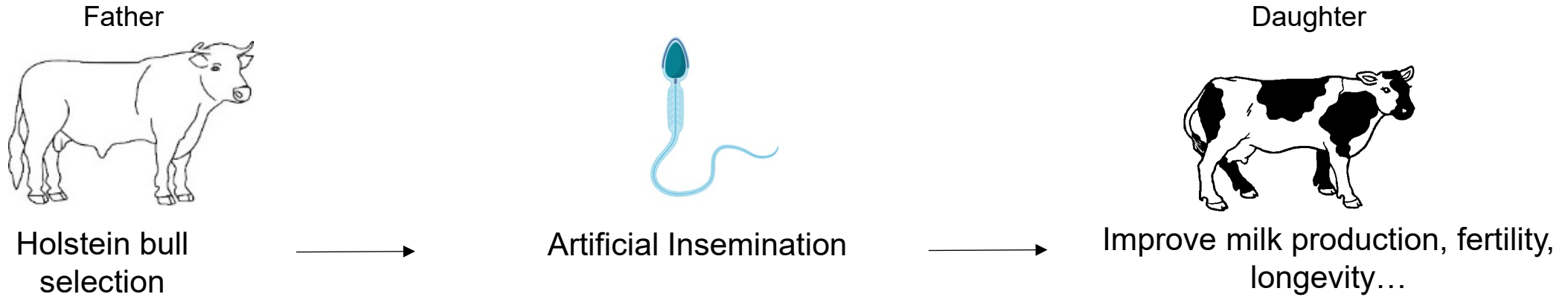


➤ Transmission of sperm DNA methylation patterns to the embryo in cattle: a genome-wide analysis

75th EAAP 2024 Annual meeting – Florence, Italy
03/09/2024

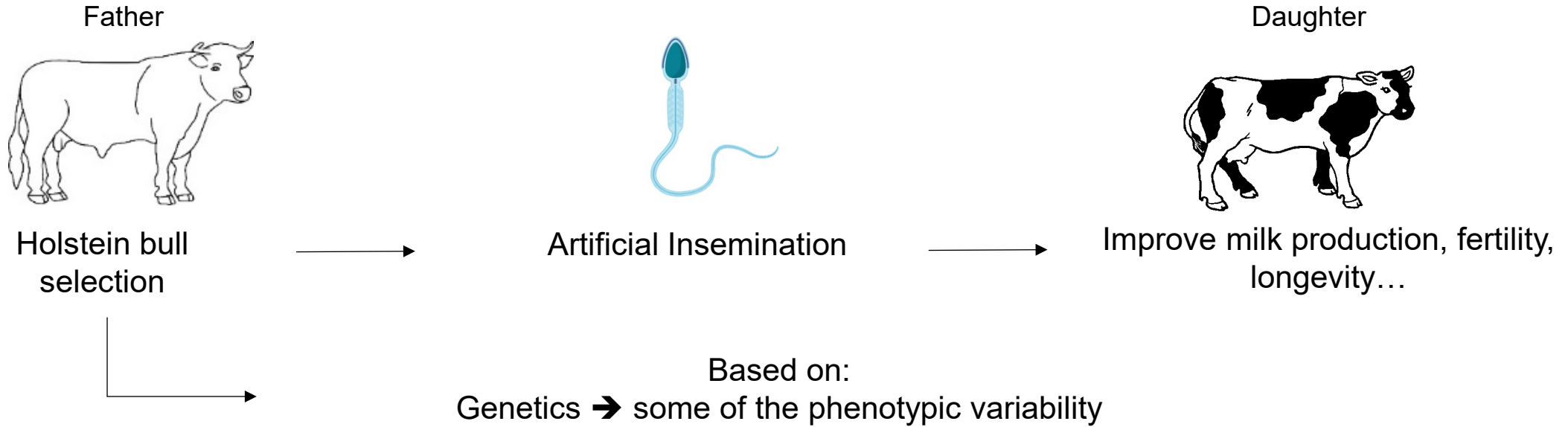
PhD student
Amrita Raja Ravi Shankar
amrita.raja-ravi-shankar@inrae.fr

