# Whole genome association analysis of resistance / susceptibility to paratuberculosis in French Holstein and Normande cattle

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UMR BioEpAR UMR GABI UMR ISP
GDS Bretagne GDS France GDS Pays-de-la-Loire GDS Orne
Gdma SEINE MARITIME GROUPEMENT DE DÉFENSE CONTRE LES MALADIES DES ANIMAUX

# Paratuberculosis (MAP)

**Paratuberculosis** (**MAP**) or Johne's disease
= endemic transmissible disease in ruminants due to
*Mycobacterium avium subsp. paratuberculosis* (*Map*)

Gut lesions
Rapid weight losses & diarrhea

Economic losses
Negative effects on animal welfare

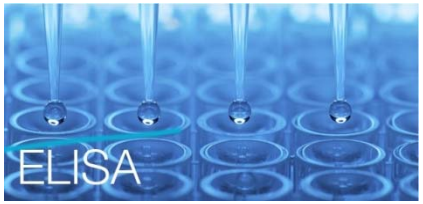
# Paratuberculosis (MAP)

Difficulties to control MAP because

✓ **No efficient treatment** available

✓ Young animals infected (<6 months) but **long latency period** before first symptoms (after 2-6 years)

✓ Tests to detect infected animals (ELISA on blood or PCR on faeces) have a **poor sensitivity** that can be compensated by repeated testing



A better **genetic resistance**
could contribute to control the disease

# French PICSAR & PARADIGM projects

2 related on-going projects on integrated control of MAP

**PICSAR**

INRA

**PARADIGM**

APIS-GENE and GDS France

**Preliminary results**

**Genomic analysis in cattle**

# Objectives

**Genomic study** of confirmed host phenotypes to identify genomic regions (**QTL**) affecting susceptibility to MAP

in two dairy cattle breeds

***Holstein***

***Normande***

➔ Better understand the **genetic determinism** of the susceptibility to MAP

➔ Build a first reference population to pave the way for **genomic selection** against the disease

# Material & methods: accurate phenotypes

***Herds with detected cases enrolled in surveillance program***

regular blood **ELISA** + fecal **PCR** tests

***Selection of cows in these herds***

1) With 2 ELISA & 2 PCR tests distant from > 8 months and
2) With **clear** and **concordant** negative or positive results, excluding intermediate ones

| | Negative | Intermediate | Positive |
|---|---|---|---|
| ELISA IDDEX S/P | < 45 | | ≥ 45 |
| PCR ADIAGENE Ct | ≥ 40 | | ≤ 35 |

3) Controls are negative cows **at least 72 months old** and **born in the same herd and in the same time period** as affected cows

# Material & methods: accurate phenotypes

*Selection of cows in these herds*

Cows with uncertain phenotypes removed

***Additional analyses to confirm phenotypes***
1 ELISA & 1 PCR test

Cows with discordant phenotypes removed

**647 CASES**
**210 Normande**
**437 Holstein**

**747 CONTROLS**
**195 Normande**
**552 Holstein**

# Material & methods: genotypes

All the **1,394** cows with phenotypes
genotyped with the 50k Beadchip
with about 40,000 informative genetic markers
distributed over the 29 autosomes (BTA)

**405**
Normande
**NOR**

**989**
Holstein
**HOL**

~ 60% of the total targeted population in the project

# Material & methods: case-control GWAS

**Within breed case-control GWAS** (Genome Wide Association Study) with GCTA software (Yang et al., 2011)

Each genetic marker tested **one by one**

MAF (Minor Allele Frequency) > 5%

**Structure of the population** accounted for through a **polygenic effect** of animals, in order to avoid spurious associations

Estimated with a Genomic Relationship Matrix
calculated with the markers of the 50K Beadchip

