

# Evaluating the role of injectable trace minerals in modulating cytokine responses to bovine respiratory vaccination in dairy calves

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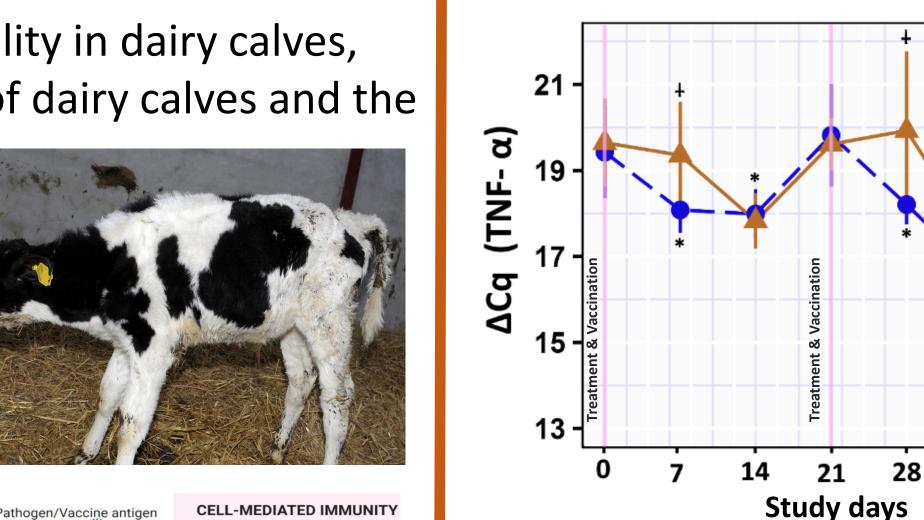
## Introduction

## **Bovine respiratory diseases (BRD)**

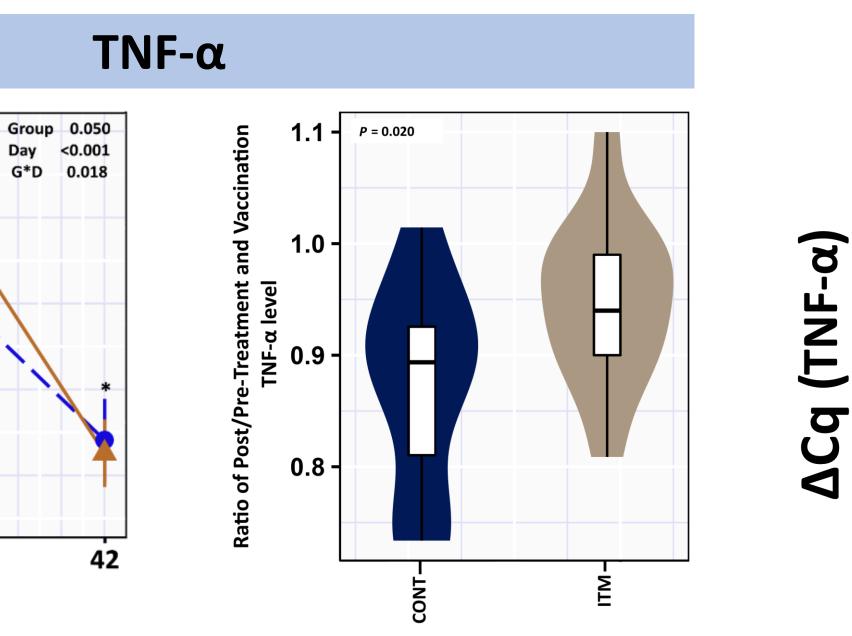
- Bovine respiratory disease (BRD) is considered the most common cause of morbidity & mortality in dairy calves, reflecting negatively on the welfare of dairy calves and the economics of dairies<sup>1,2</sup>
- BRD is responsible for 46.5% of calf deaths after weaning<sup>2</sup>