Bovine selection



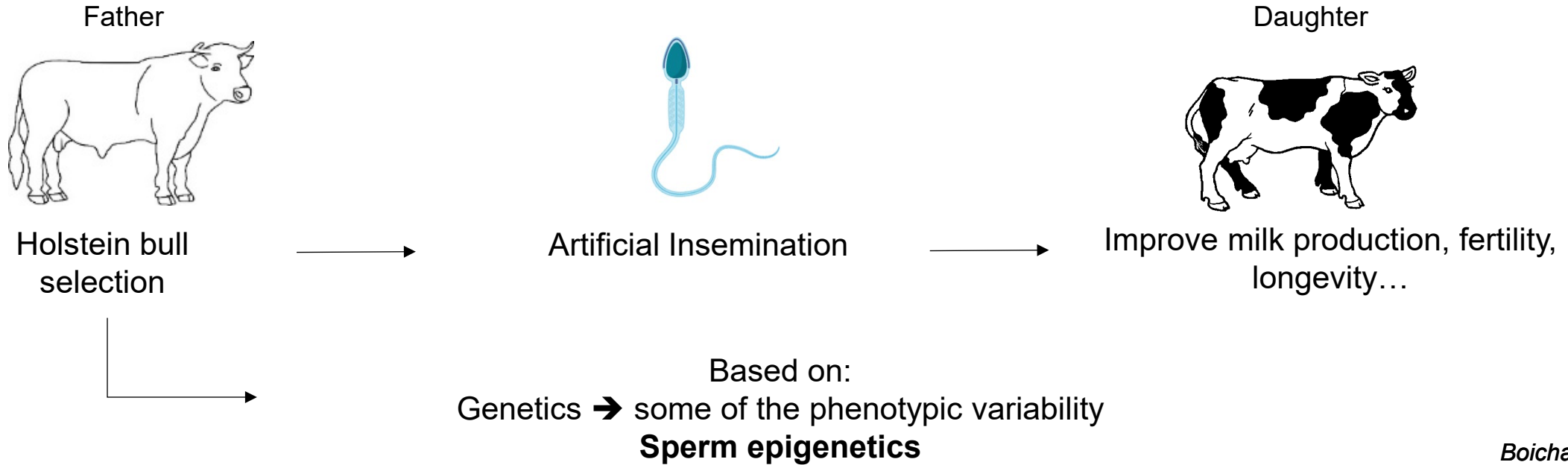
Boichard et al, 2012

Bovine selection



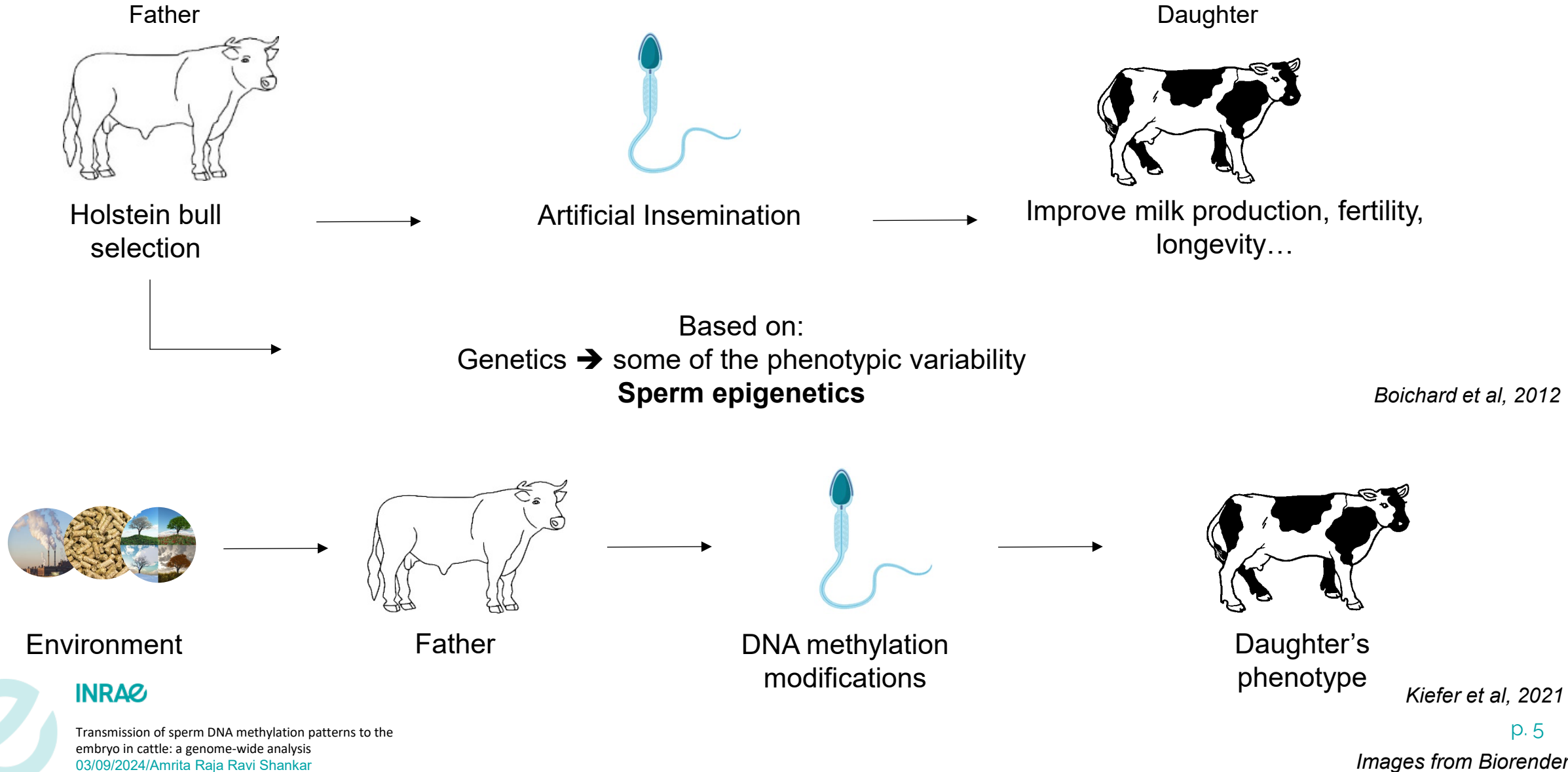
Boichard et al, 2012

Bovine selection

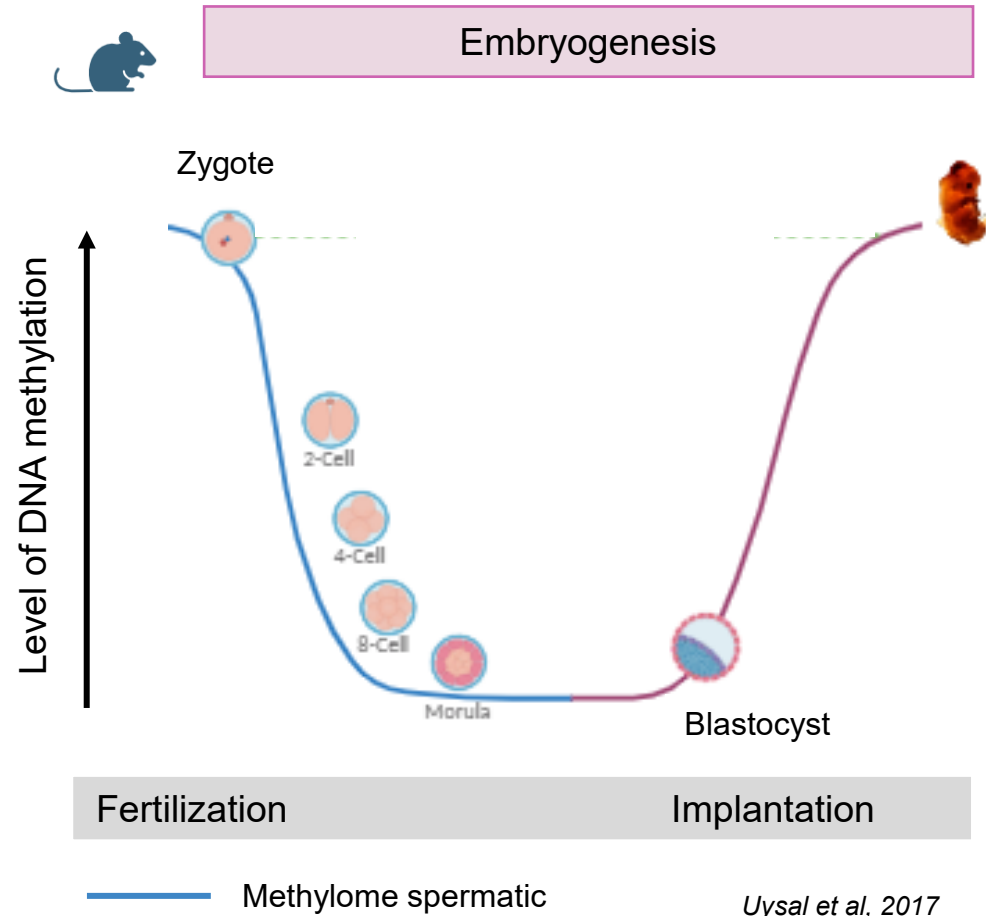


Boichard et al, 2012

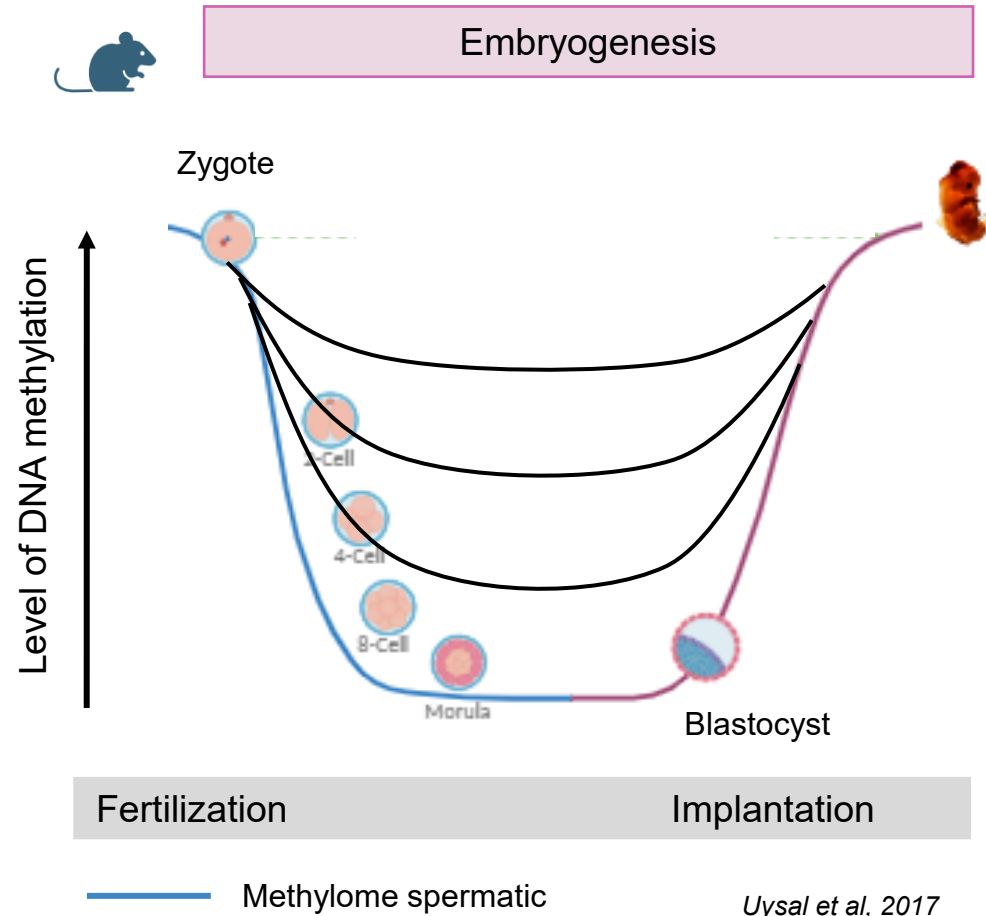
Impact of the father's environment on offspring's phenotype



Epigenetic reprogramming during embryogenesis



Some loci remain partially methylated during embryogenesis



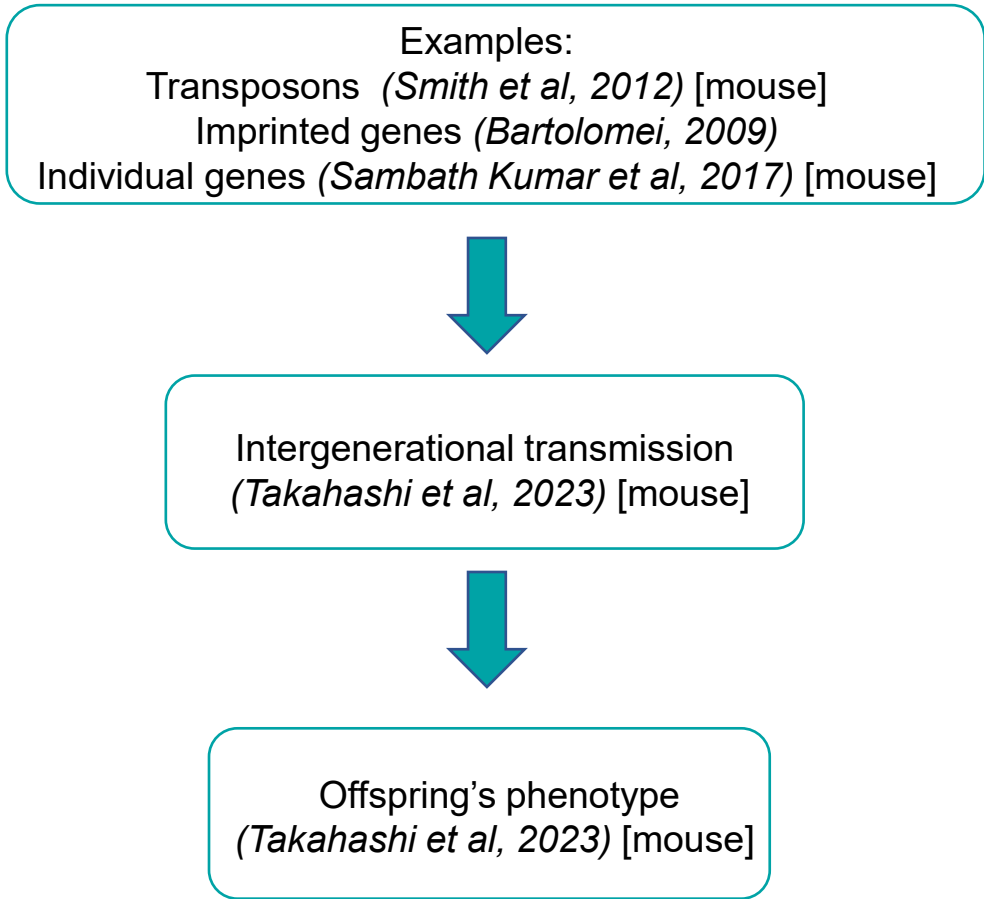
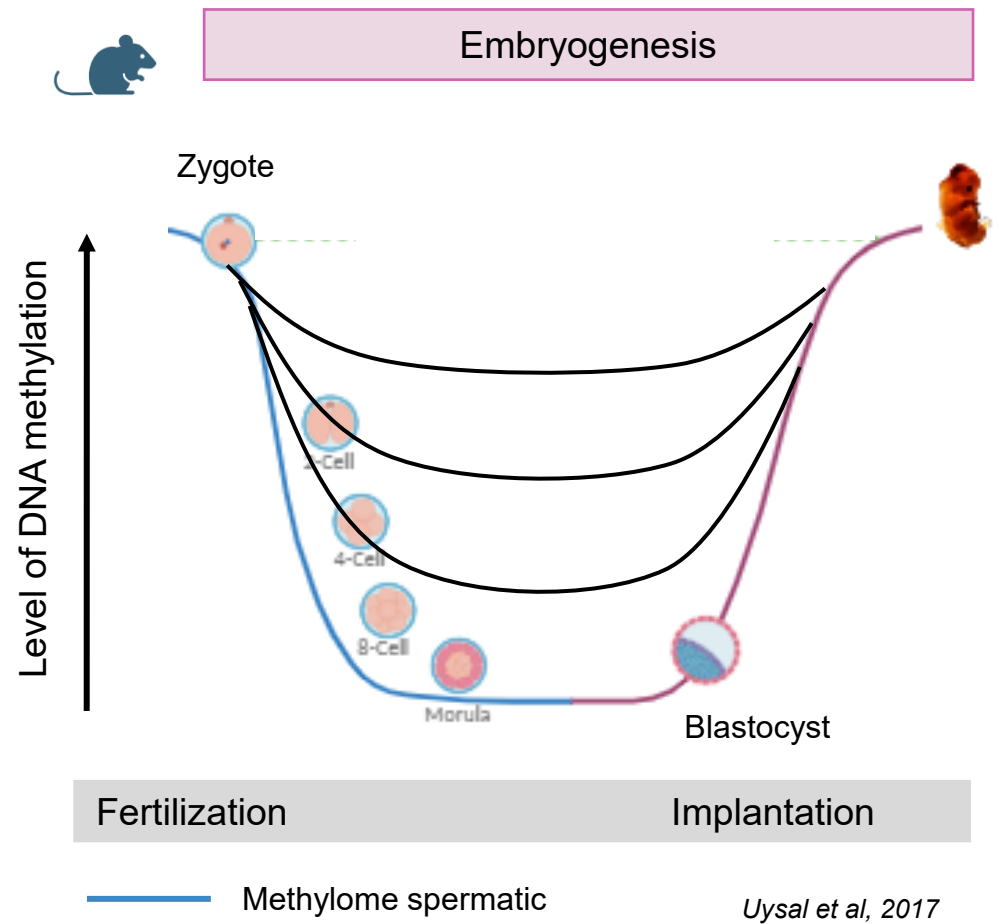
Examples:
Transposons (*Smith et al, 2012*) [mouse]
Imprinted genes (*Bartolomei, 2009*)
Individual genes (*Sambath Kumar et al, 2017*) [mouse]