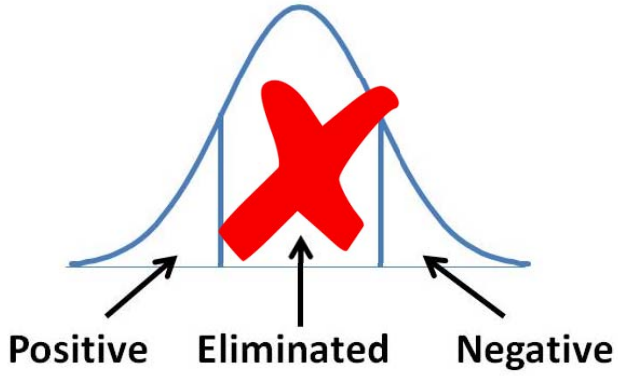
# Results: $h^2$ of resistance to MAP

## Heritability $h^2$

$h^2$ = 60%

$h^2$ = 46%

Higher than expected ~30%
=> probably due to selection of phenotypes (elimination of intermediate phenotypes)



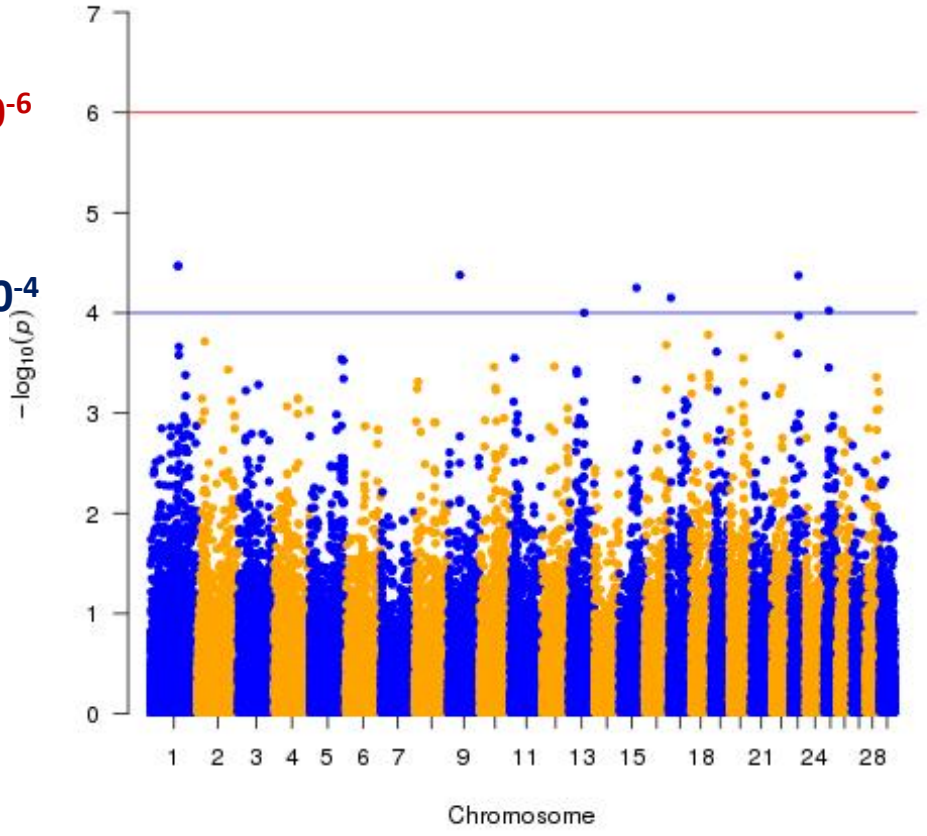

Nevertheless, **genetic variation** of susceptibility to MAP appears to be **large**

