

GENOME-WIDE STUDY OF STRUCTURAL VARIANTS IN FRENCH BOVINE DAIRY AND BEEF BREEDS



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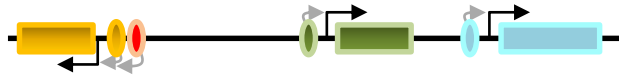


Structural Variations (SVs)

❖ Definition

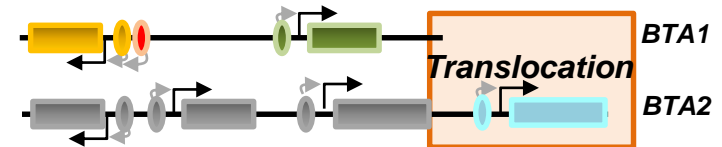
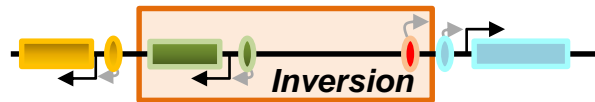
- ❑ Genomic alterations → DNA segments ≥ 50 nucleotides

A - Genomic reference regions

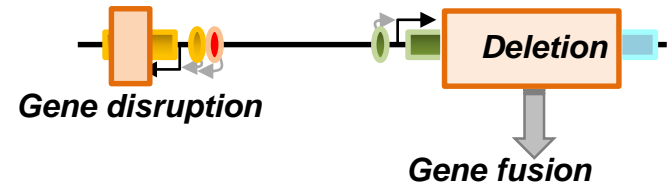
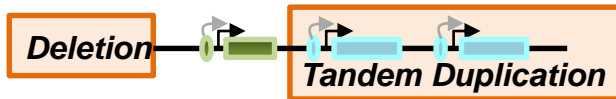


❖ Two classes of SVs

I - **Balanced SVs:** modify DNA structure



II - **Unbalanced SVs:** modify DNA copy number





Major goal of the project

❖ Scientific context:

- ❑ Rapid evolution of sequencing technologies

- ❑ Development of bioinformatics tools

➔ Identification of all genomic variations in several species

❖ In livestock species:

- ❑ Most studies mainly focused on SNPs and InDels

- ❑ Few studies dealt with SVs

➔ **Genome-wide study of SVs in cattle using whole genome sequences (WGSs)**

Origin of WGSs

❖ **Samples:** WGSs of 339 animals

Type	Breed	Number
Dairy	Holstein	68
	Montbéliarde	63
	Normande	43
	Abondance	9
	Tarentaise	8
Beef	Limousine	52
	Charolaise	43
	Blonde d'Aquitaine	38
	Salers	8
	Rouge des Prés	7

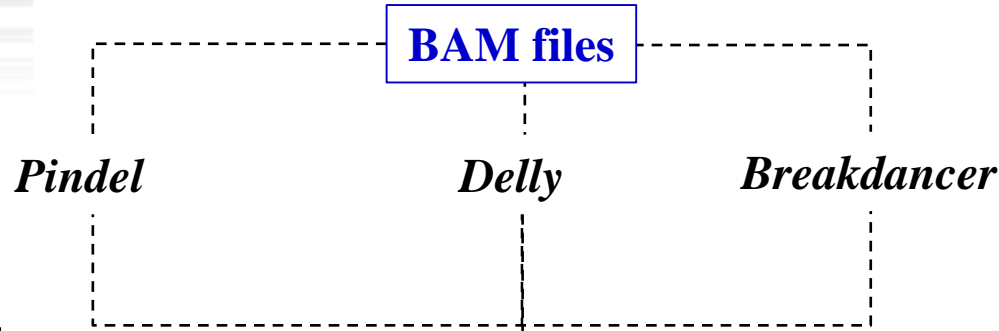
❖ **Sequencing technology:**

- ❑ 100 nt Paired-end sequences
- ❑ Illumina Hiseq2000

Strategy to study SVs



Step1: SV search



Step2: SV filtering/merging

- 1 - SVs common to the 3 tools
- 2 - no stretches of « N » around BP
- 3 - merge (90% RO threshold)

Filtered SV Regions (SVRs)

