Agriculture et Agroalimentaire Canada

Agriculture and Agri-Food Canada

Effect of epigenetic modifications on immune cells functions

P. Lacasse*, C. Ster, N. Vanacker, and A. Roostaee

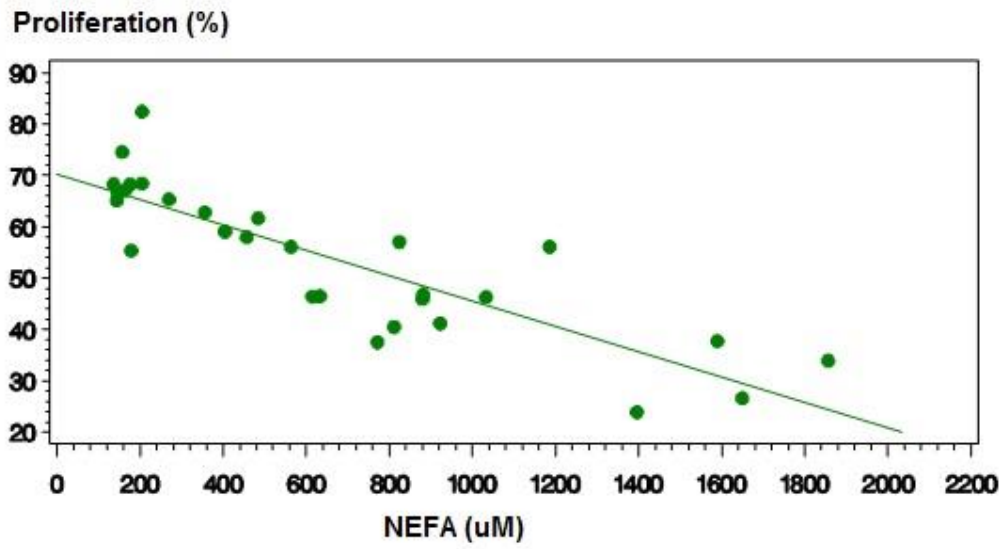
Agriculture and Agri-Food Canada
Sherbrooke Research and Development Centre

Canada

Variability of lymphocytes functions

- Lymphoproliferative response varies during the lactation cycle and among animals

- An important source of variation during lactation is the composition of the lymphocyte's environment, notably NEFA concentration



Ster et al., JDS (2012)

Variability of lymphocytes functions

- Nevertheless, lymphocytes from animals at similar stage of lactation, similar nutritional status and without any symptoms of diseases still show differences in functionality
- These differences are maintained when animals are sampled several time
- Genetic variation probably explains some variation, but heritability of resistance to infection (mastitis) is low

→ Epigenetic modifications?

Epigenetic modifications

- Alterations in gene expression
- Independent of DNA sequence
- Changes in chromatin structure
  - Euchromatin: Gene expression
  - Heterochromatin: Gene silencing