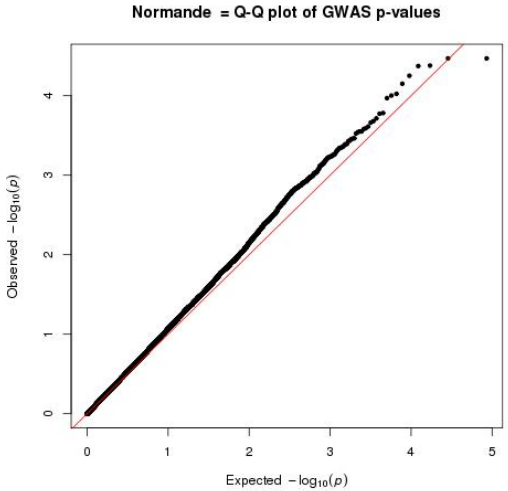
# Results: GWAS in Normande

**Manhattan Plot at the genome level**

p-value = $10^{-6}$

p-value = $10^{-4}$



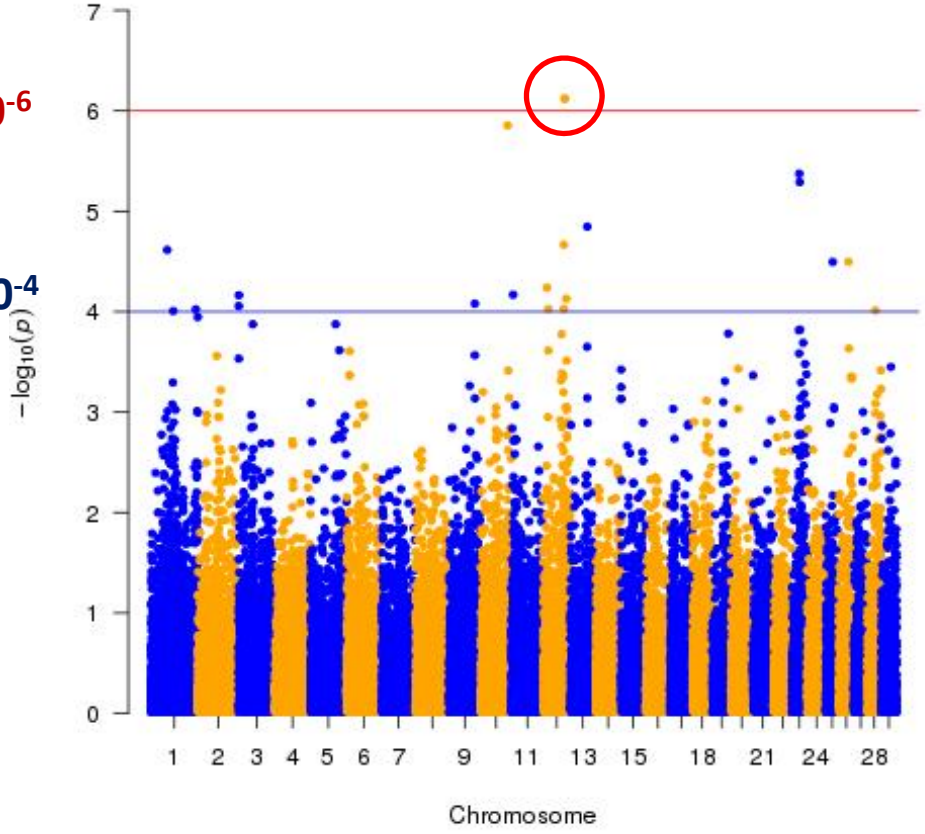



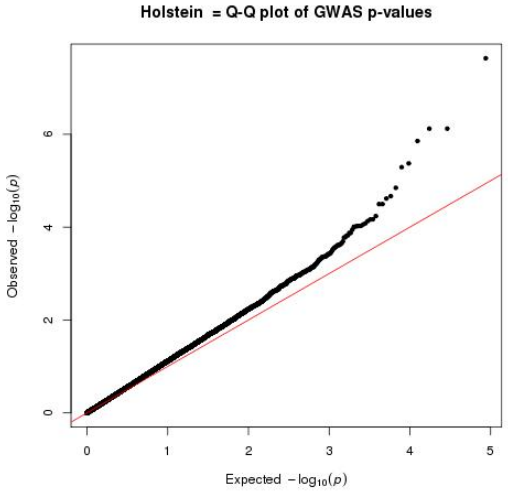