Step3: SV validation/characterization

- Annotation
- Experimental validation
- Link with genetic defects and complex traits

Results

❖ **Total number of identified SV regions :** **22 276**

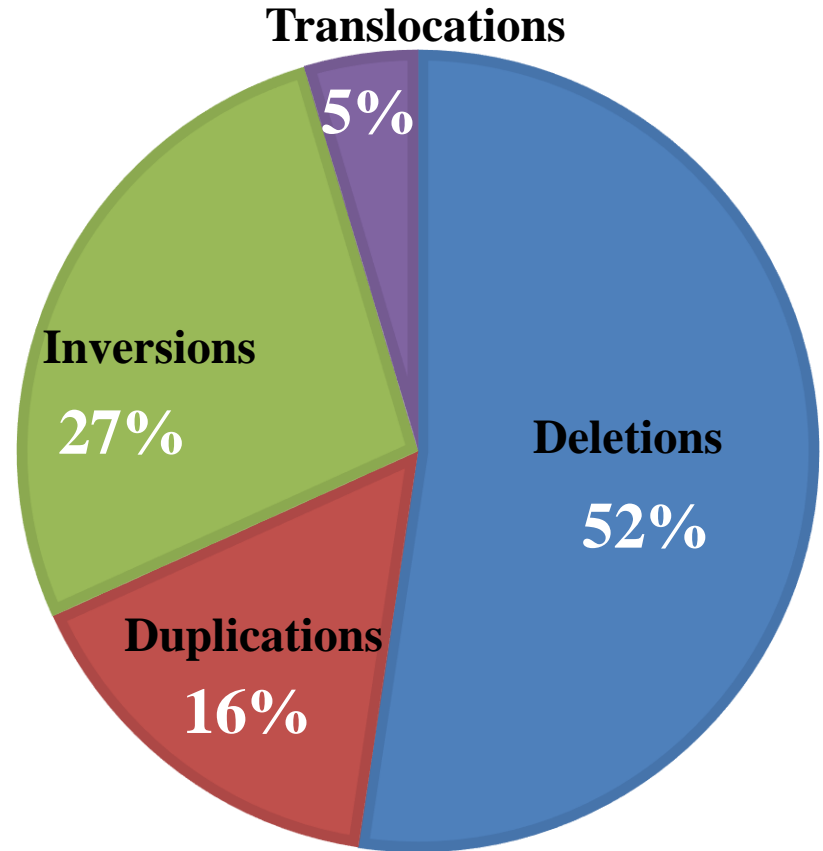
❖ **Similar fractions in other species**