Intergenerational transmission
(*Takahashi et al, 2023*) [mouse]

Offspring's phenotype
(*Takahashi et al, 2023*) [mouse]

Context	Question	Experimental design	Results	Conclusion / Prospects
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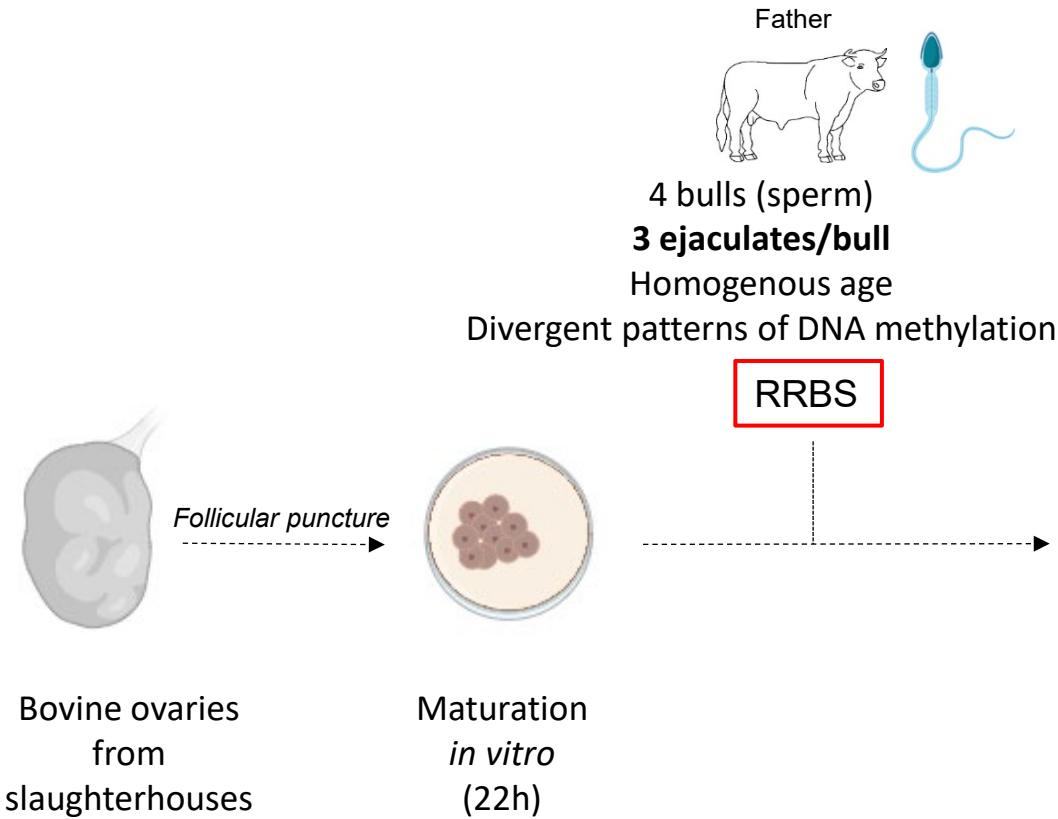
Some loci remain partially methylated during embryogenesis



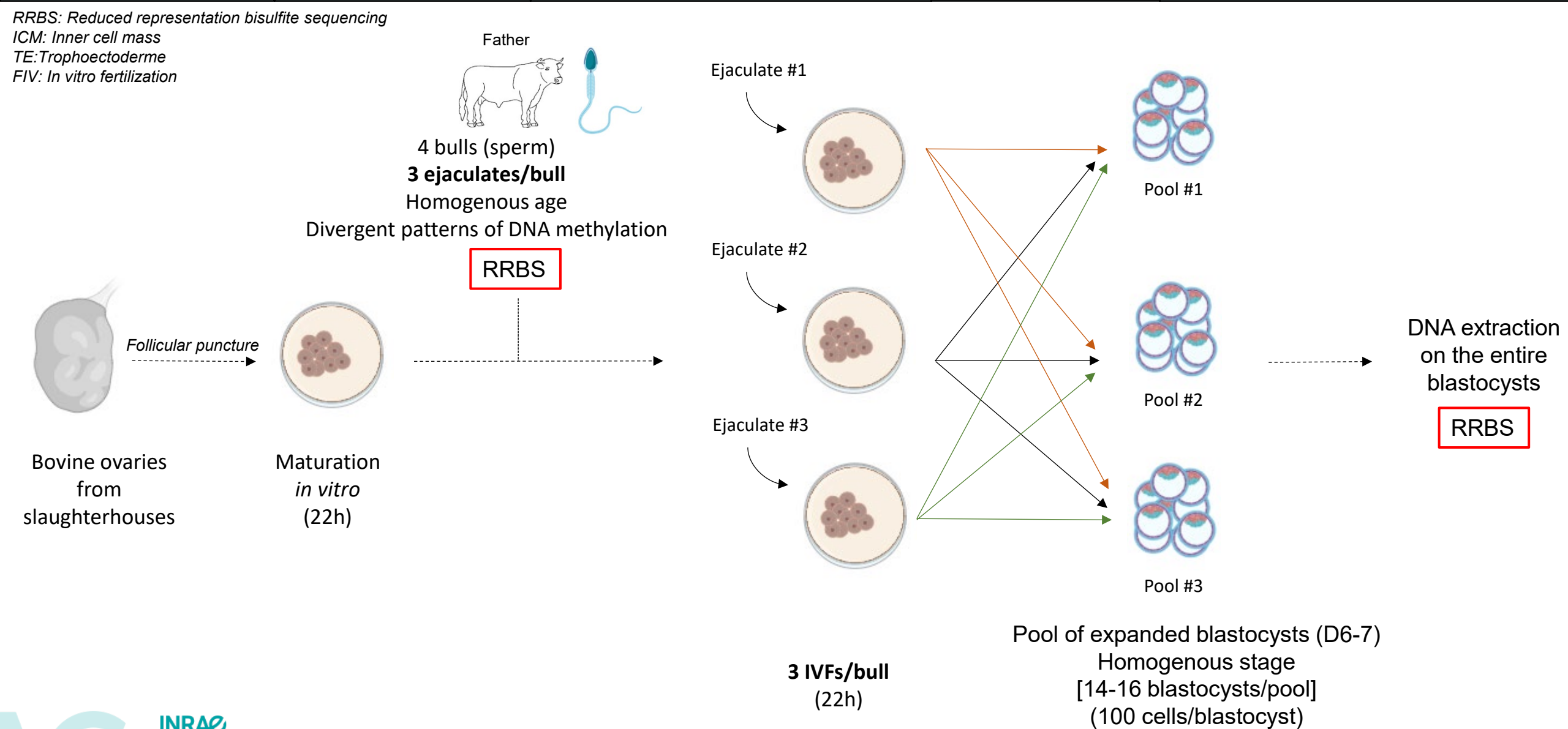
Which CpGs remain partially methylated in bovine embryos that could mediate paternal effects?