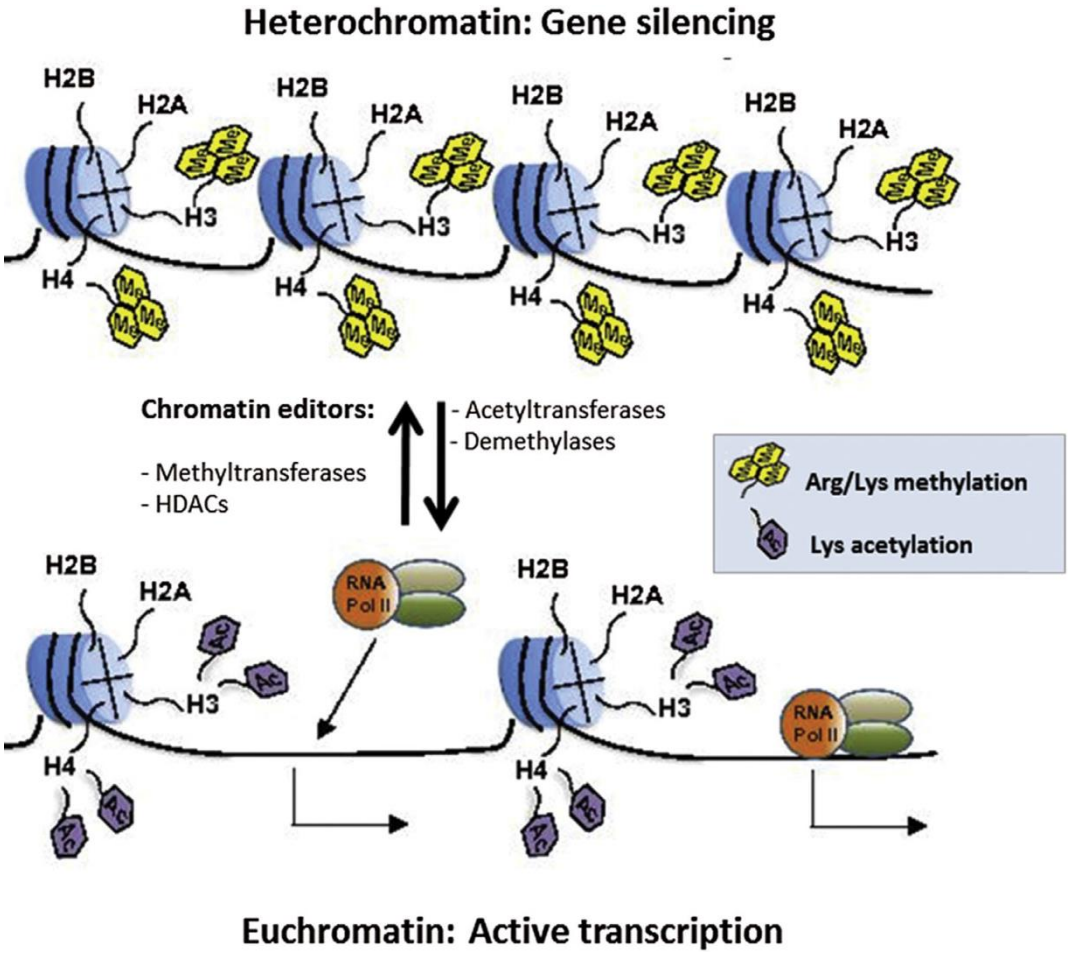
Epigenetic Modifications

Histone **methylation**

Heterochromatin
(Gene **silencing**)