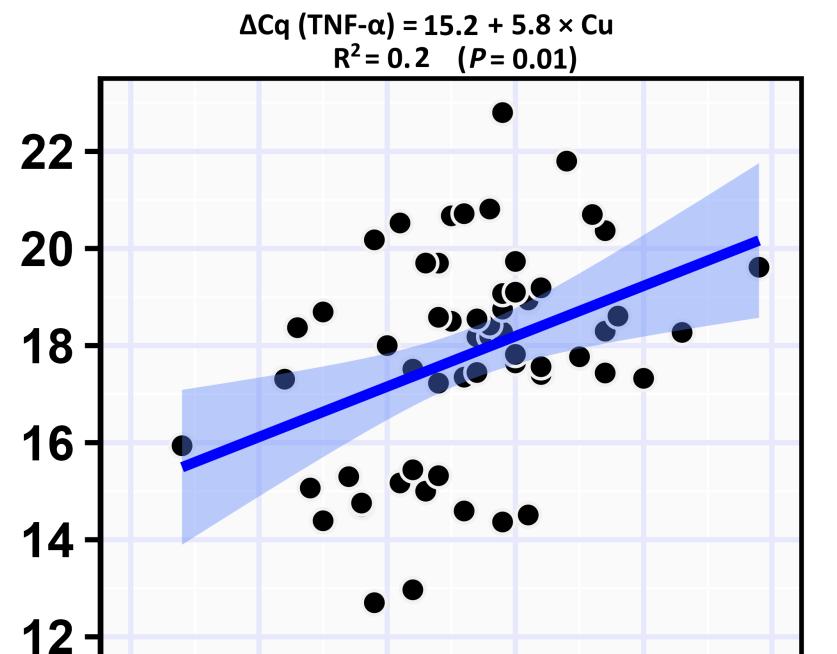
### **Cytokines**

• Cytokines response



Results





post-vaccination is an important driver of both humoral and cellular immunity<sup>3,4</sup>