❖ **Large number of deletions**

❑ **> 63% of deletions < 1Kb in size**

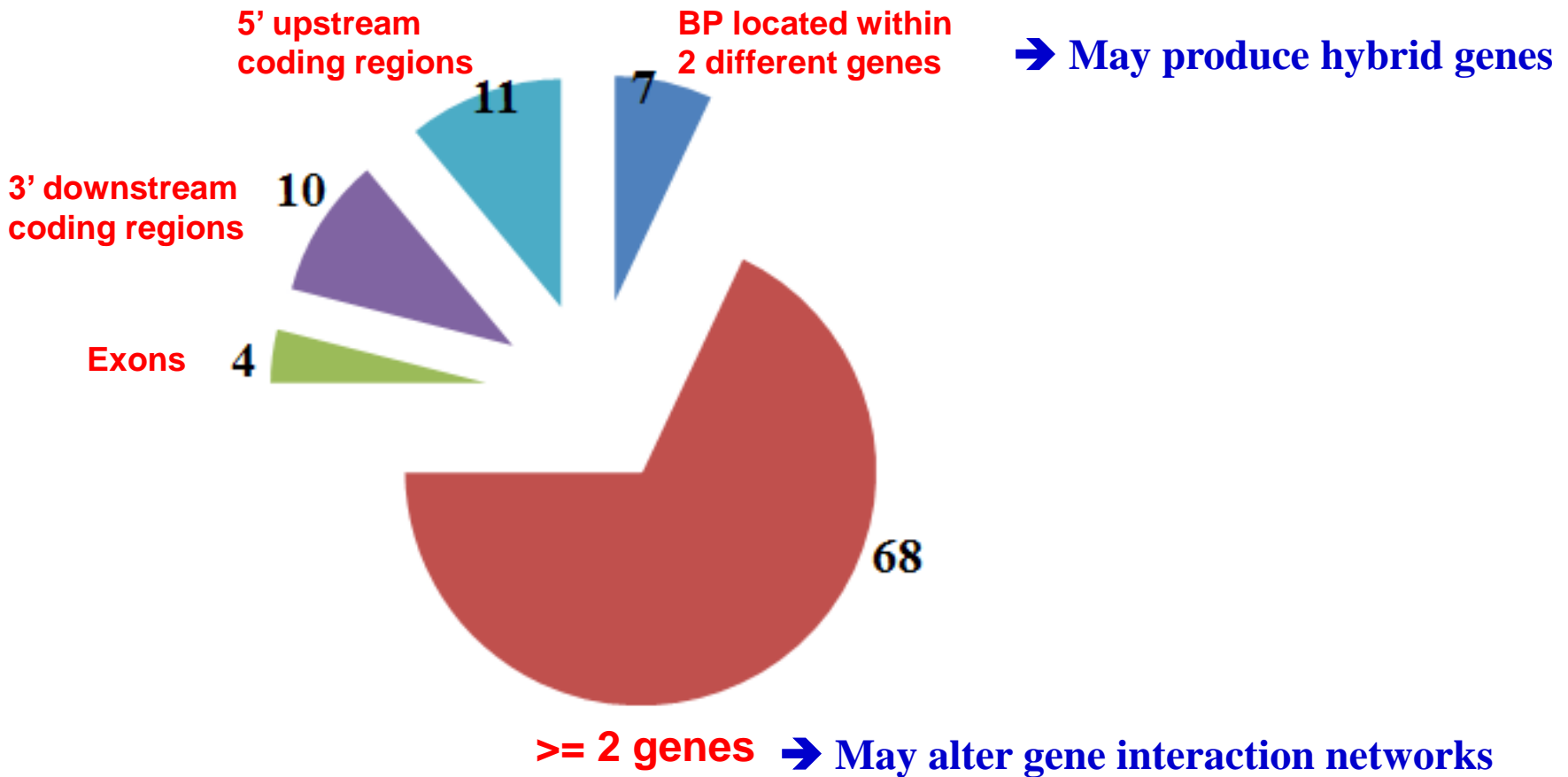
❑ **> 70% of inversions and duplications > 10 Kb in size**