Context	Question	Experimental design	Results	Conclusion / Prospects
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RRBS: Reduced representation bisulfite sequencing

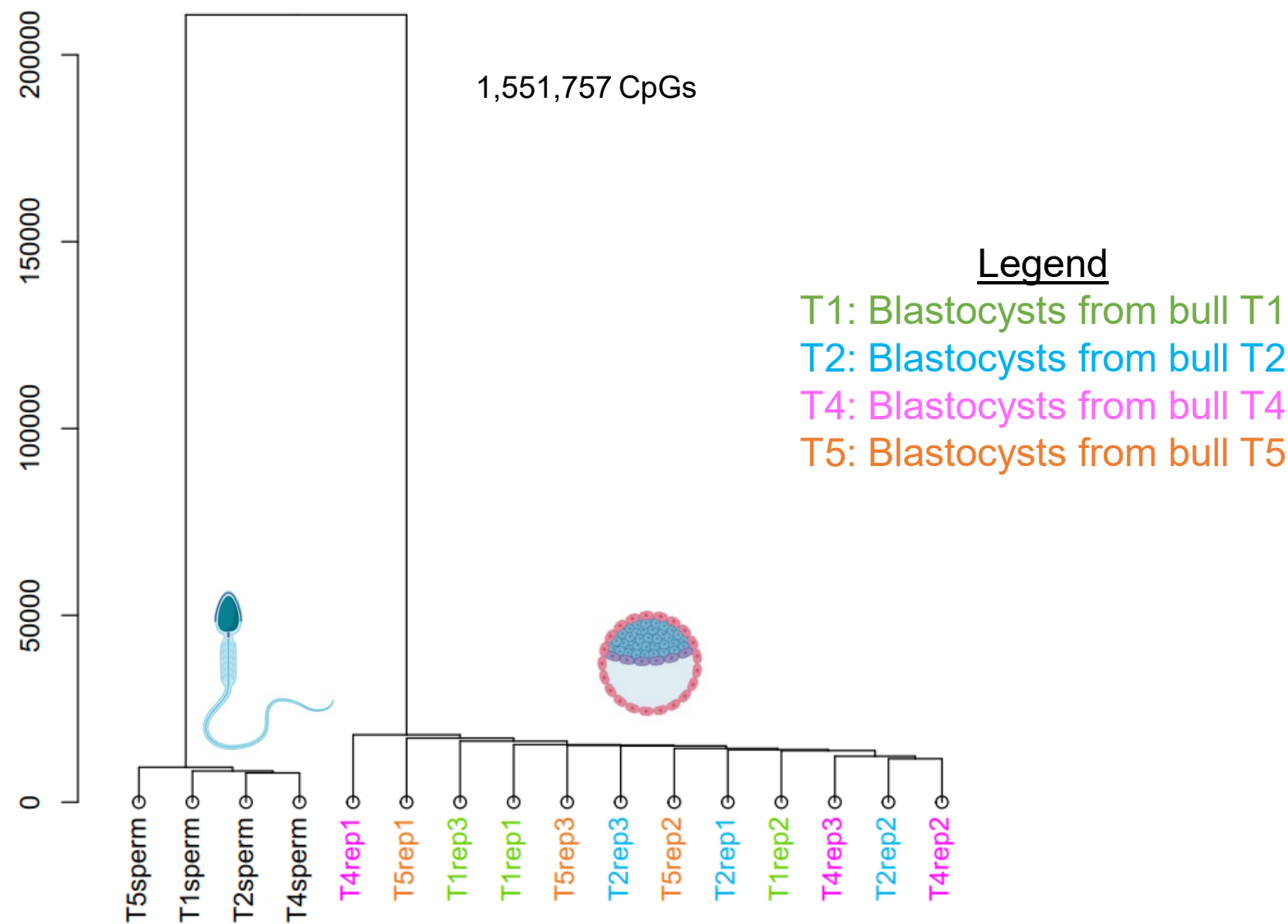


Context	Question	Experimental design	Results	Conclusion / Prospects
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Context	Question	Experimental design	Results	Conclusion / Prospects
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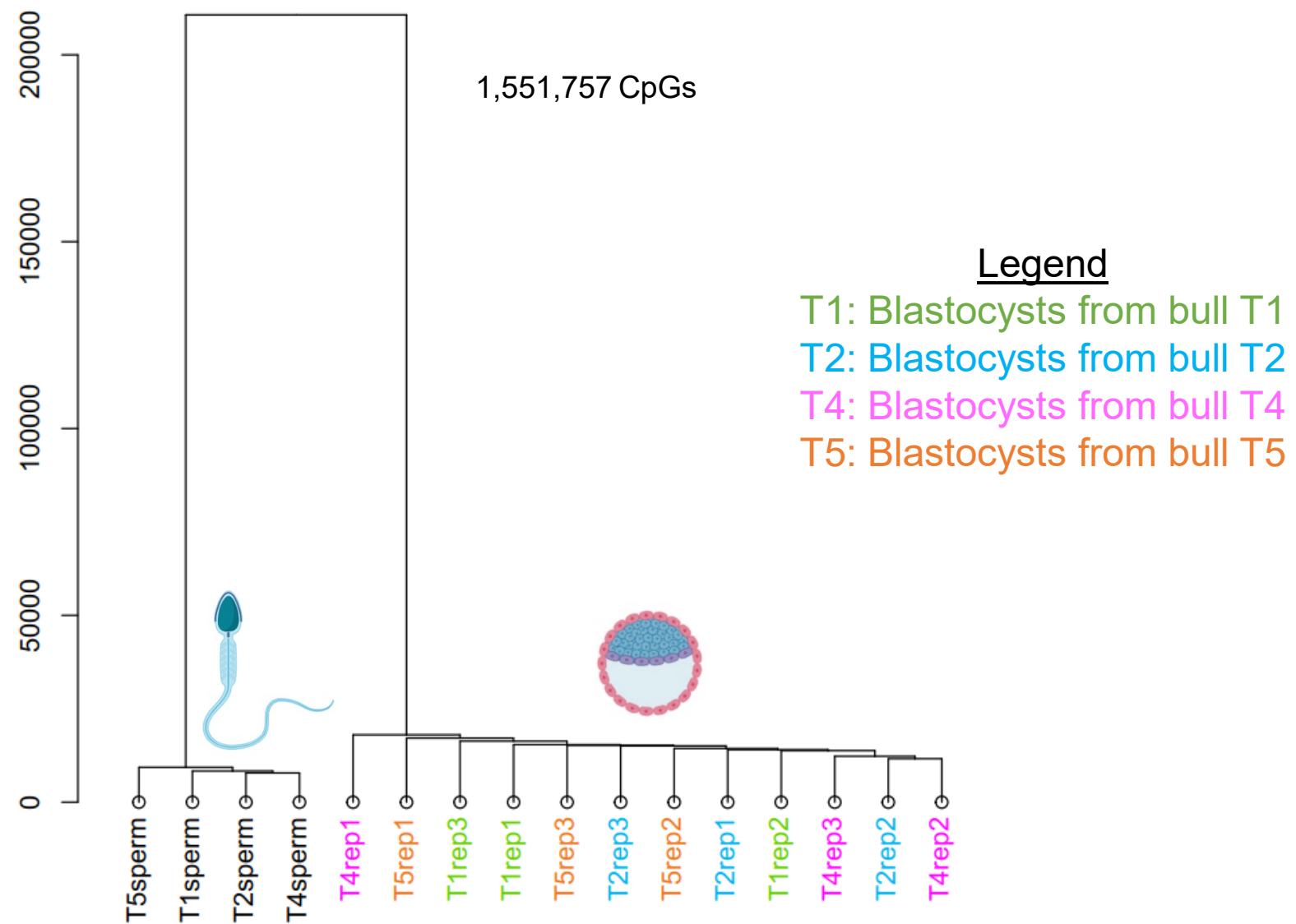
Segregation between sperm and blastocysts by unsupervised hierarchical clustering