## Trace minerals

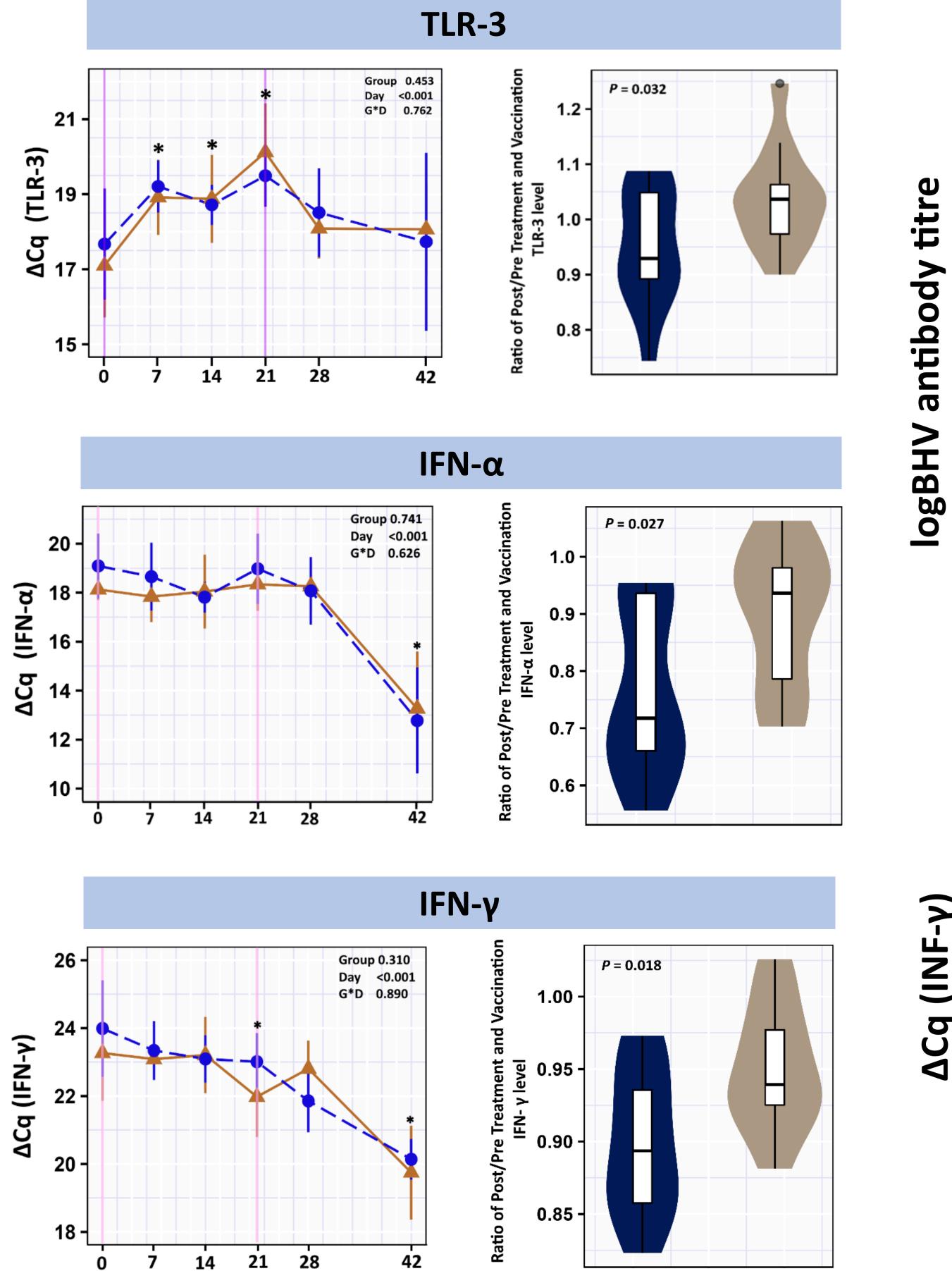
• It has been found that trace mineral supplementation concurrent with a modified-live virus (MLV) vaccine containing bovine viral diarrhea virus 1 and 2 (BVDV), bovine herpesvirus 1 (BHV), bovine respiratory syncytial virus (BRSV), and parainfluenza 3 virus (PI3V) in dairy calves resulted in earlier and robust antibody titers and leukocyte proliferation.<sup>5</sup> However, the mechanism underlying this is not well understood