❑ **Recombination mechanisms (in particular NAHR) → 2 fold deletions**



SV annotation

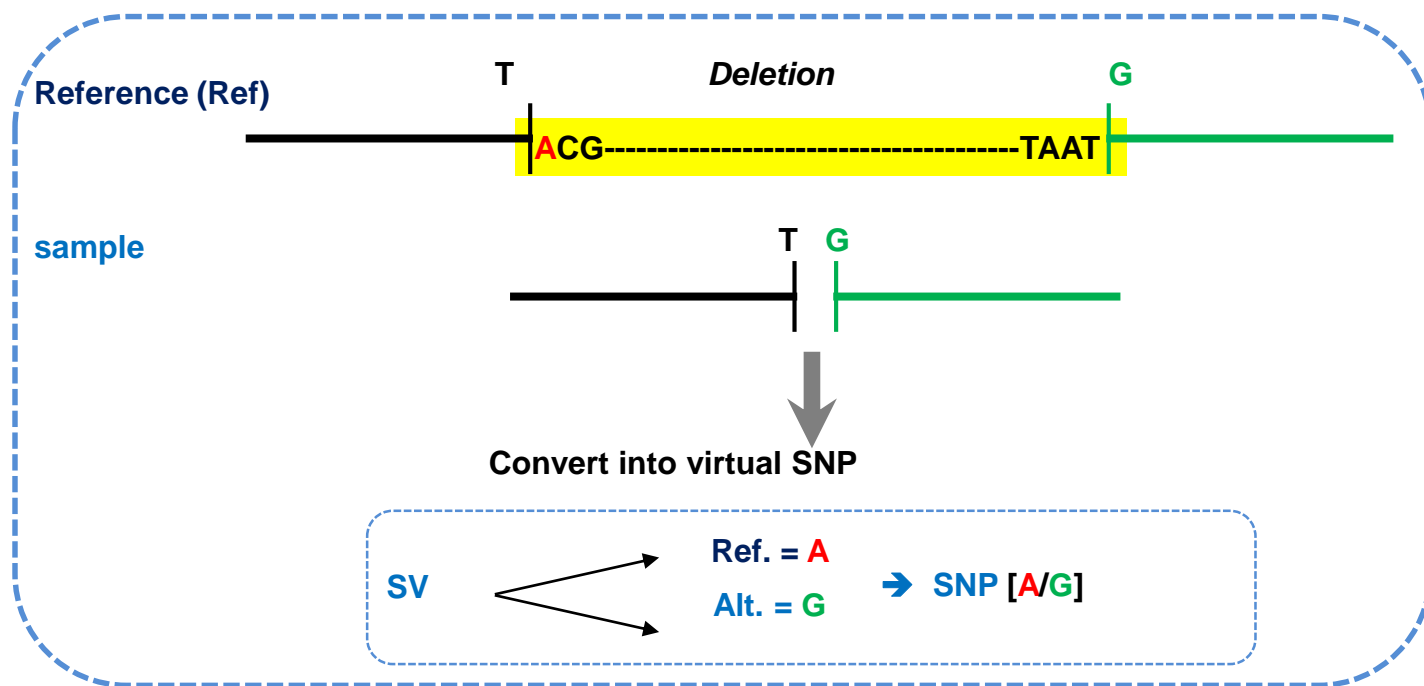
❖ Total number of annotated SVs: 2 415 (11%)



SV detection efficiency

❖ Experimental validation approach

- ❑ Large scale genotyping using the bovine EuroG10K SNP chip
- ❑ Conversion of SVs to “Virtual SNPs” (Nucleotide change around the 2 SV breakpoints)



Results of the experimental validation of SVs

Number of tested SVs : 331

Validation panel: 382 animals (15 dairy and beef breeds)

Number of successfully genotyped SVs: 255 (77%)

Validated SVs: 191 (75%)

❖ **Observed Minor Allele Frequency (MAF) : 0.2**

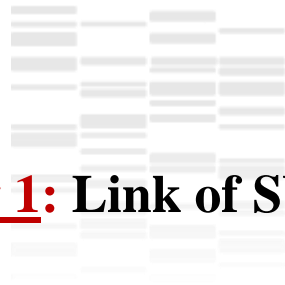
❖ **Observed heterozygosity rate (He): 0.28**