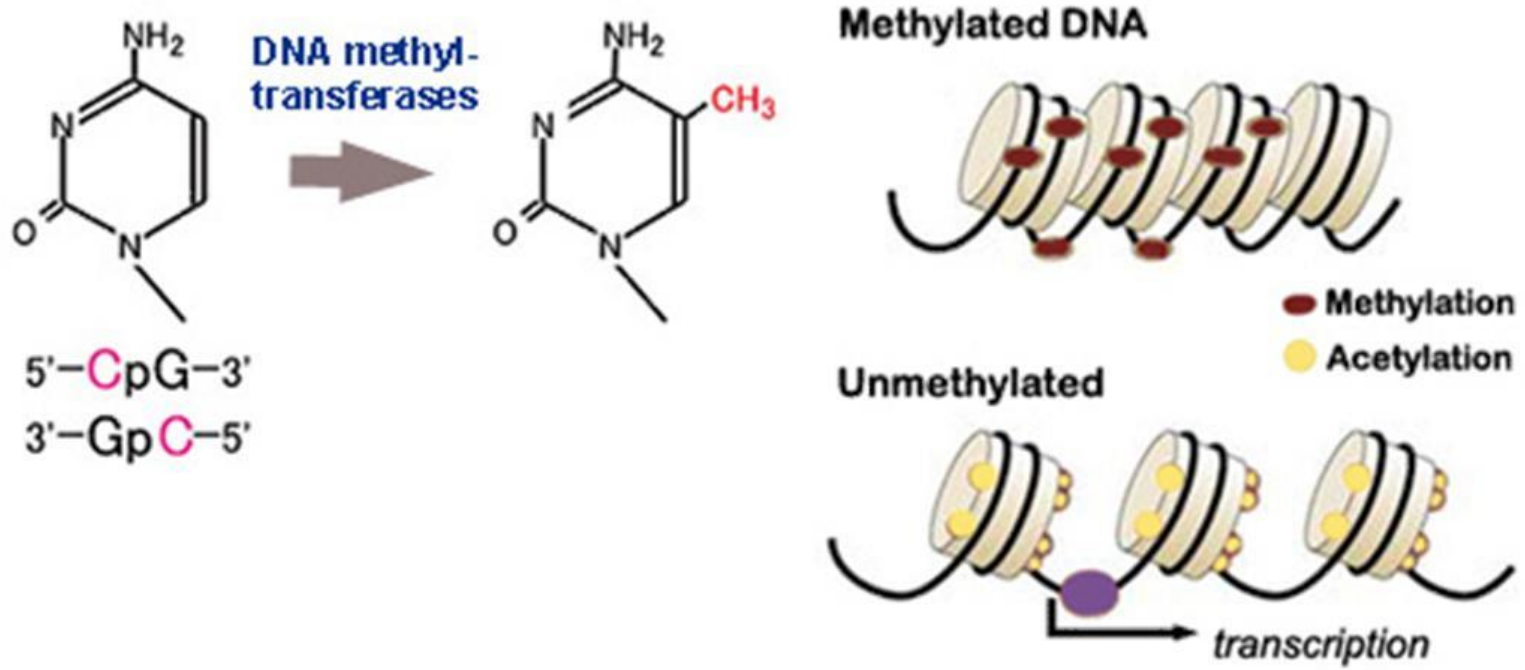
Histone **acetylation**

Euchromatin
(Gene **expression**)



Roostaee et al. 2016

DNA methylation



Epigenetic agents

- Histone deacetylase inhibitor

↑Histone acetylation ⟶ ↑Gene expression

- Histone methylation inhibitor

↓Histone methylation ⟶ ↑Gene expression

- DNA methylation inhibitor

↓DNA methylation ⟶ ↑Gene expression

Epigenetic contribution to individual variation in response to LPS in bovine dermal fibroblasts

Parameters : IL-8, IL-6, TNF-α

High responding → Histone acetylation agent / DNA demethylation agent → **High responding**

Low responding → Histone acetylation agent / DNA demethylation agent → **High responding**

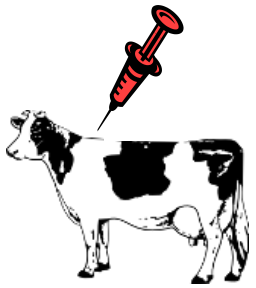
Green *et al.* Vet Immunol Immunopathol, 2014

Objective

Our objective was to determine if epigenitic modifications can explain some of the variation between cows in their lymphoproliferation capacity

Identification of cows with high and low lymphoproliferative response

40 multiparous Holstein cows in mid-late lactation lactation



Blood was sampled twice (4 weeks apart)