IL-9, IL-10, IL-13, IL-25

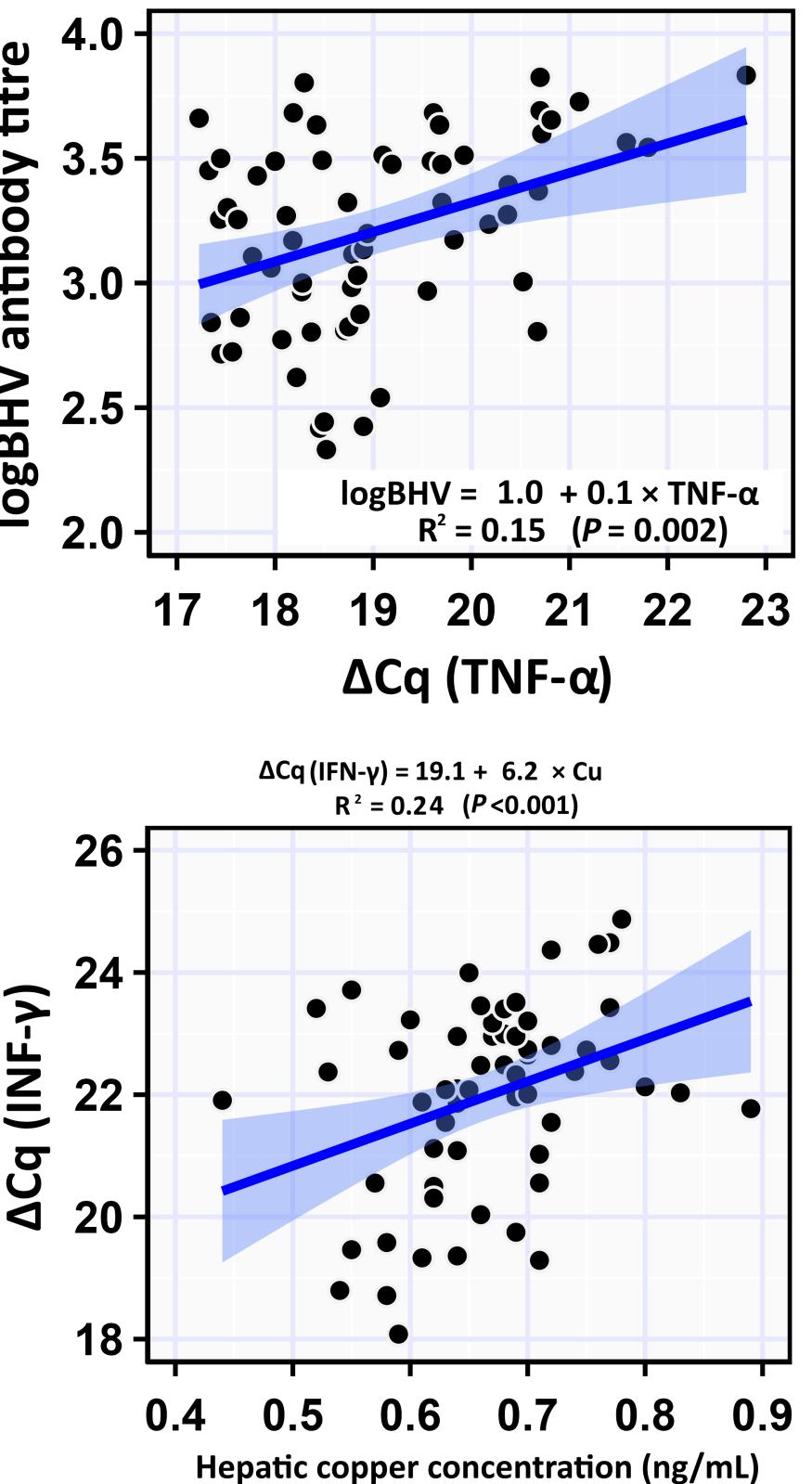
## **Hypothesis & Objectives**

### **Hypothesis**

Injectable trace minerals (ITM) supplementation boosts antibody titers and leukocyte proliferation after BRD vaccination by modulating pro-inflammatory cytokines



### 0.4 0.9 .5 Hepatic copper concentration (ng/mL)



### **Objective**

To characterize the effects of ITM on the cytokines response to the modified-live virus (MLV) vaccine, and attenuated-live *Mannheimia-Pasteurella* (MP) bacterin in dairy calves

## Materials and Methods

Thirty weaned Holstein bull calves (~ 3.5 months) – **Randomized clinical trial** 