❖ **Polymorphic Information Content (PIC) : 0.23**

→ **Informative markers**

Linkage analyses

Biological impact of SVs



❖ **Strategy 1: Link of SVs with genetic defects**

❖ **Strategy 2: Link of SVs with complex traits**

Biological impact of SVs

❖ Strategy 1: Link of SVs with genetic defects

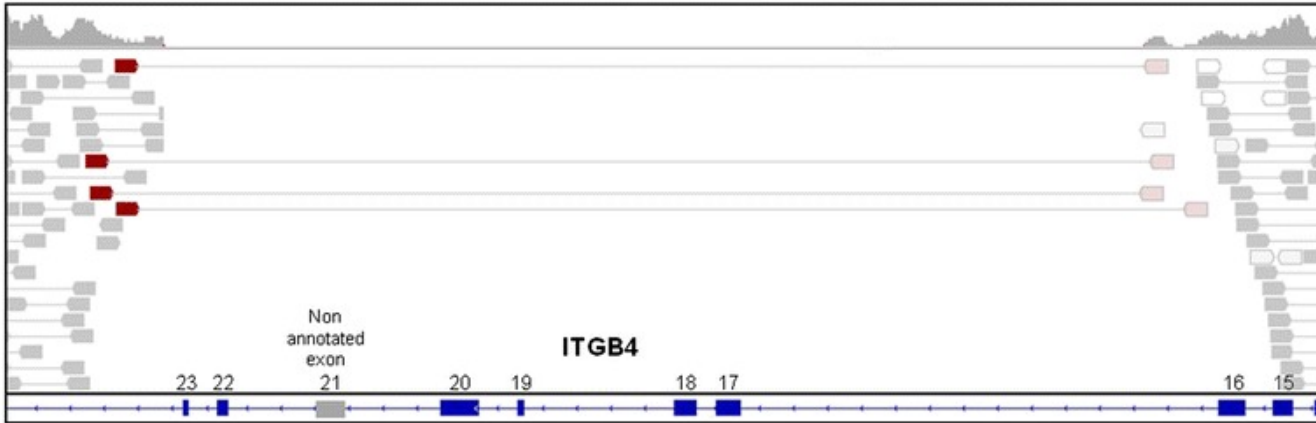
- ❑ Search for SVs in all animals already sequenced and declared as sick and/or carrying genetic defects.
- ❑ Interaction with ONAB (National Observatory of Bovine Abnormalities):
 - Identification of bovine defects resembling human diseases caused by SVs.
 - Sequencing of these animals to look for SVs responsible for these genetic defects.

❖ Strategy 2: Link of SVs with complex traits

Link of SVs with genetic defects

Example: a homozygous deletion located within the *ITGB4* gene coding regions

IGV screenshot of BTA19: 56.86 to 56.96 Mb



[Genet Sel Evol.](#) 2015; 47(1): 37.

Published online 2015 May 3. doi: [10.1186/s12711-015-0110-z](https://doi.org/10.1186/s12711-015-0110-z)

PMCID: PMC4417276

PMID: [25935160](https://pubmed.ncbi.nlm.nih.gov/25935160/)

Whole-genome sequencing identifies a homozygous deletion encompassing exons 17 to 23 of the *integrin beta 4* gene in a Charolais calf with junctional epidermolysis bullosa

[Pauline Michot](#), [Oscar Fantini](#), [Régis Braque](#), [Auréli Allais-Bonnet](#), [Romain Saintilan](#), [Cécile Grohs](#), [Johanna Barbieri](#), [Lucie Genestout](#), [Coralie Danchin-Burge](#), [Jean-Marie Gourreau](#), [Didier Boichard](#), [Didier Pin](#),[✉] and [Aurélien Capitan](#)[✉]

Biological impact of SVs



❖ Strategy 1: Link of SVs with genetic defects

- ❑ Search for SVs in all animals already sequenced and declared as sick and/or carrying genetic defects.
- ❑ Interaction with ONAB (National Observatory of Bovine Anomalies):
 - Identification of bovine defects resembling human diseases caused by SVs.
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❖ Strategy 2: Link of SVs with complex traits

- ❑ Use of validated SVs to genotype a large number of animals with phenotypes
- ❑ Search for potentially causal variants (GWAS + functional annotation of variants).

Association between SVs and production traits

❖ Genotyping data

Breed	Polymorphic SVs	Number of genotyped animals
Montbéliarde	160	10 920
Normande	125	4 679
Holstein	138	25 777

❖ Phenotypes

❑ Five milk production traits

- milk yield
- fat yield
- protein yield
- fat percentage
- protein percentage

❖ Software

- ❑ GCTA (Yang et al., 2011)