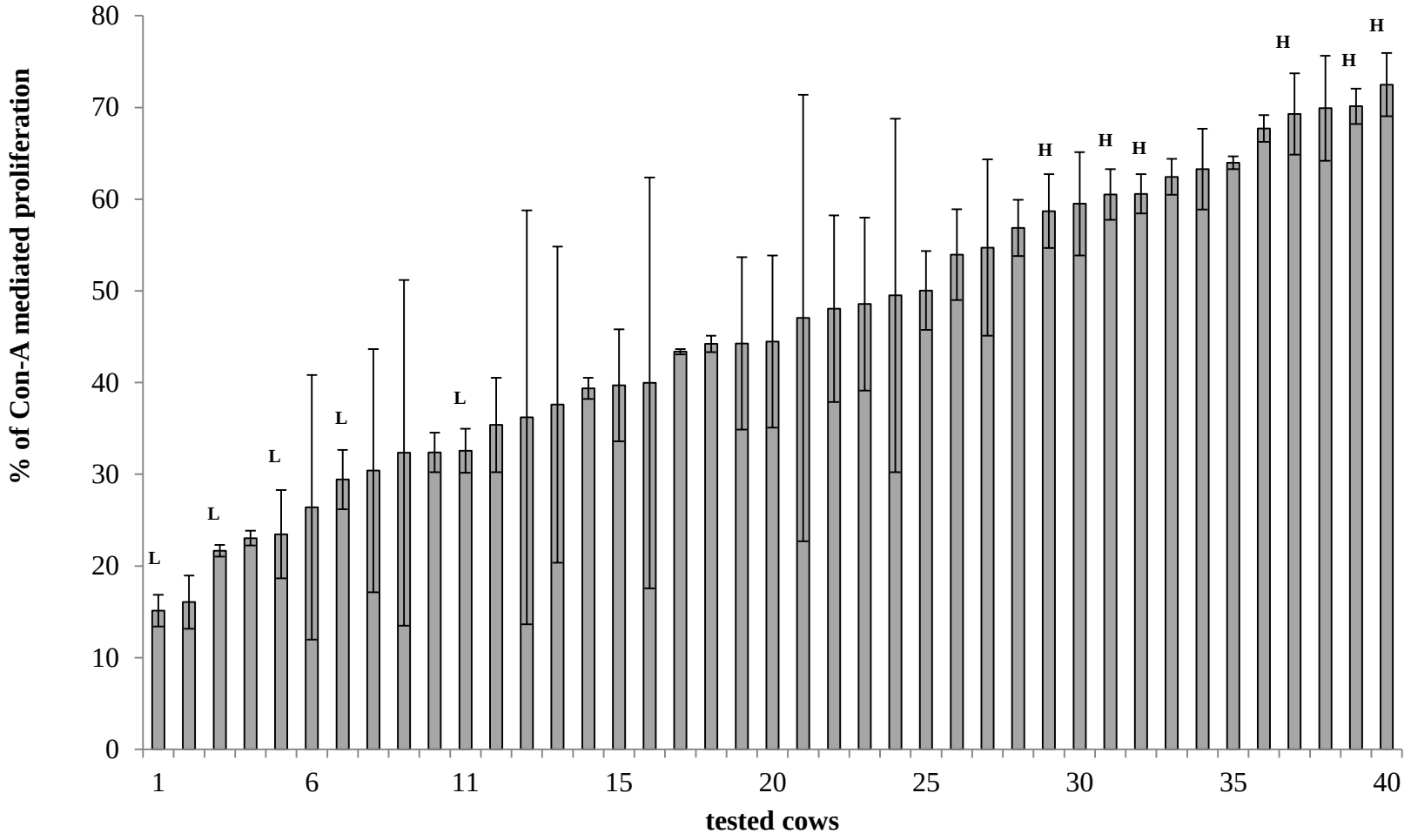
↓

PBMCs were isolated

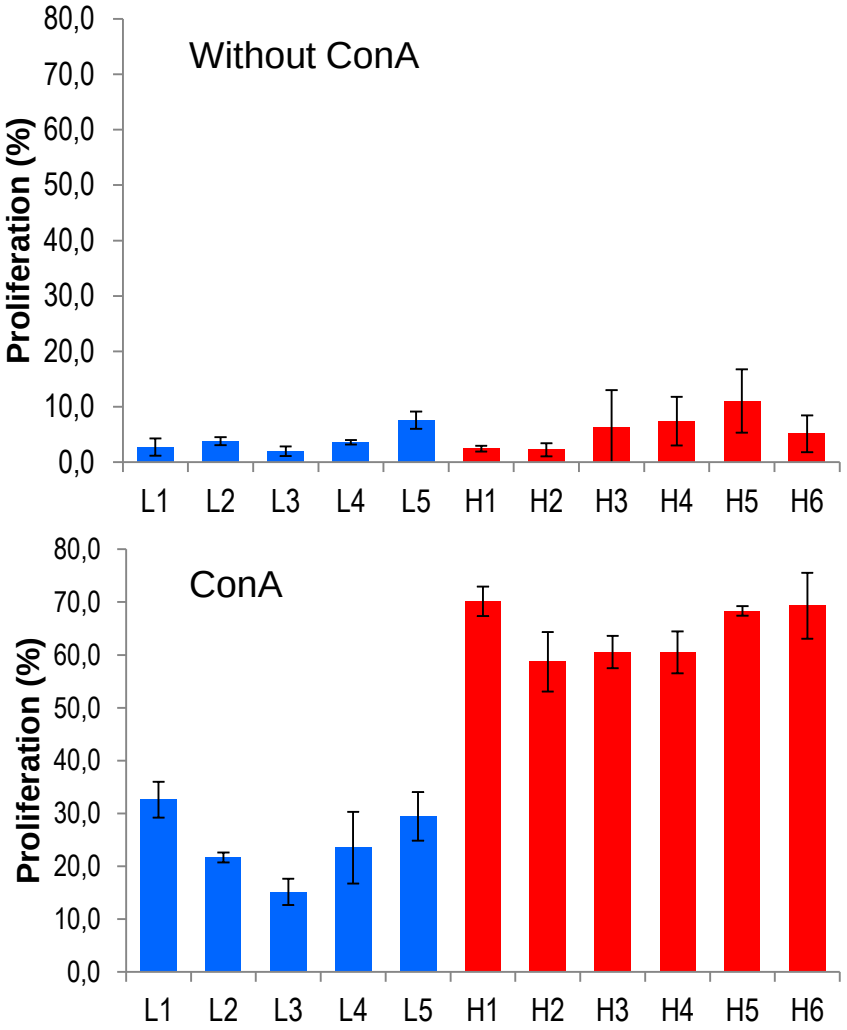
↓

Lymphoproliferative response was evaluated with and without ConA

Identification of cows with high and low lymphoproliferative response



Identification of cows with high and low lymphoproliferative response



Effect of epigenetic agents on lymphoproliferative response

- PBMCs were isolated from 11 cows (6H 5L)