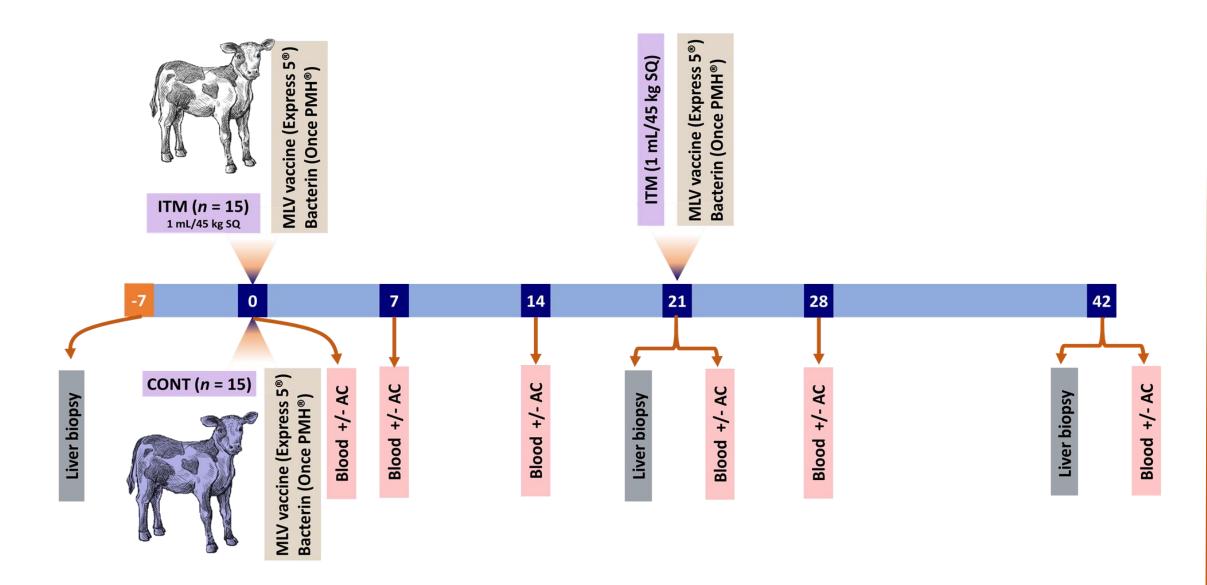


Figure 1. Mean ± SD of cytokines **ΔCq values in ITM-treated calves** (tan solid line) and CONT-treated calves (**blue dashed line**). +indicates a significant difference between treatment groups. \*indicate a significant difference in sampling days compared to day 0.

Figure 2. Boxplot of the ratio of the post (d28) to pre-treatment and vaccination (d0) of cytokine levels in ITMtreated calves (tan violin **plot**) and CONT-treated calves (**blue violin plot**).

Figure 3. The associations between ΔCq values of cytokines and variables of **interest.** The solid blue line is the regression line, and the blue shaded area is the 95% confidence interval for the regression line.

## References

### **Methods**

- Trace minerals were assessed in the liver biopsy samples and serum-neutralizing antibody titers against BVDV, BHV, BSV, and MP were determined.
- The level of cytokines mRNA present in a sample was measured using RT-PCR.
- $\Delta Cq = Cq$  (gene of interest) Cq (housekeeping gene)

## Statistical analysis

- Because of the naturally occurring variability of cytokine levels, the ratios of the post (d28) to pretreatment & vaccination (d0) values for each animal were calculated
- Repeated-measures analysis of variance was used

## Conclusions

Injectable trace mineral supplementations seem to upregulate the expression of inflammatory cytokines, including TNF-α, TLR-3, INF-α, and INF- γ post-BRD vaccination in dairy calves. However, no impact was observed on TLR-7, IL-1 β, IL-10, IL-12, and CD80<sup>+</sup>, CD86<sup>+</sup>.

### Significance

Injectable trace mineral supplementations likely potentiate the immunogenicity of the BRD complex vaccines through the regulation of inflammatory cytokines release postvaccination in dairy calves; however, it is unclear whether these effects contribute to improved protection against BRD in dairy calves.

1. Panciera, et al. 2010. Pathogenesis and pathology of bovine pneumonia Vet. Clin. North Am. Food Anim. Pract. 26:191-214.

2.USDA Dairy 2007, Heifer calf health and management practices on U.S. dairy operations, 2007 USDA: APHIS:VS 3. Forbester, et al. 2021. Genetic influences on viral-induced cytokine responses in the lung. Mucosal Immunol. 14:14-25. 4. Schijns, et al. 2021. