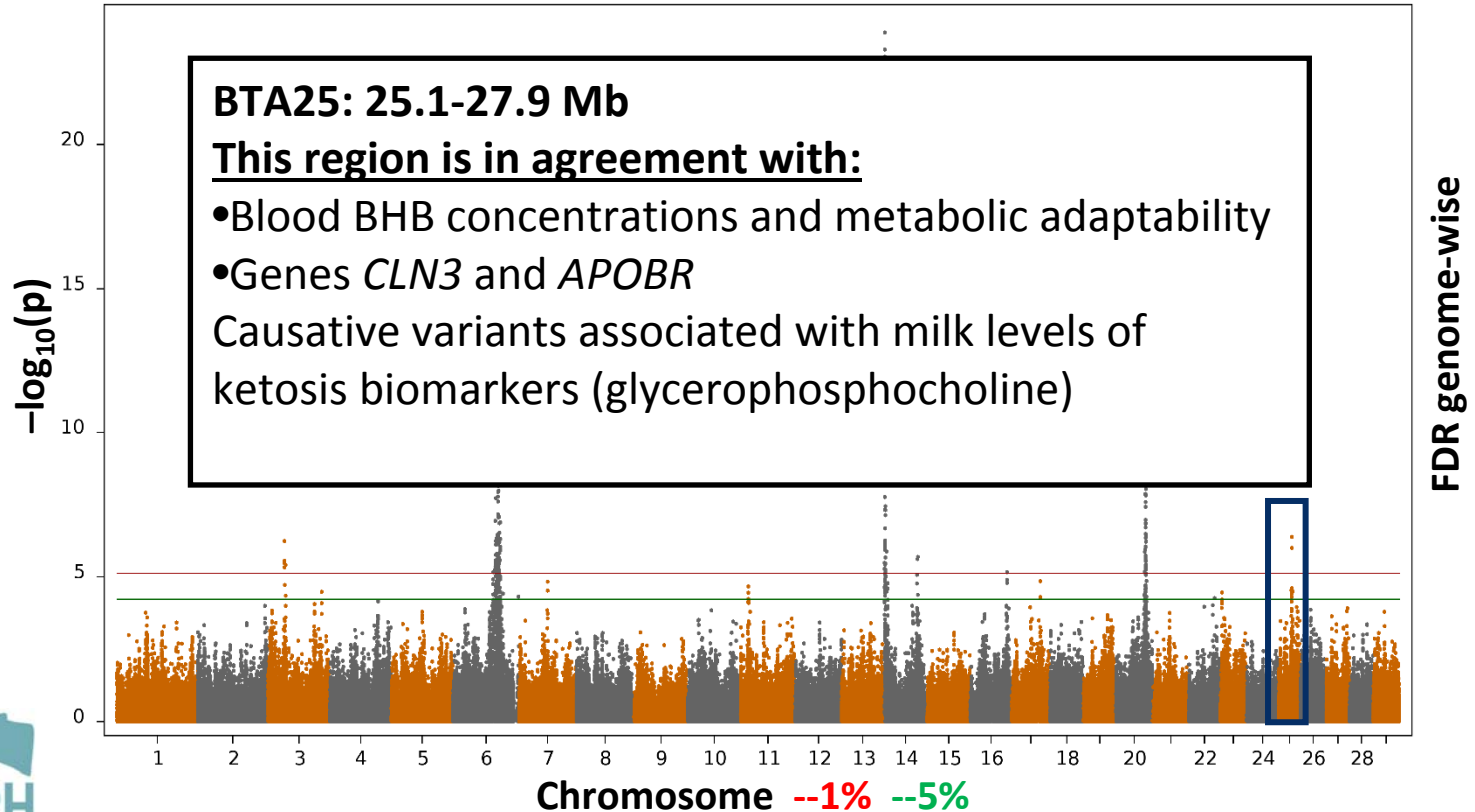
BHB2+ - GWAS result

Distribution of $-\log_{10}(p)$ for BHB2+



BHB2+ - GWAS result

Distribution of $-\log_{10}(p)$ for BHB2+



Conclusions

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- GWAS resulted in significant regions associated with milk BHB in first and later lactations
 - Mainly on chromosomes 6, 14 and 20
- Some of the regions identified in agreement with previous regions associated with mastitis, fat metabolism and immune response

Acknowledgements

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Agriculture and
Agri-Food Canada