# Results: GWAS in Holstein

**Manhattan Plot at the genome level**

p-value = $10^{-6}$

p-value = $10^{-4}$



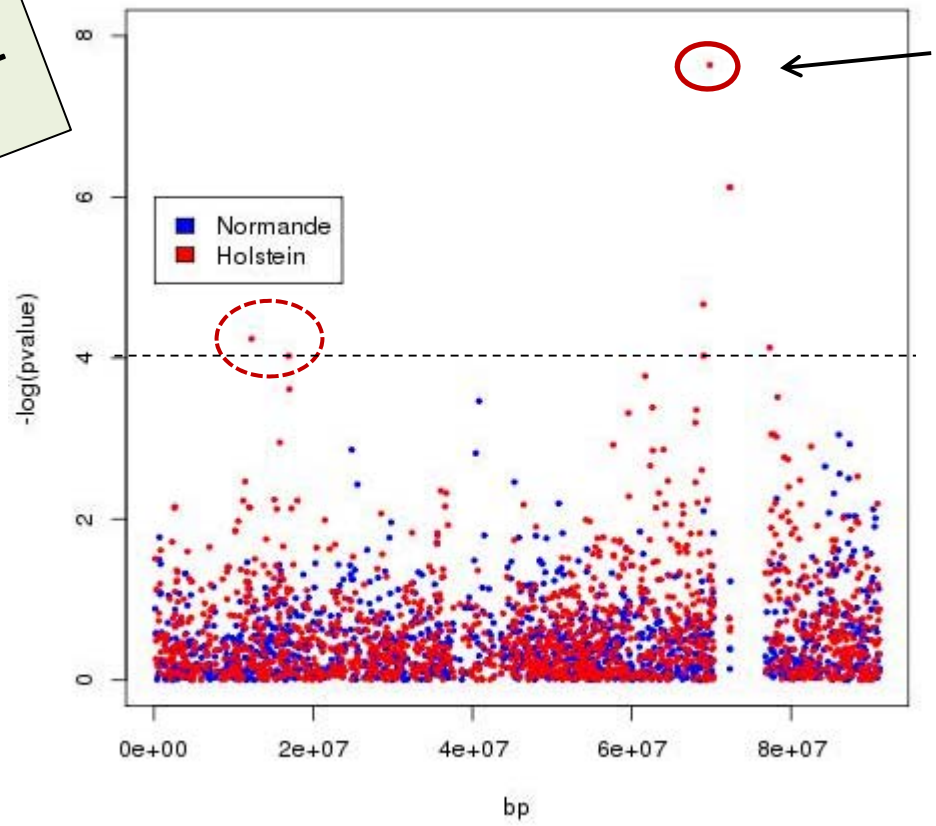



# GWAS results: the most significant QTL

**Manhattan Plot on one chromosome**

The most significant QTL is located at 70Mb in HOL (p-value = $2.10^{-8}$)