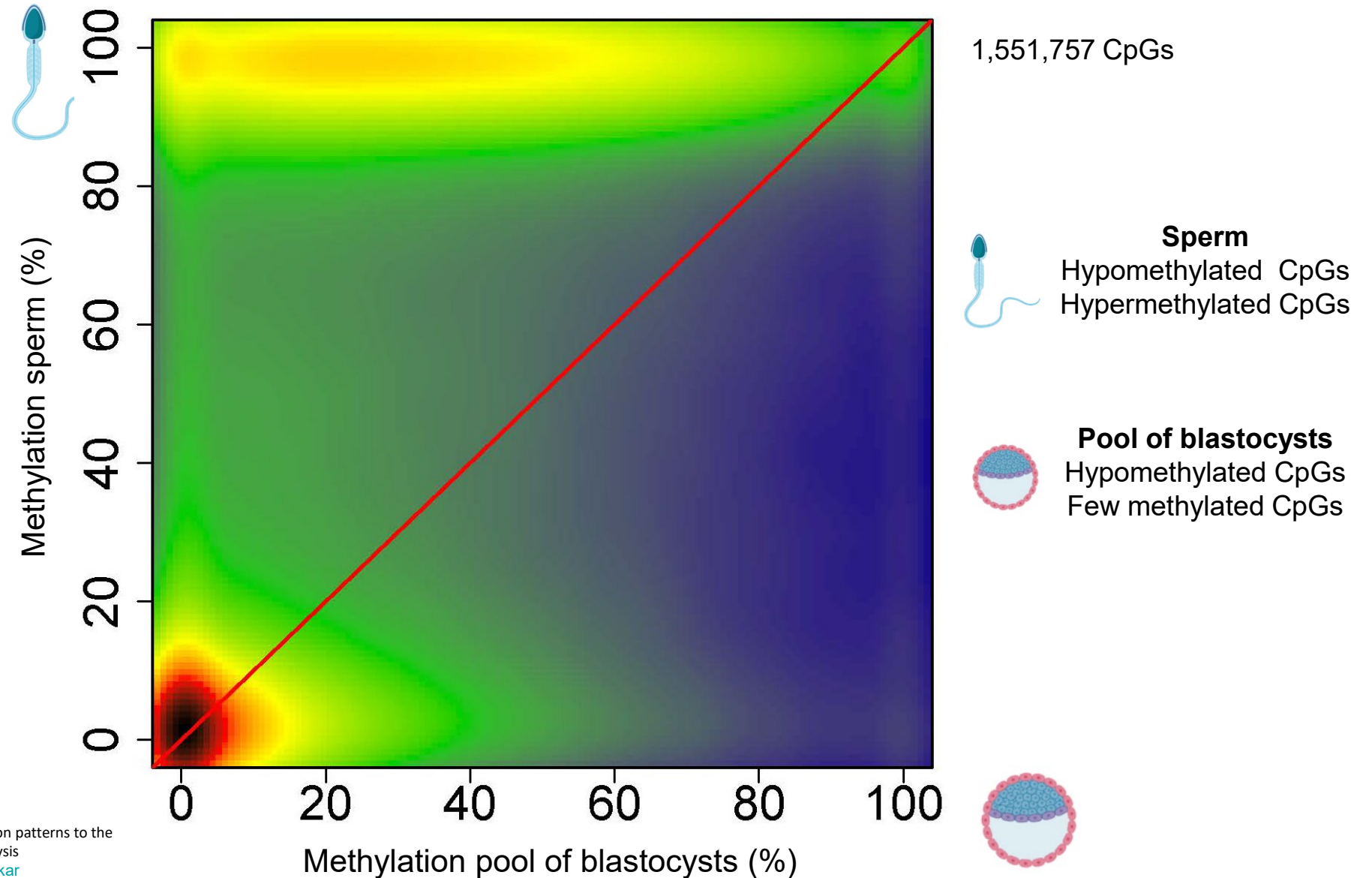
INRAE

Context	Question	Experimental design	Results	Conclusion / Prospects
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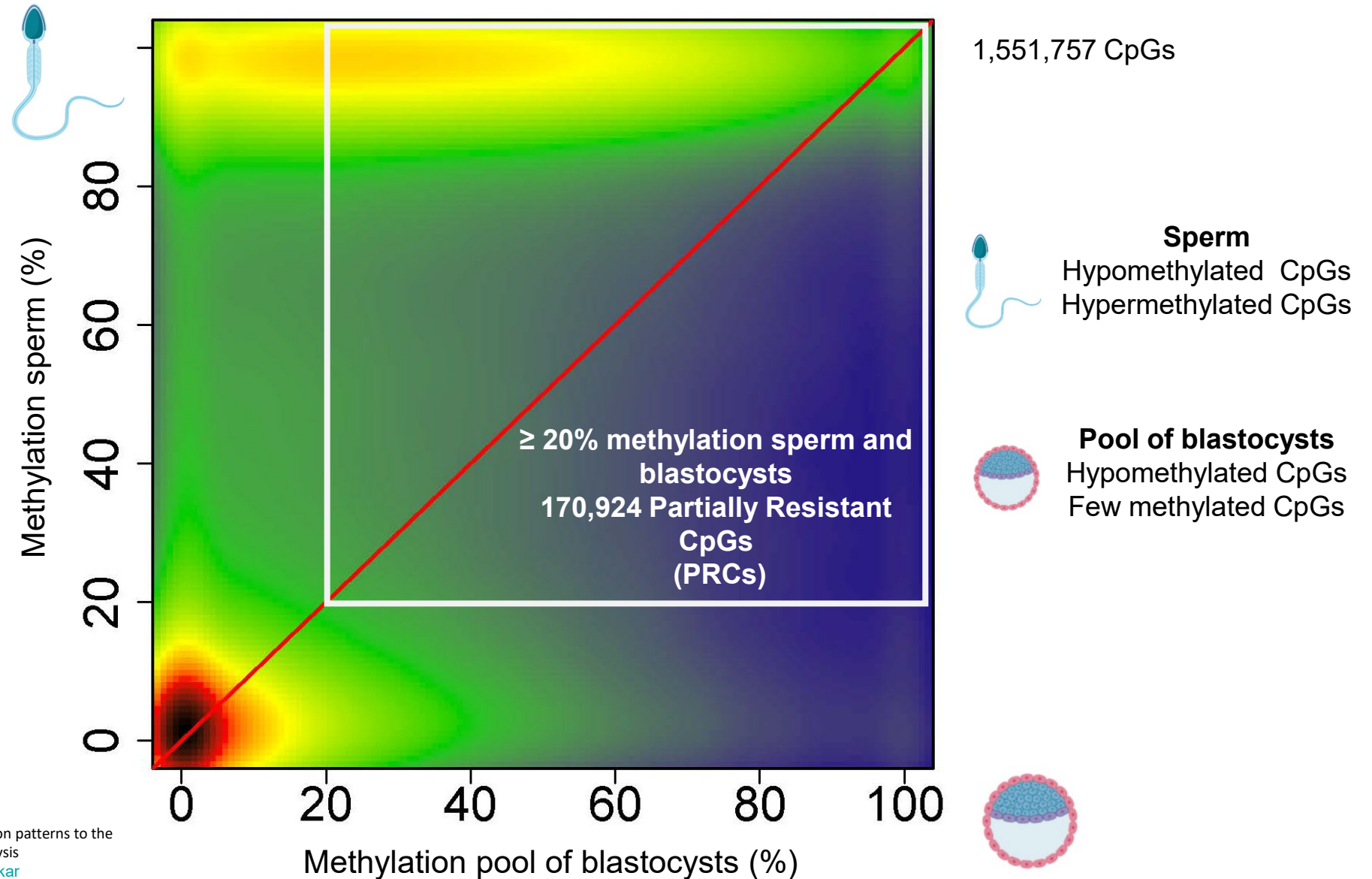
No segregation between blastocysts according to the bull by unsupervised hierarchical clustering



Methylation status of CpGs in sperm and blastocysts



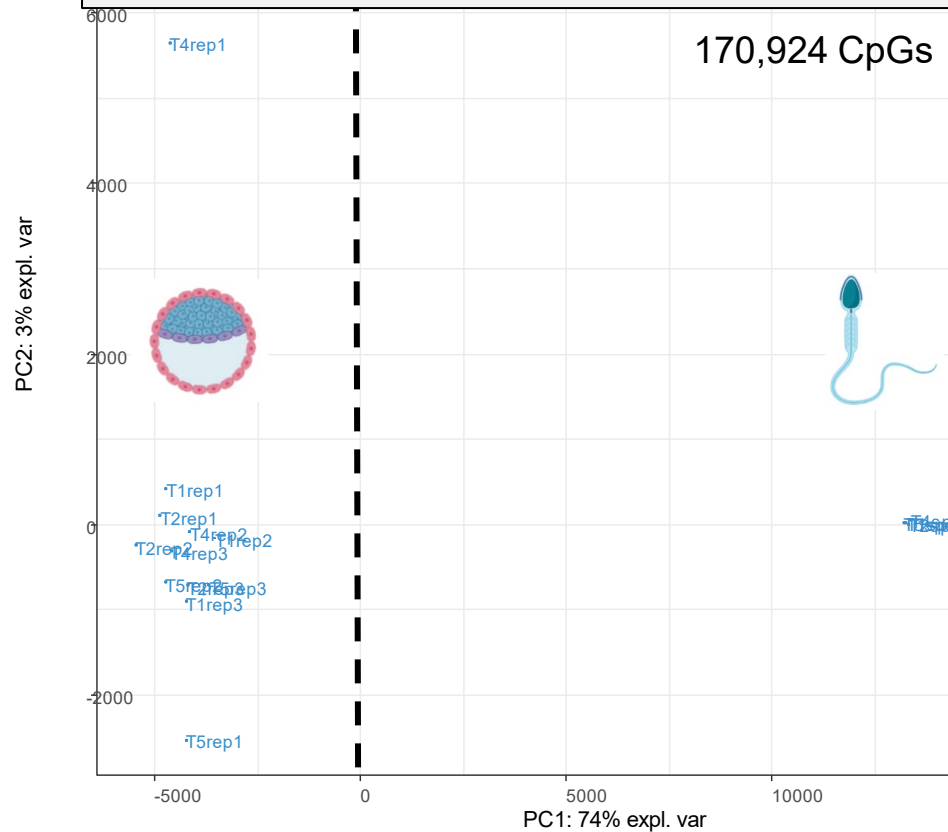
170,924 CpGs remain partially methylated in blastocysts



PRCs' methylation status allows segregation between sperm and blastocysts

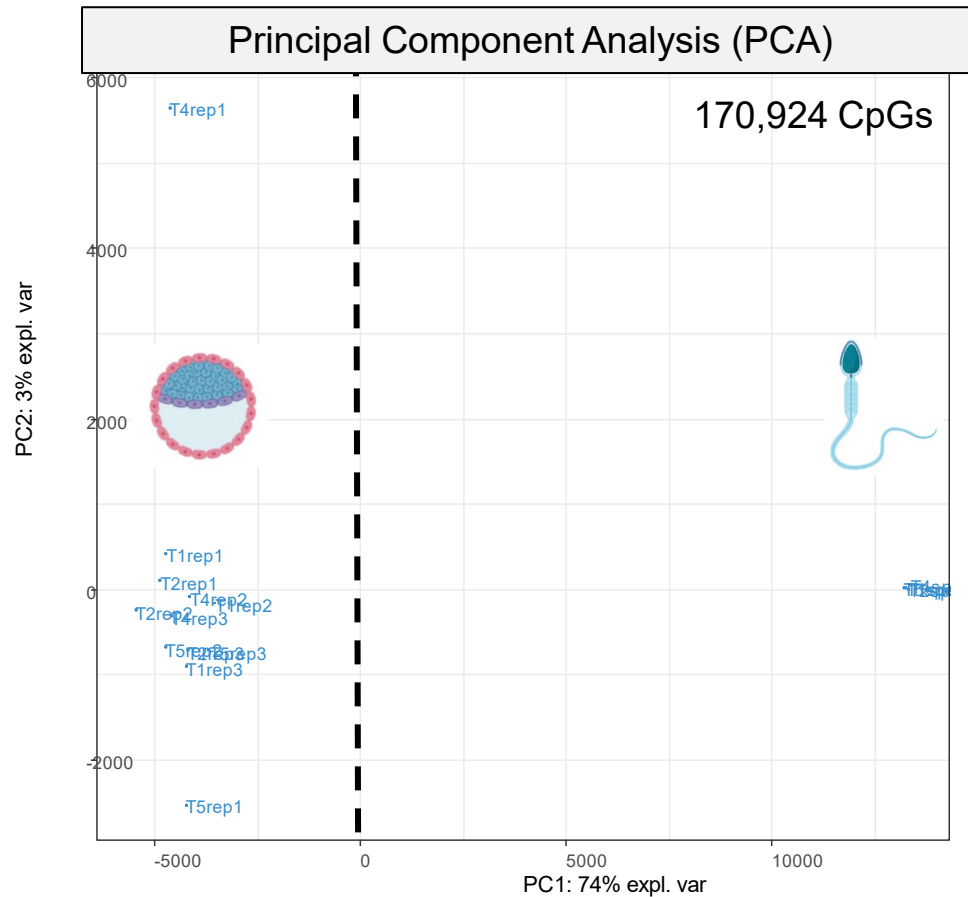
Unsupervised method

Principal Component Analysis (PCA)