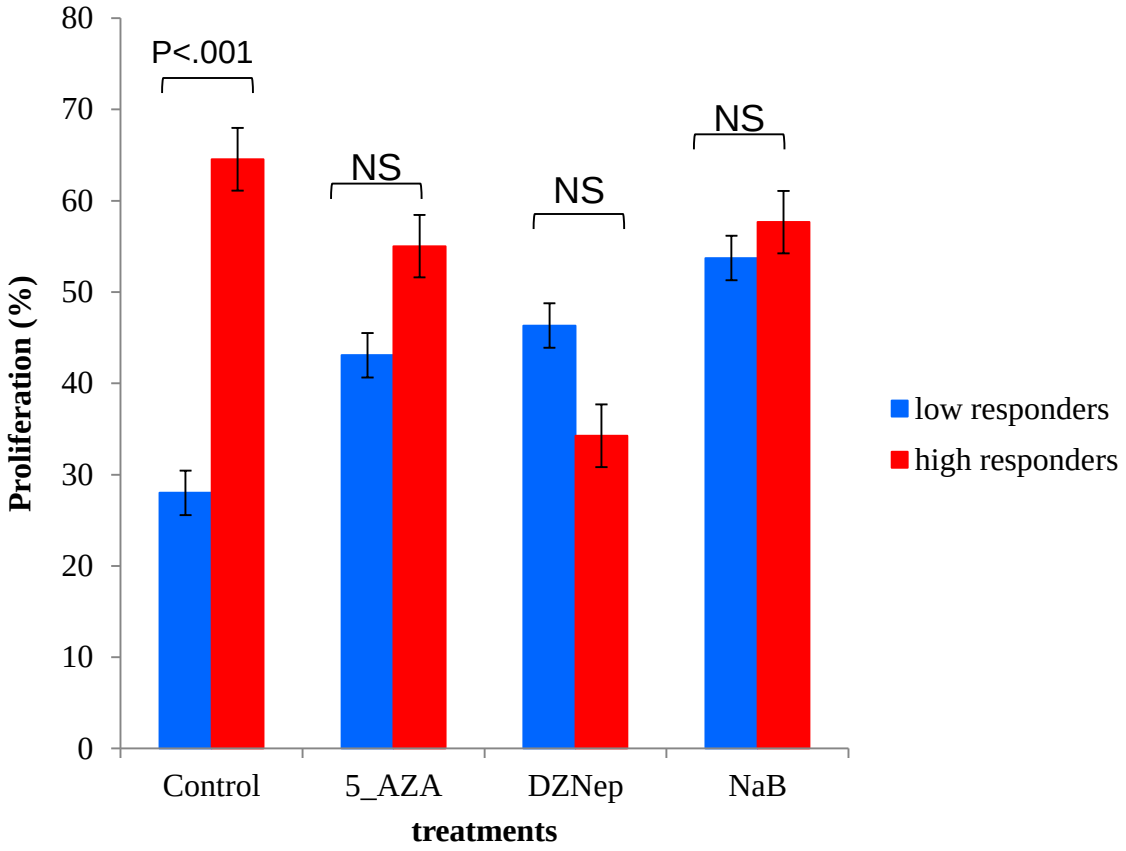
- Lymphoproliferative response was evaluated with and without ConA and the following epigenetic agents :
  - None (control)

  - ***5-aza-2'-deoxycytidine*** **(5-AZA)** a DNA de-methylation agent
    ↓ DNA methylation ⟶ ↗ Gene expression