Zoom on **chromosome 12** in **2 breeds**



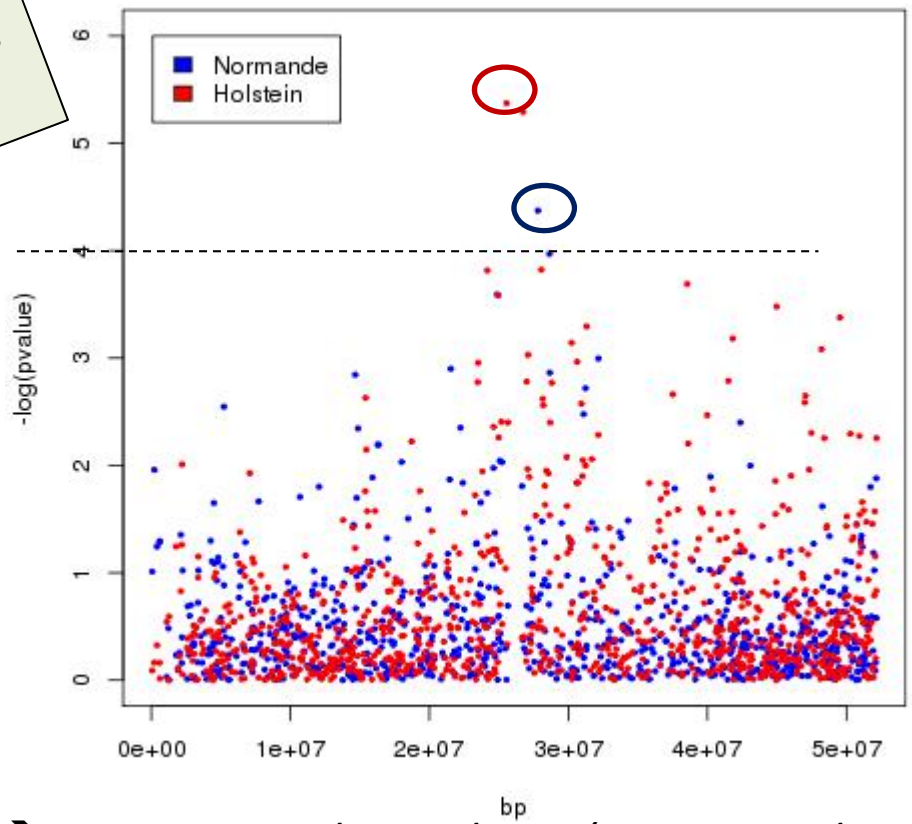

ABCC4 gene (Crohn's disease)

➔ *1 QTL at 70 Mb in Holstein (Minozzi et al., 2012)*

# GWAS results: a QTL shared between breeds

A QTL found in both breeds located at 27Mb in the **MHC region** ($4.10^{-6}$ < p-value < $4.10^{-5}$)

Zoom on **chromosome 23** in **2 breeds**



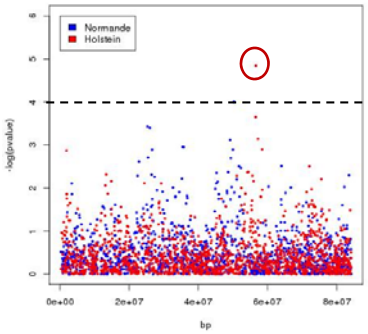
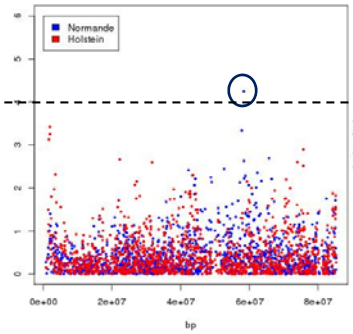
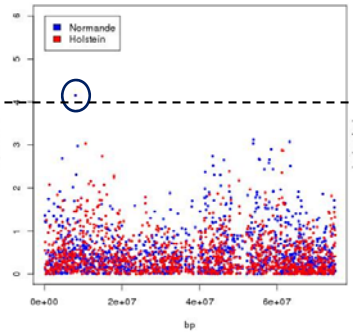
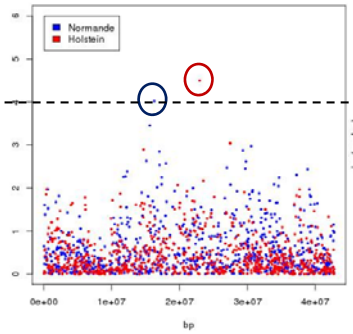
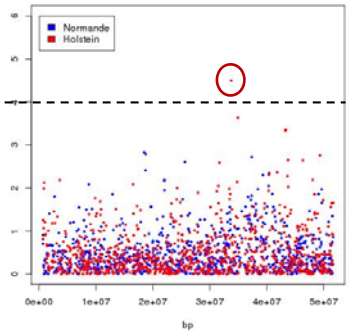
➔ *1 QTL at 32 Mb in Holstein (Minozzi et al., 2012)*