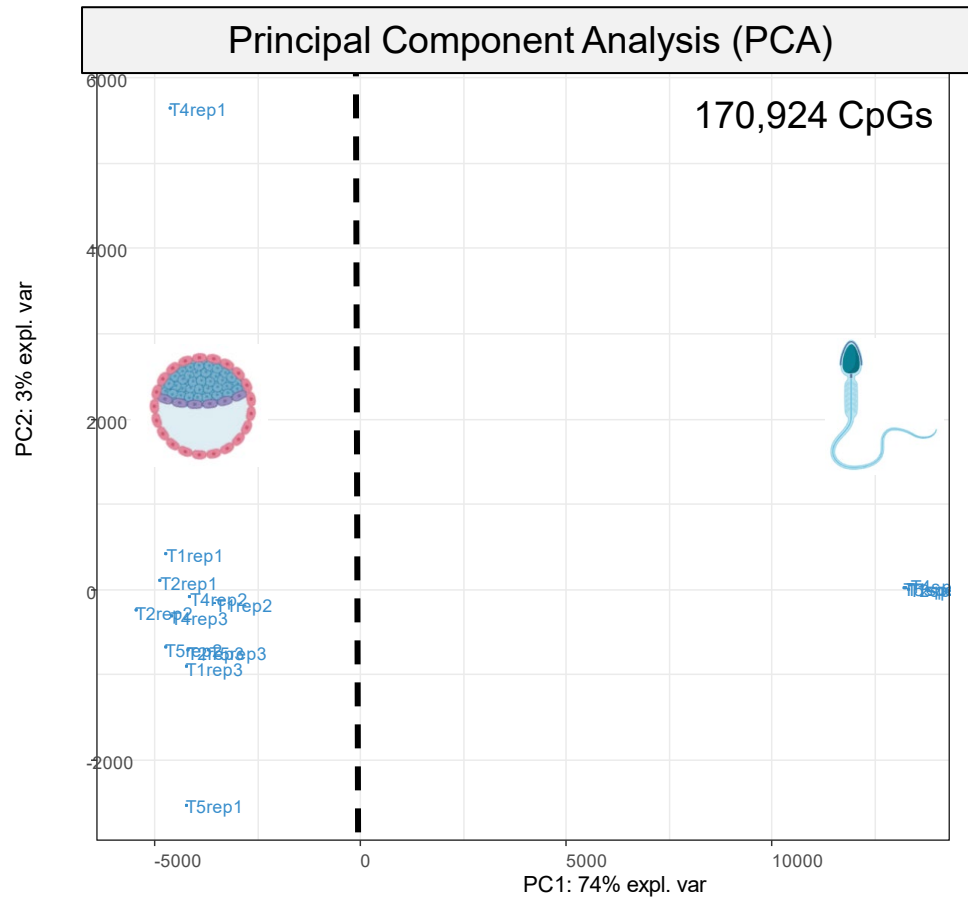
PRCs' methylation status doesn't allow segregation between blastocysts according to the bull

Unsupervised method



Switch to a supervised method: Partial Least Square –Discriminant Analysis on sperm and blastocysts according to the bull

Unsupervised method



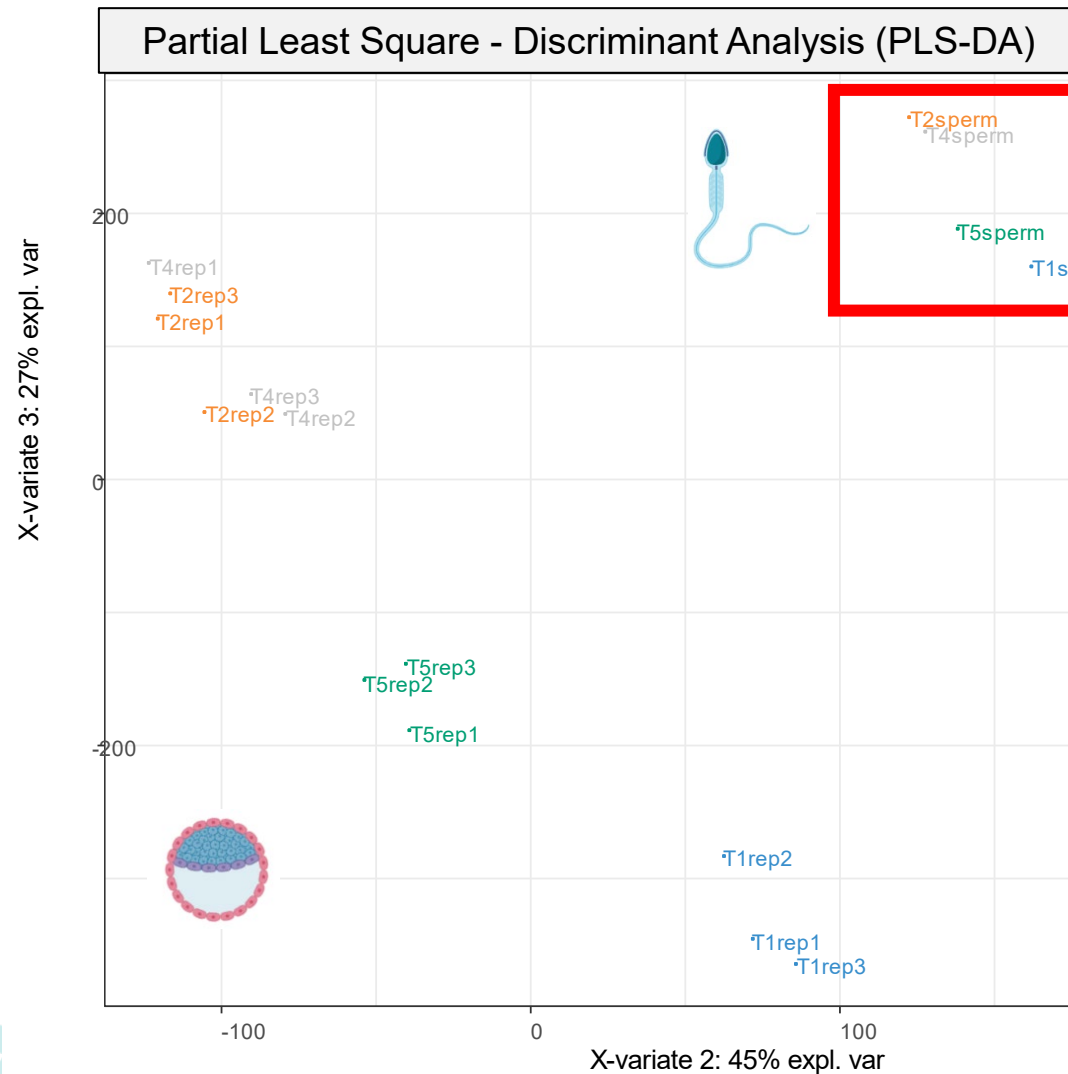
Supervised method

Partial Least Square - Discriminant Analysis (PLS-DA)

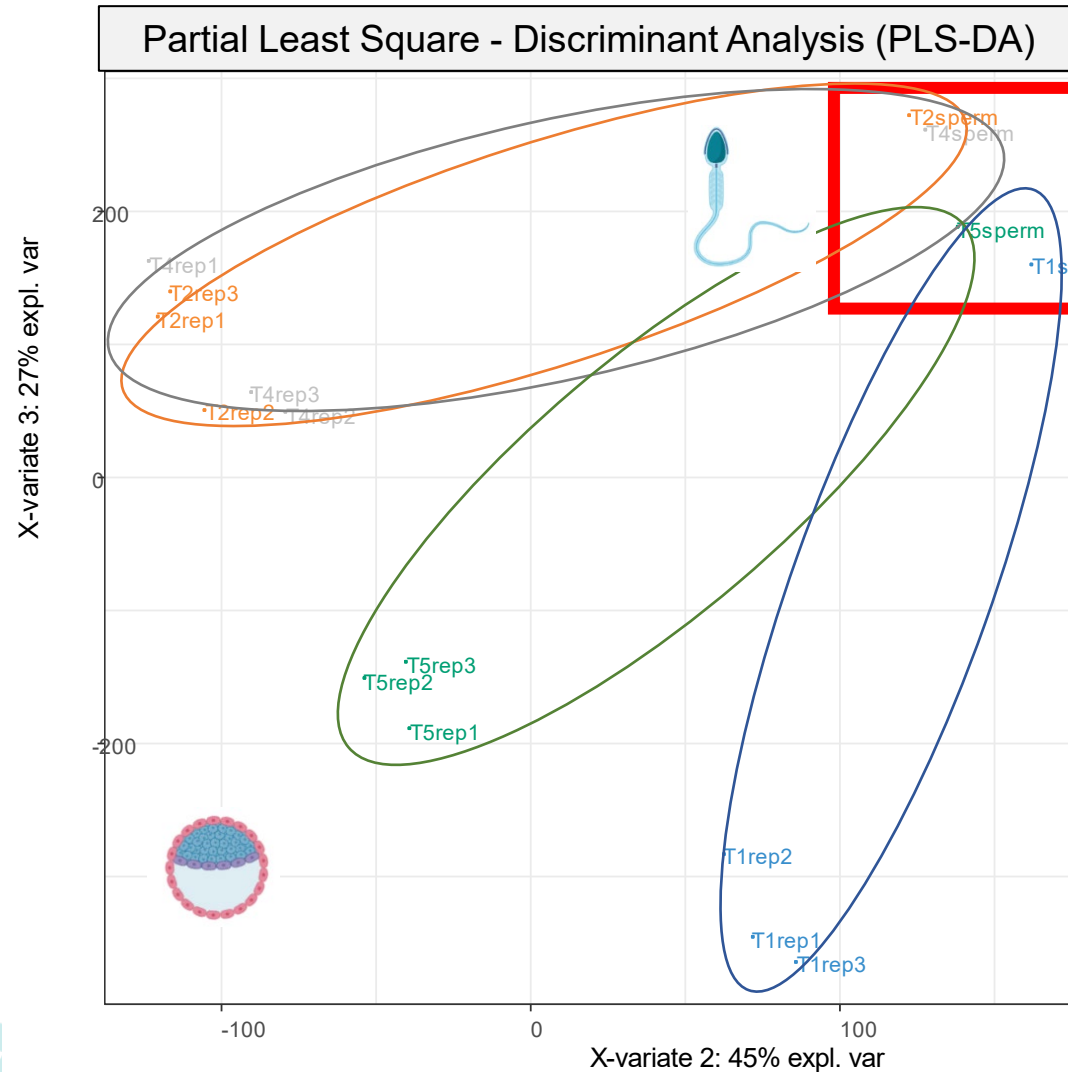
→ Impose a point of view to cluster blastocysts according to the bull