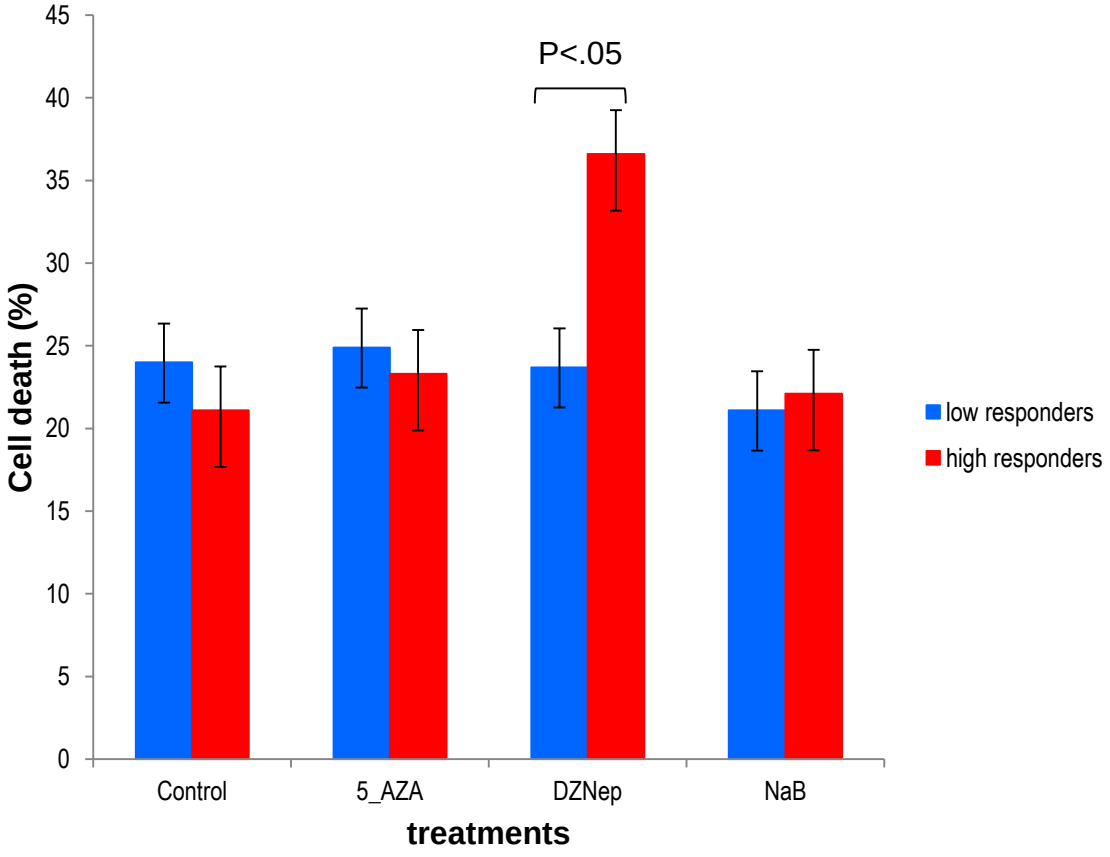
  - ***Sodium Butyrate*** **(NaB)** a histone deacetylase inhibitor
    ↑ Histone acetylation ⟶ ↗ Gene expression

  - ***3-Deazaneplanocin*** **(DZNep)** a histone methylation inhibitor
    ↓ Histone methylation ⟶ ↗ Gene expression

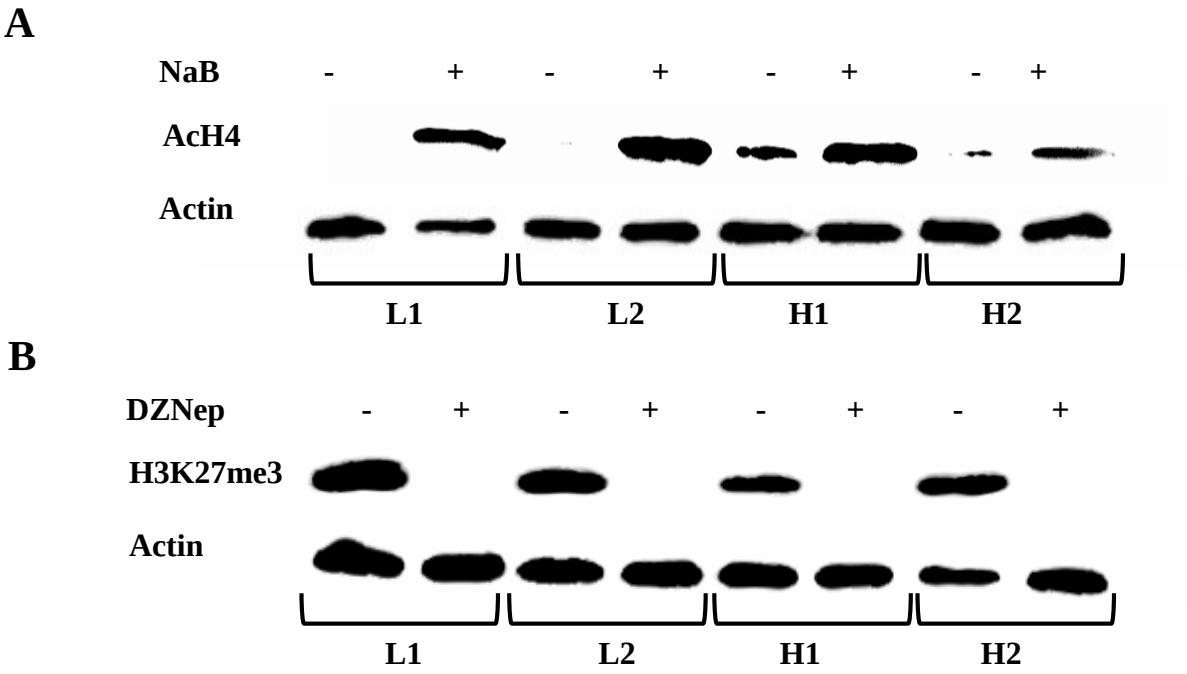
Effect of epigenetic agents on lymphoproliferative response