➔ *1 QTL at 27 Mb in Jersey (Zare et al., 2014)*

# GWAS results: QTL previously described

Normande
Holstein

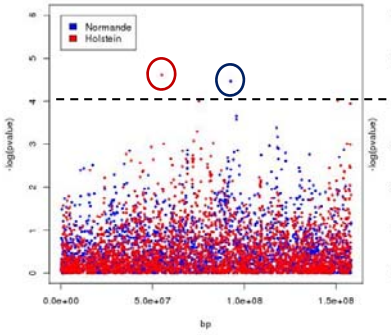
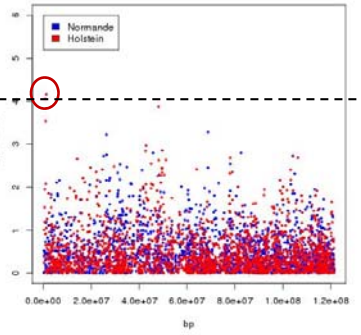
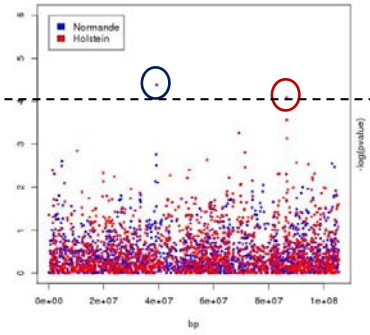
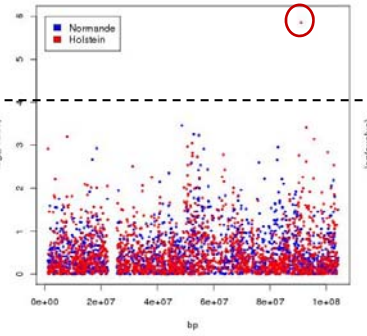
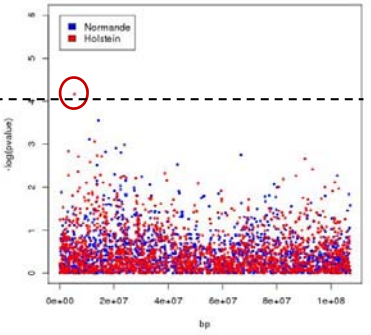
$7.10^{-5} < \text{p-value} < 10^{-4}$

| BTA 13 | BTA 15 | BTA 17 | BTA 25 | BTA 26 |
|---|---|---|---|---|
|  |  |  |  |  |
| HOL at 57Mb | NOR at 58Mb | NOR at 8Mb | NOR at 16Mb<br>HOL at 23Mb | HOL at 34Mb |
| 65 Mb<br>in HOL | 60 Mb<br>in HOL-Friesian | 9 Mb<br>in HOL | 19 Mb<br>in Jersey & HOL | 37 Mb<br>in HOL-Friesian |
| *Minozzi et al., 2012* | *Van Hulzen et al., 2012* | *Alpay et al., 2014* | *Sallam et al., PAG2015*<br>28 Mb<br>in HOL<br>*Minozzi et al., 2012* | *Van Hulzen et al., 2012* |

# GWAS results: QTL newly described

Normande
Holstein

$7.10^{-5} < \text{p-value} < 10^{-4}$

| ***BTA 1*** | ***BTA 3*** | ***BTA 9*** | ***BTA 10*** | ***BTA 11*** |
|---|---|---|---|---|
|  |  |  |  |  |
| HOL at 55Mb<br>NOR at 93Mb | HOL at 1.2Mb | NOR at 39Mb<br>HOL at 87Mb | HOL at 91Mb | HOL at 5.5Mb |

# Conclusions

**Relevant & accurate** contrasted phenotypes

↓

**Large genetic variability** of susceptibility to MAP

↓

Identification of **numerous QTL** in Normande and Holstein dairy cattle with high or moderate significant effects on **susceptibility to MAP**