Numeric data: 170,924 CpGs
Categorical dataset: bull

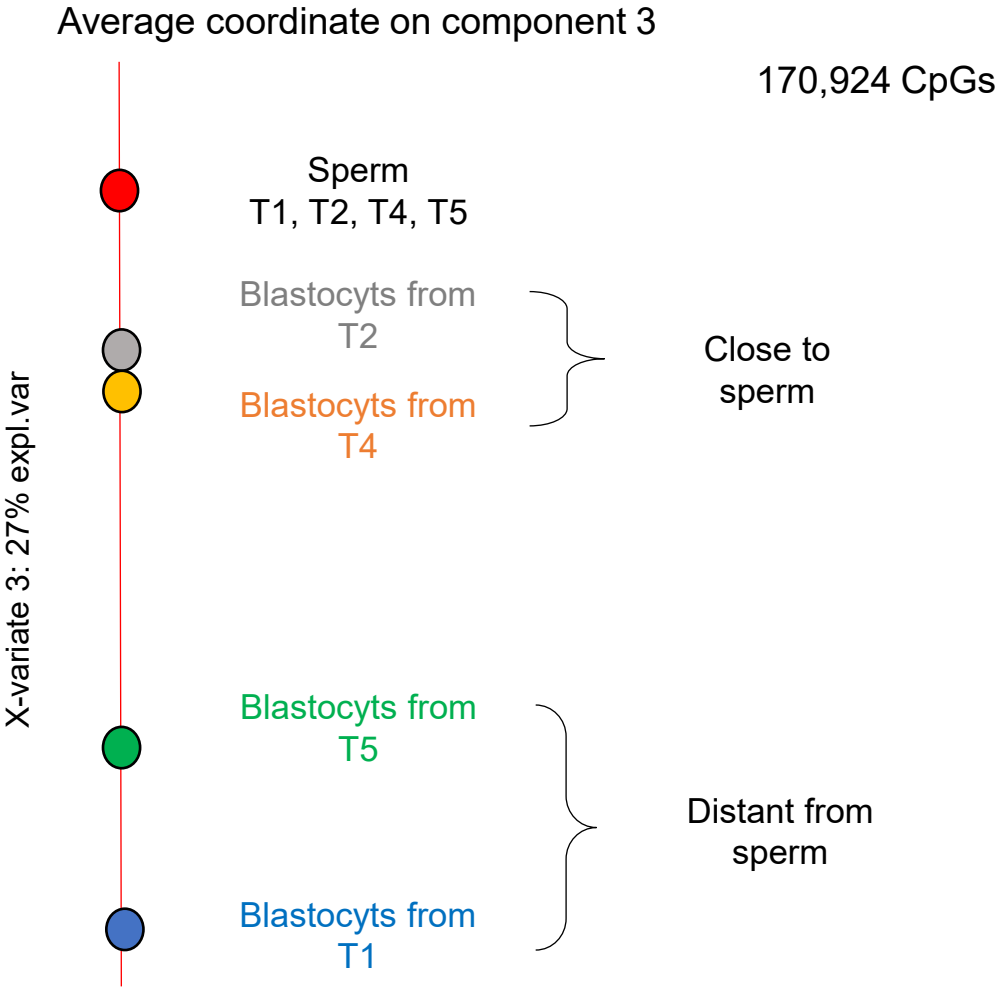
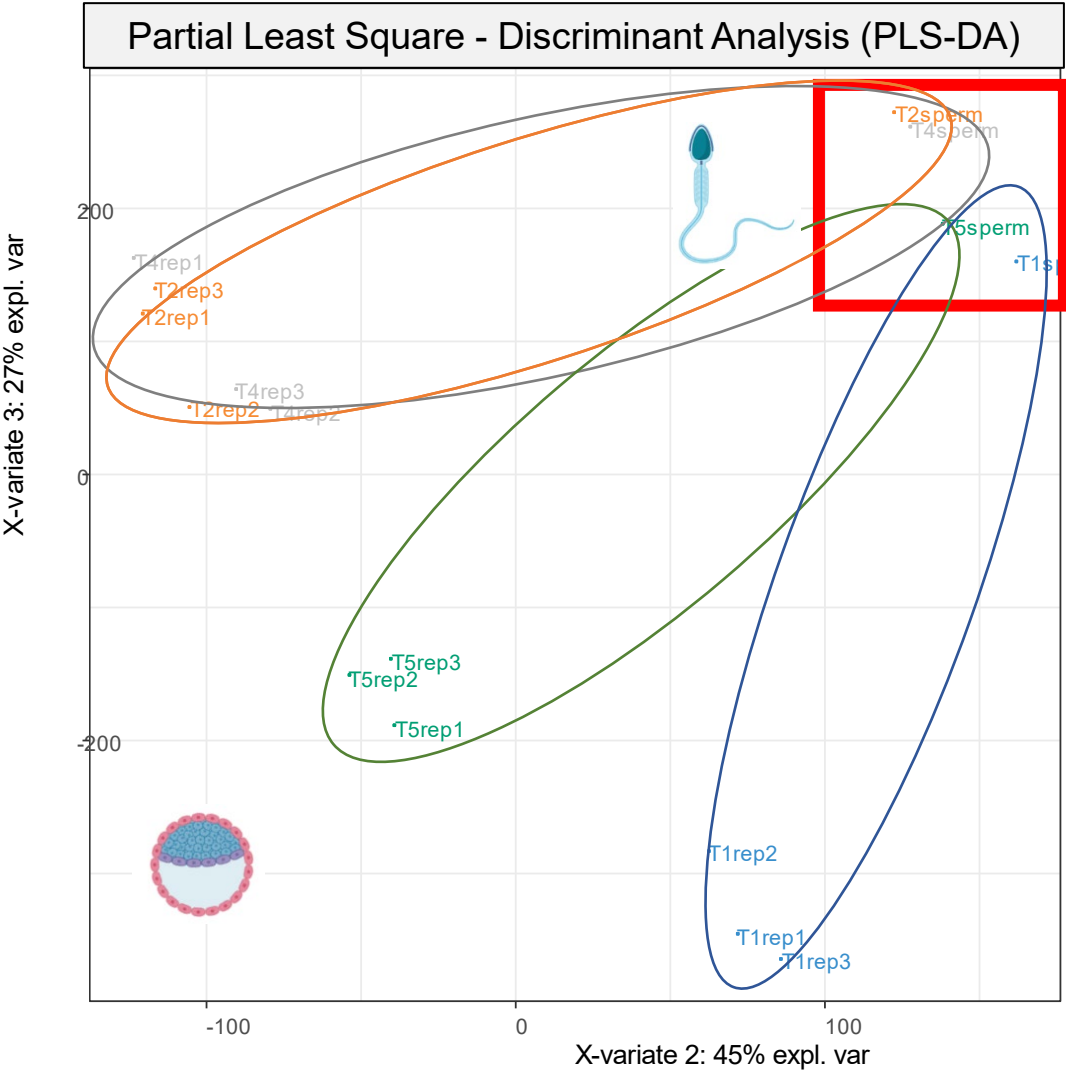
PRCs' methylation status allows segregation between sperm and blastocysts