Effect of epigenetic agents on lymphoproliferative response

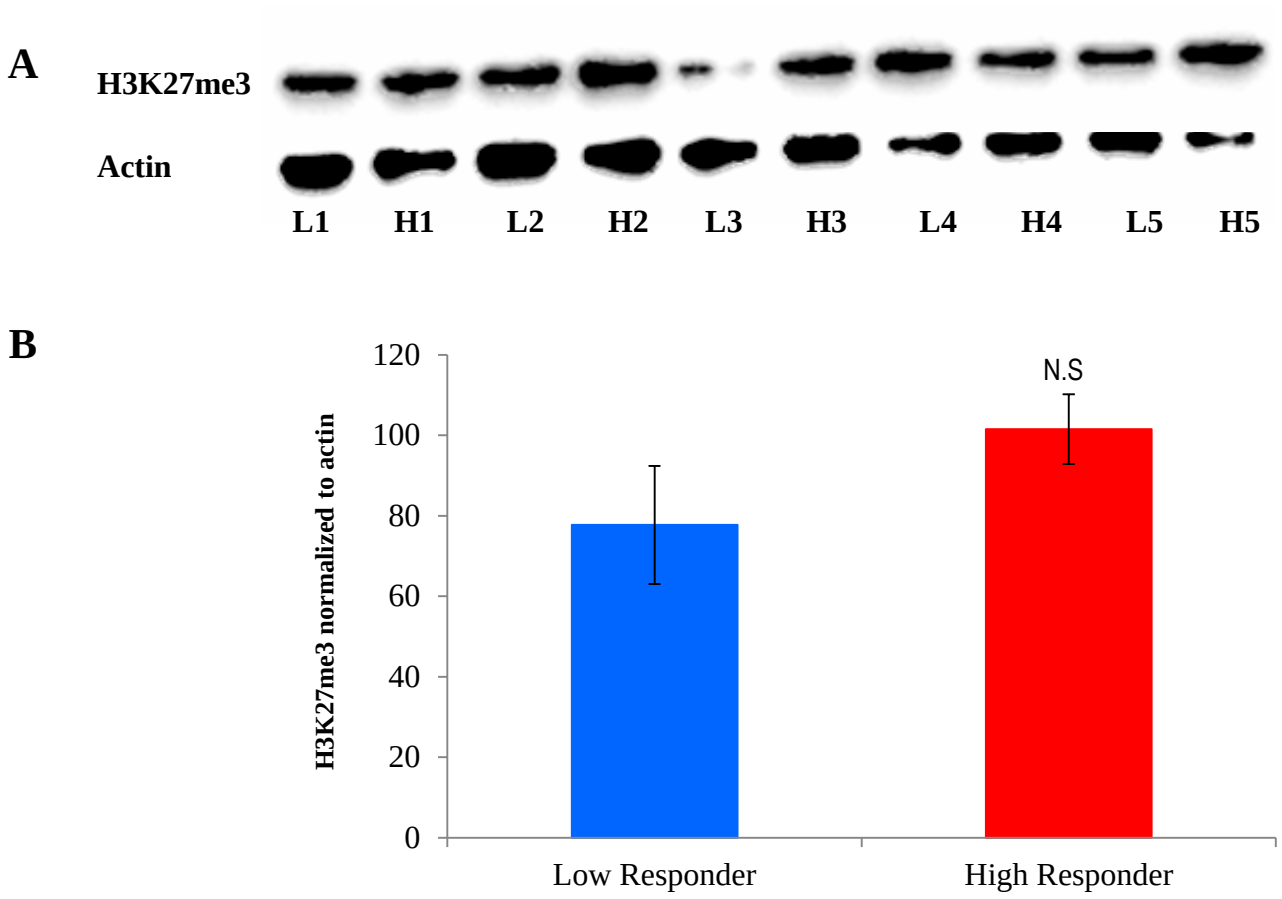


Effect of epigenetic agents on histone methylation and acetylation



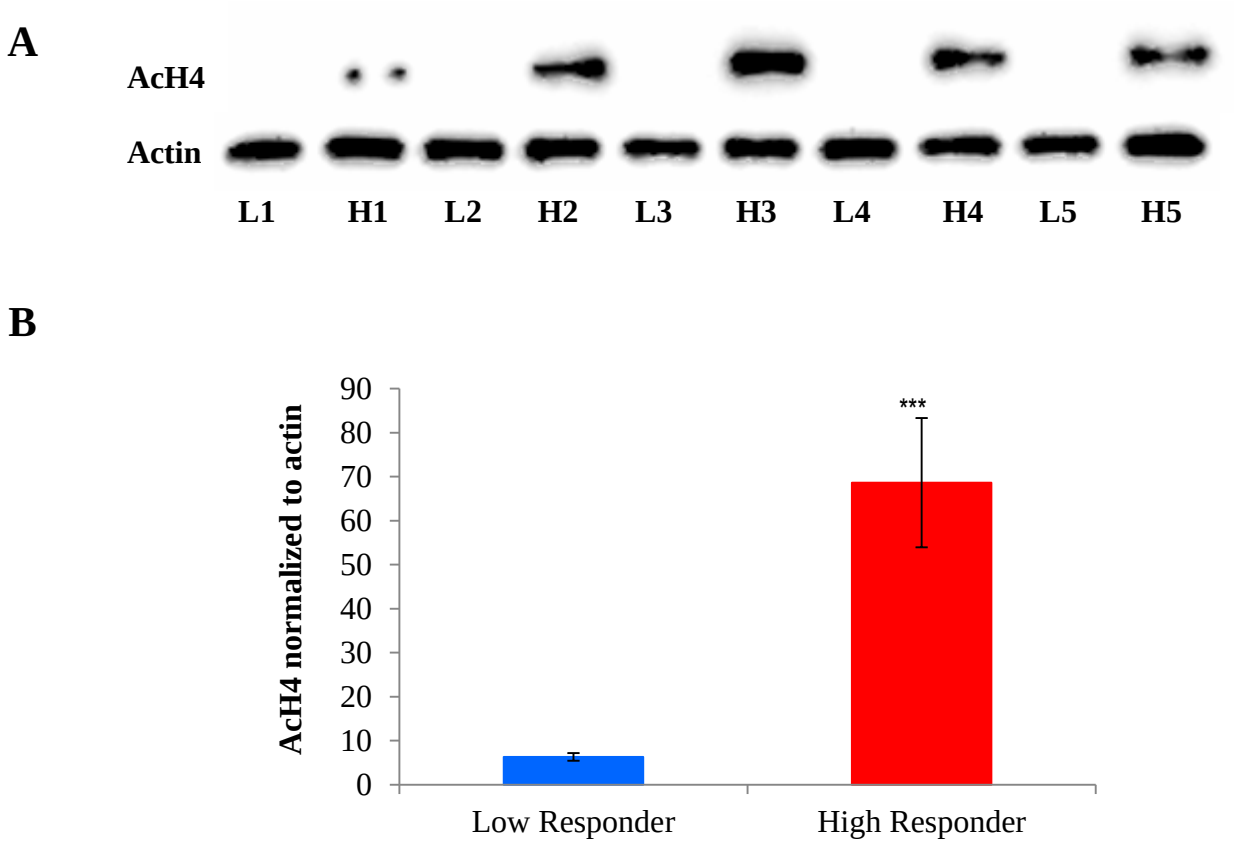
Effect of the different treatments on ConA stimulated lymphocytes on methylation of histone H3 (A), acetylation of Histone H4 (B) for low (L, n=5) and high (H, n=6) responders

Histone methylation in high and low responders cows



Methylated of histone H3 (H3K27me3) in ConA-stimulated lymphocytes of low (n=5) and high (n=5) responders (A) Westen blot. (B) quantification of the westernblots.

Histone acetylation in high and low responders cows



Acetylation of histone H4 (AcH4) in ConA-stimulated lymphocytes of low (n=5) and high (n=5) responders (A) Western blot. (B) quantification of the western blot.

Conclusions

- The results of this experiments support the hypothesis that epigenetic modifications significantly alter lymphocyte response in dairy cows

- A better understanding of factors controlling epigenetic modifications may help to raise dairy cows with a high level of immunity

Special thanks to

The authors would like to thank Catherine Thibault and Frédéric Beaudoin for providing excellent technical assistance.