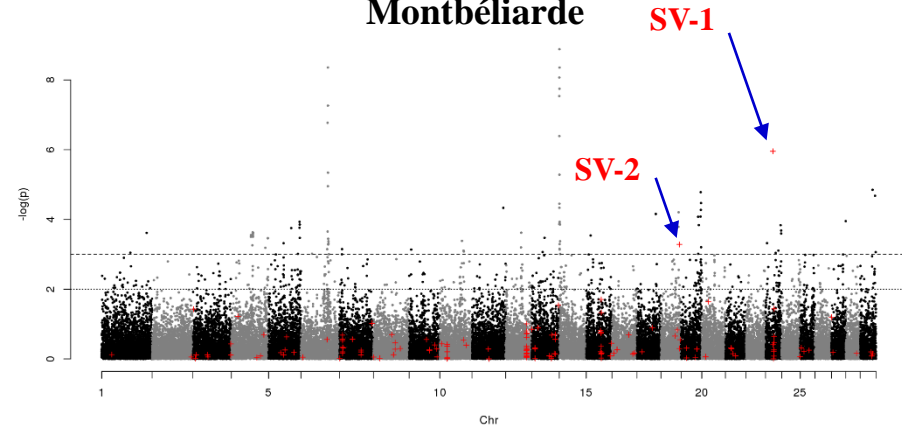
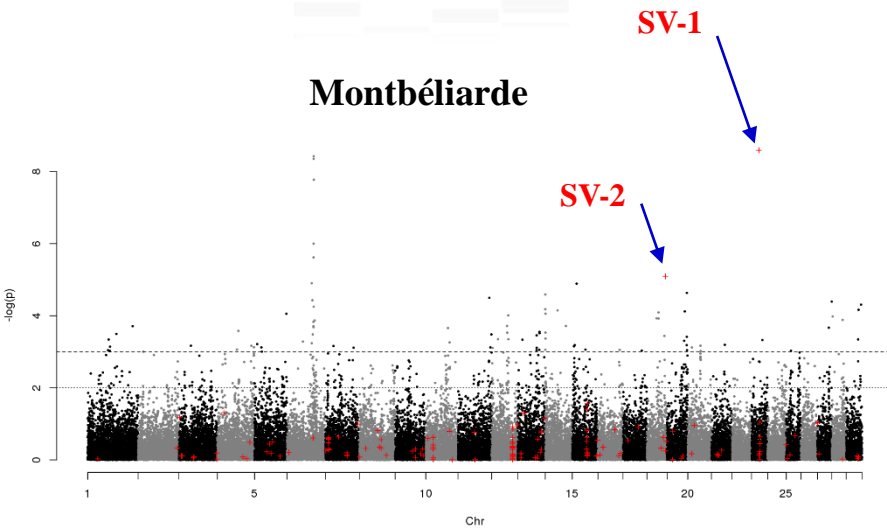
GWAS preliminary Results