Agriculture et
Agroalimentaire Canada

Canadian Dairy
Commission

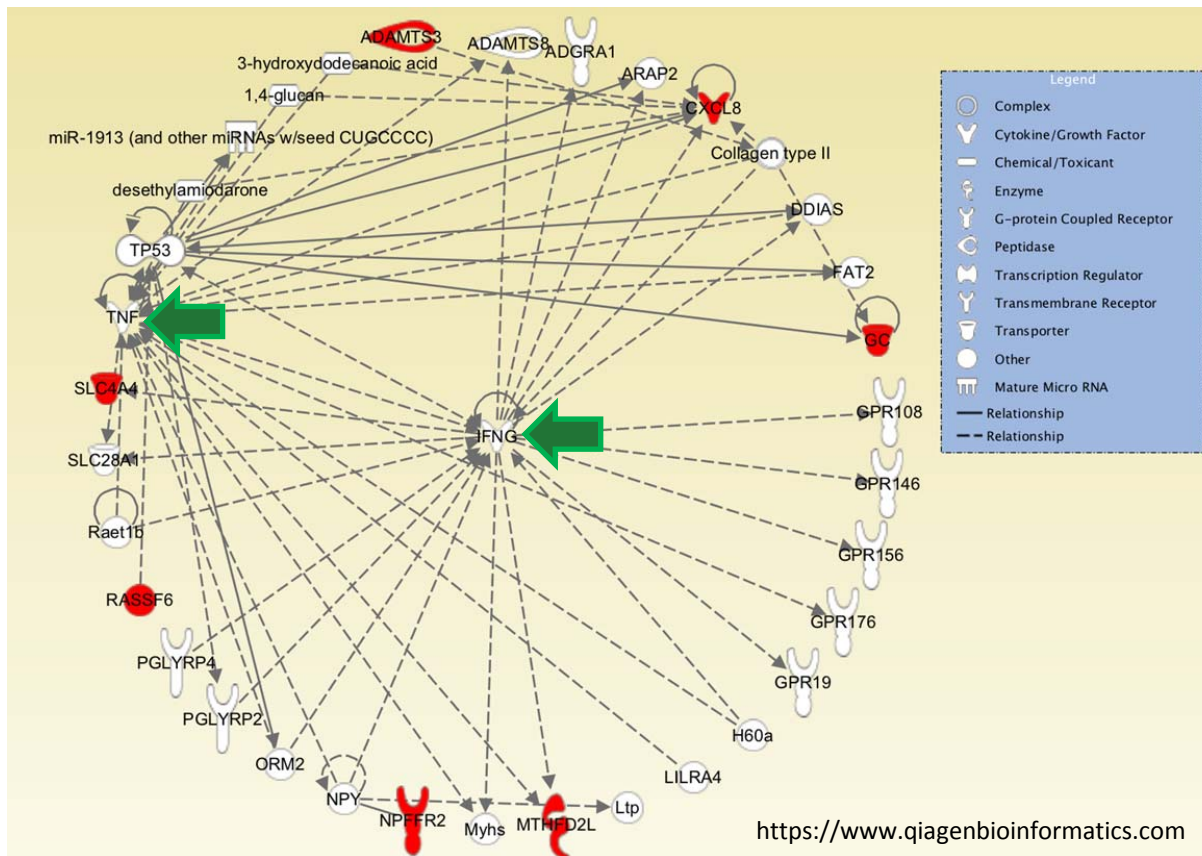
Commission
canadienne du lait



Back up slides

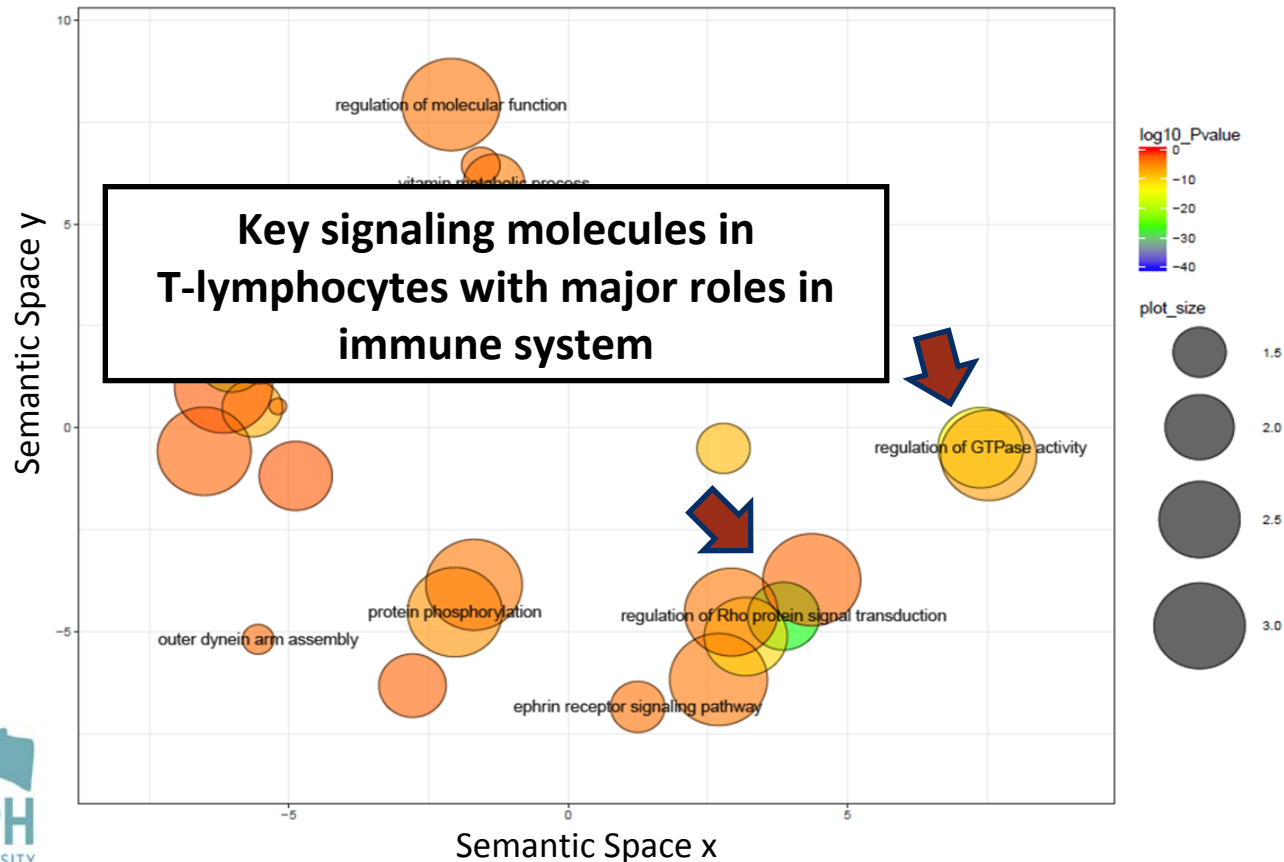


Ingenuity pathway analysis results



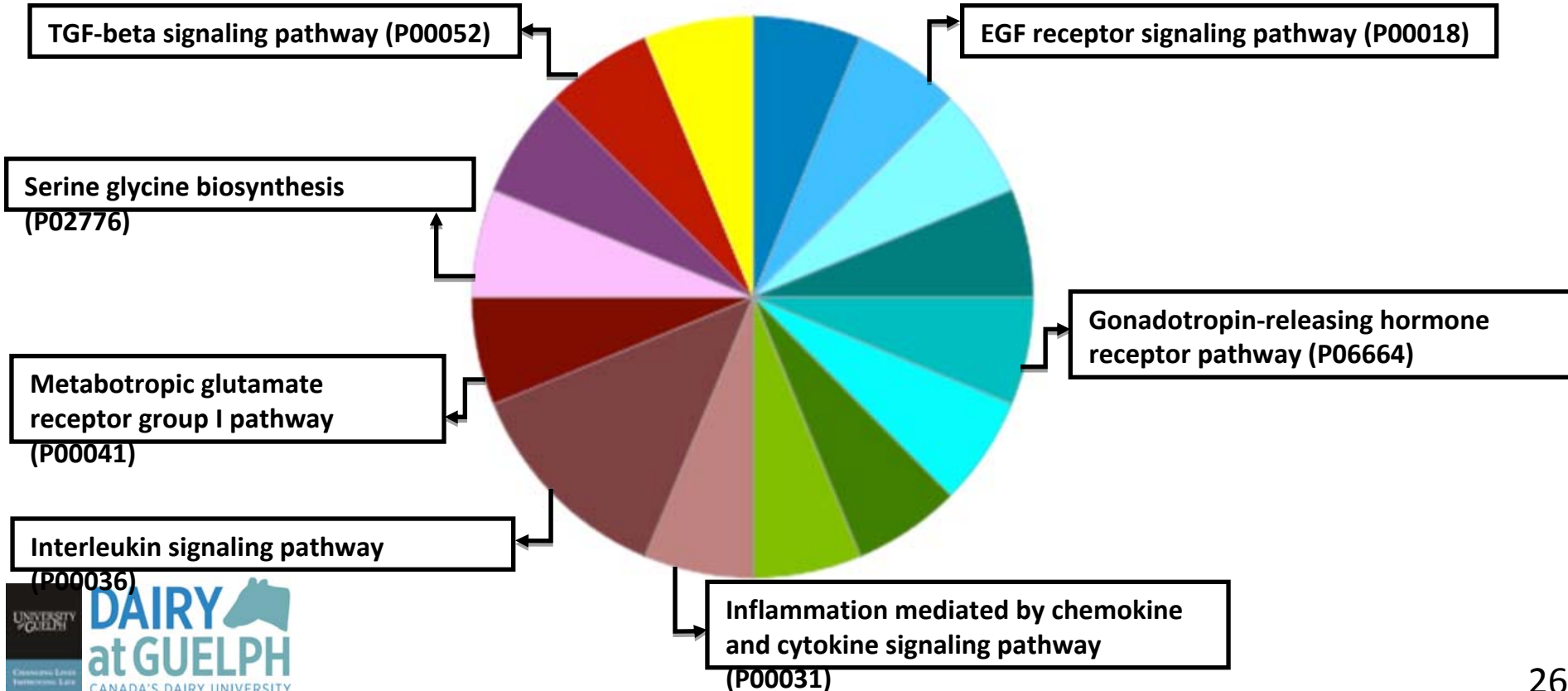
Prediction of the gene networks for candidate genes identified for (SCK1) 24

Semantic similarity scatterplot of the over-represented GO terms (REVIGO web-server)



Functional analysis-PANTHER PATHWAYS

Total # Genes: 76; Total # pathway hits: 16



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