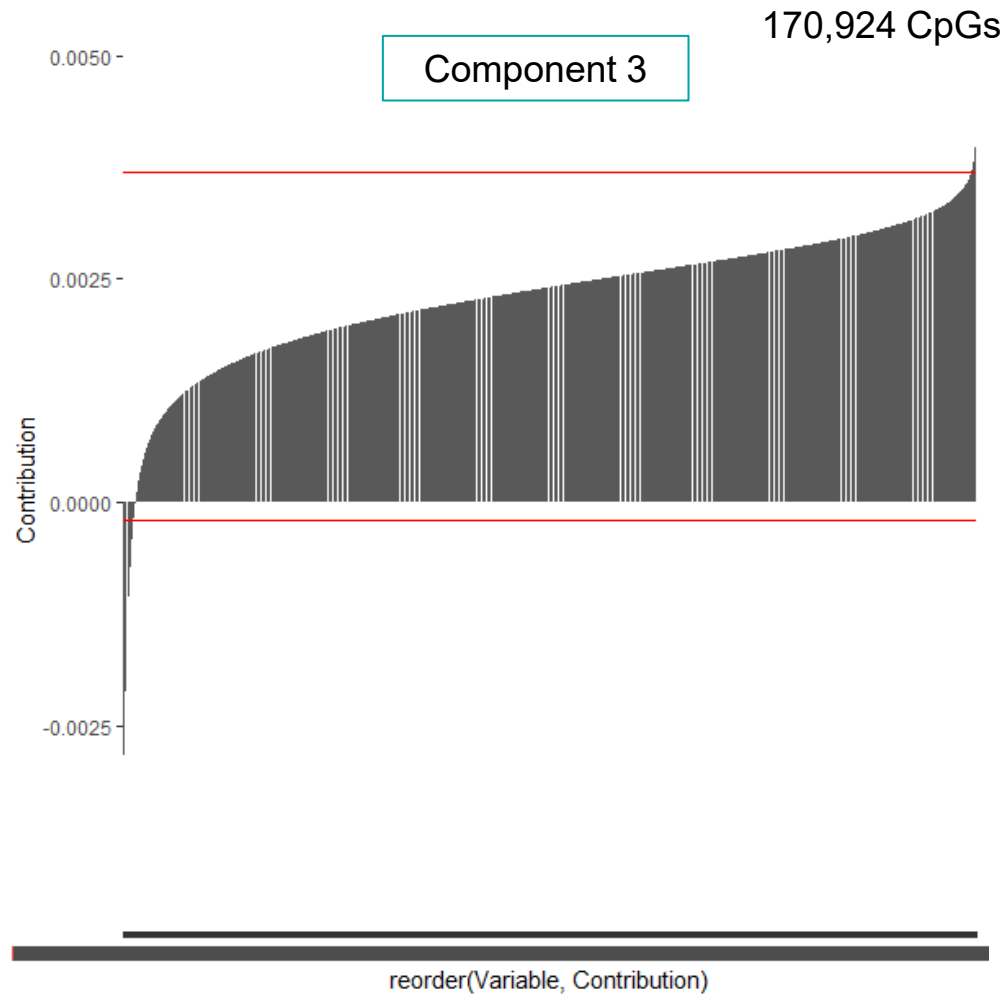
PRCs' methylation is divergent in blastocysts from bulls T5, T1



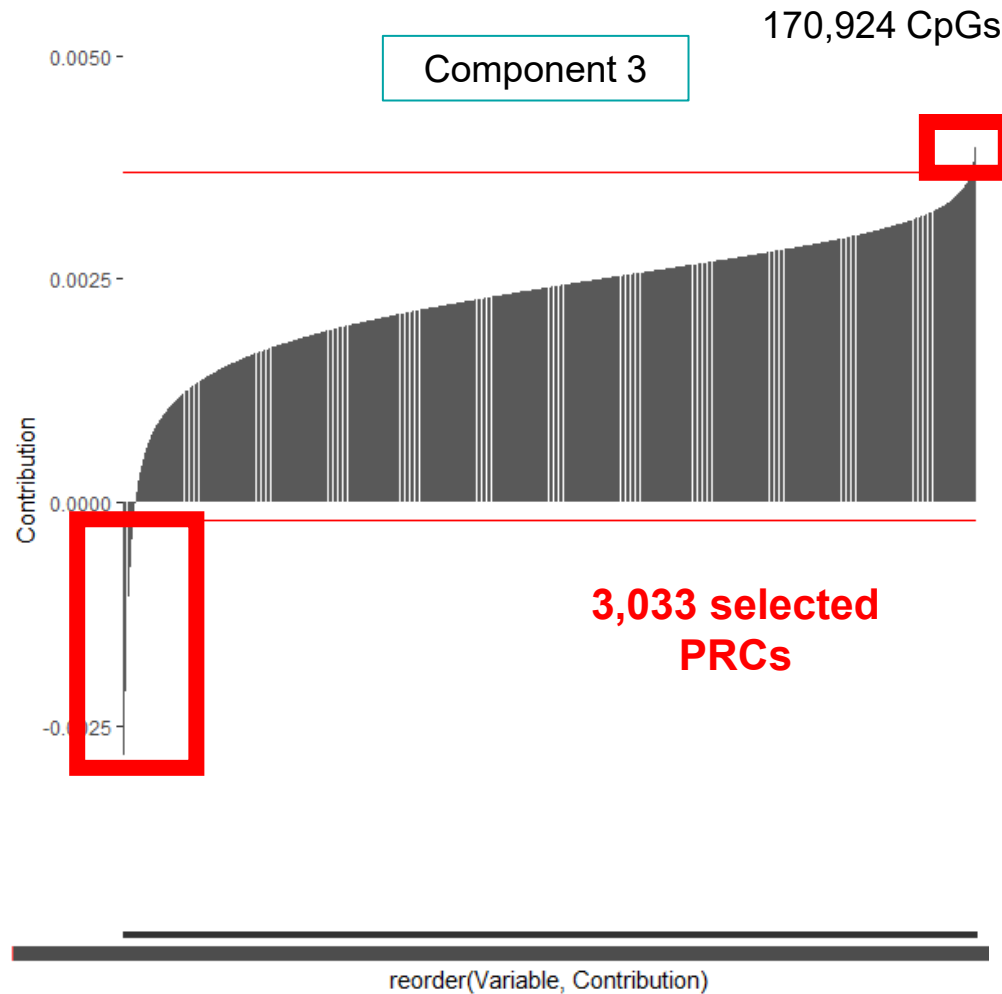
PRCs differentially reprogrammed in blastocysts according to the bull



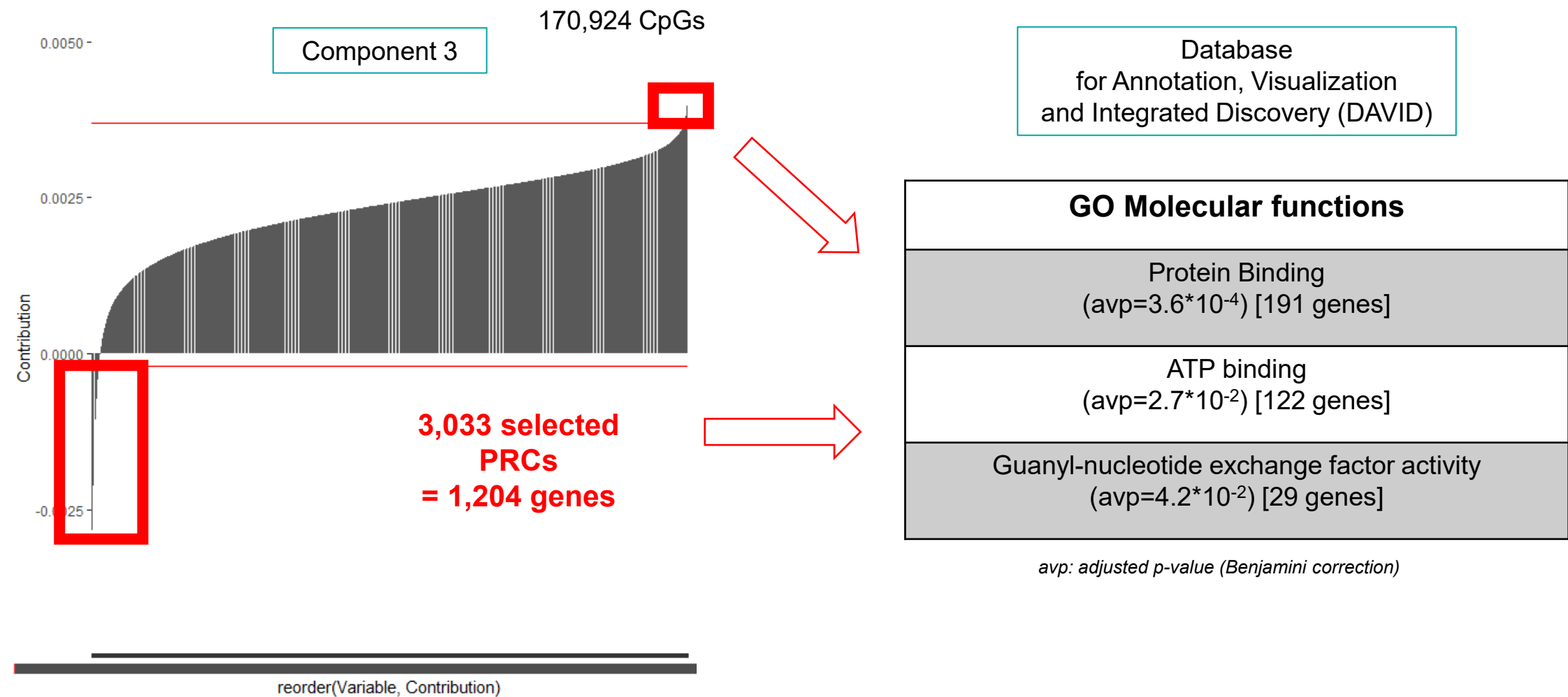
PRCs' contribution to segregate blastocysts according to the bull



Selection of 3,033 PRCs contributing to segregate blastocysts according to the bull



The 3,033 selected PRCs contribute to several molecular functions



Conclusion

- Identification of 170,924 Partially Resistant CpGs (PRCs)
 - ➔ may escape epigenetic reprogramming
- Blastocysts from bull T5, T1 have divergent methylation patterns at 3,033 PRCs compared to T2 and T4
 - ➔ Interindividual variability between bulls in the ability to be reprogrammed after fertilization
- 3,033 PRCs are enriched in molecular functions

Prospects

- As genetic contribute to interindividual variation
- What are the molecular mechanisms underlying interindividual variability between bulls? (genetics?)
- Do PRCs have an impact on the offspring's methylation status and phenotype?

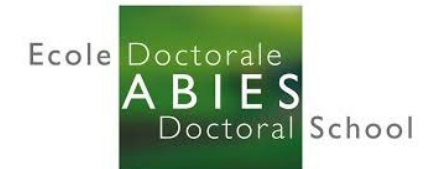
These 3,033 PRCs are present in the RUMIGEN Epichip array



Acknowledgment

Team members

Hélène Kiefer
 Aurélie Chaulot-Talmon
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 Hélène Jammes
 Véronique Duranthon



Thank you for your attention

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Transmission of sperm DNA methylation patterns to the
embryo in cattle: a genome-wide analysis
03/09/2024/Amrita Raja Ravi Shankar