Protein yield

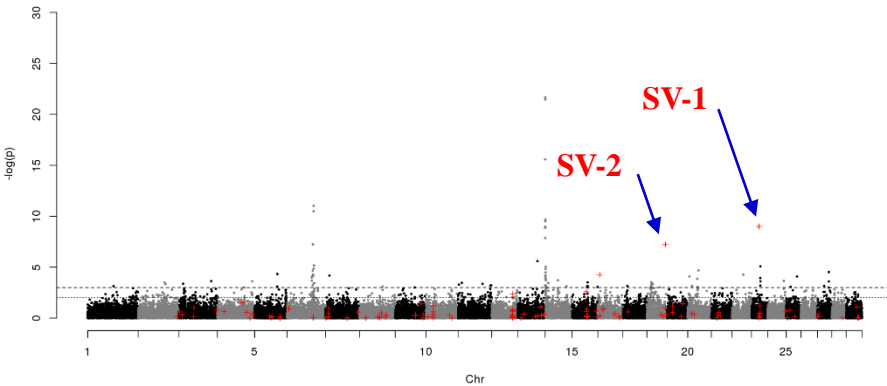
Milk yield

Montbéliarde

Montbéliarde



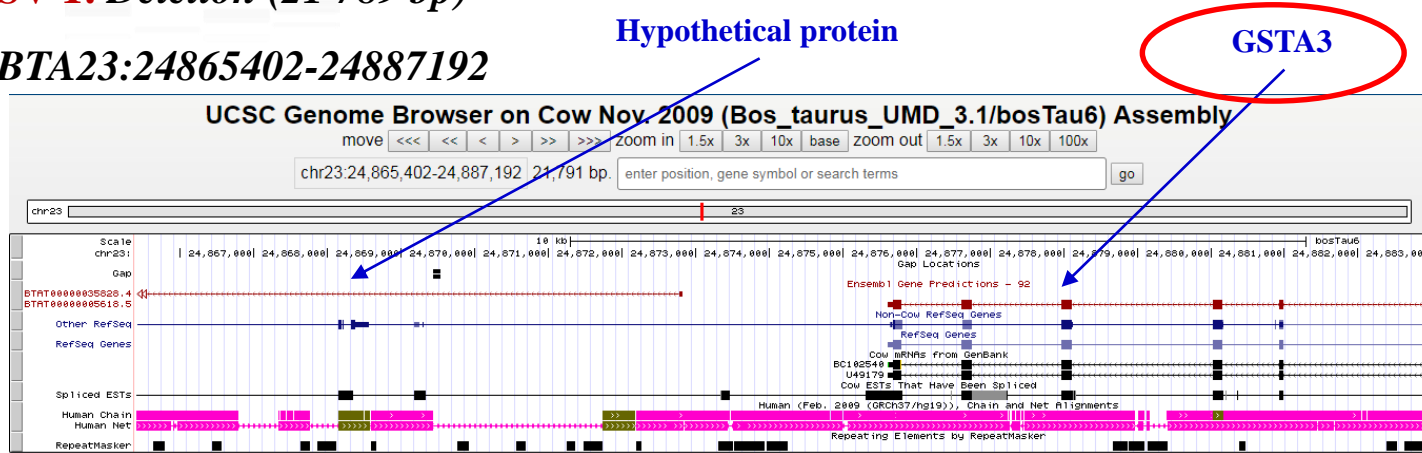
Holstein



Gene content of both deletions

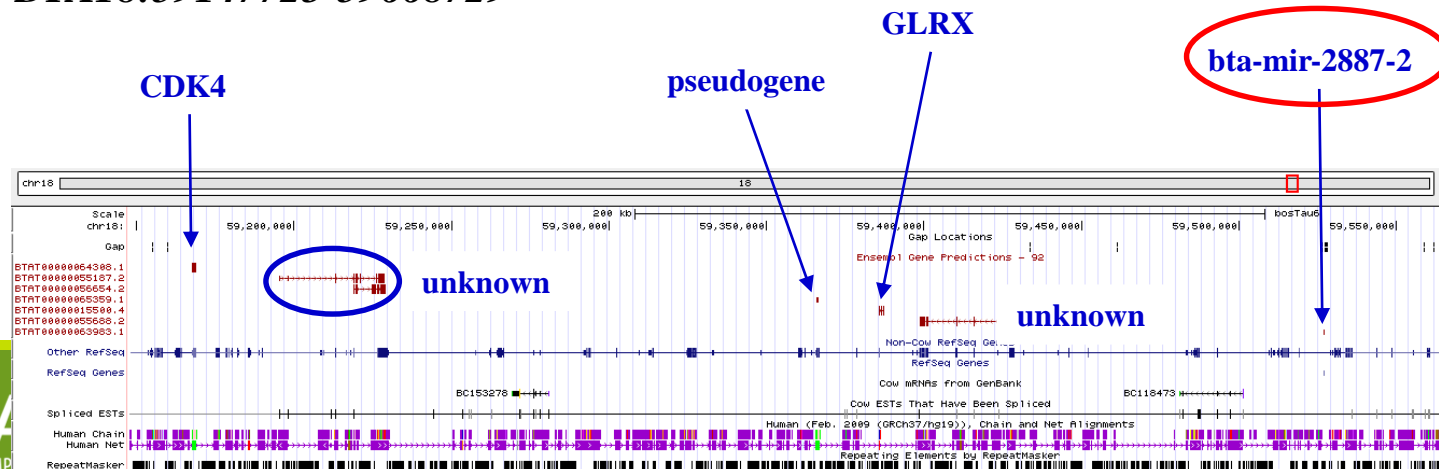
SV-1: Deletion (21 789 bp)

BTA23:24865402-24887192



SV-2: Deletion (521 005 bp)

BTA18:59147723-59668729





Conclusions

❖ Pan-genomic evaluation of SVs in cattle using whole genome sequences

❖ State of the art today

❑ Parcel Mapping of SVs

➤ ➔ Large scale identification

➤ ➔ Characterization of the genetic variability of complex traits

❖ Future spin off

❑ Improve genome knowledge

❑ Improve accuracy and persistence of genetic evaluations



❖ *Thanks for your attention*