↓

11 QTL in HOL

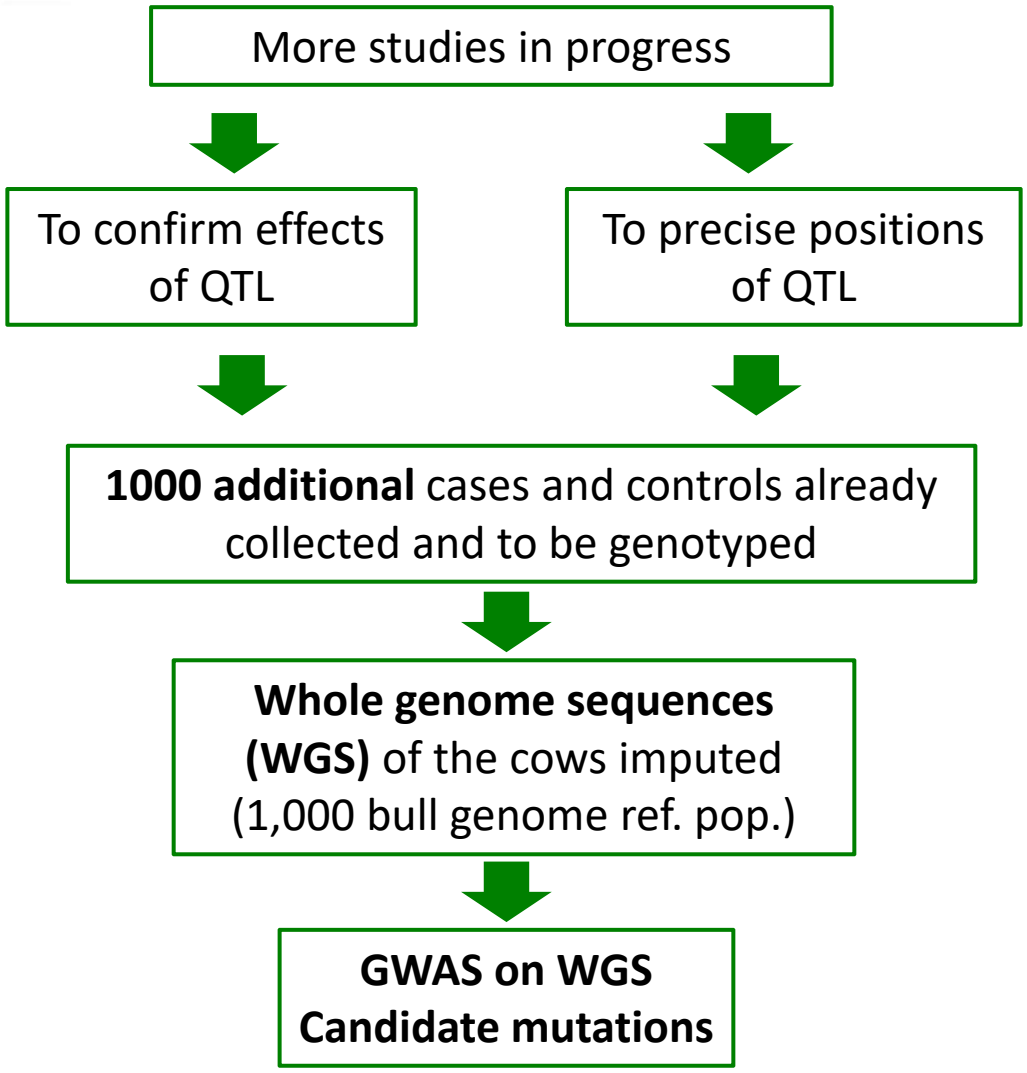
6 QTL in NOR

↓

Only one shared between breeds (but detection power higher in HOL / NOR)

**7 NEW QTL**

# Perspectives



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# Thank you for your attention

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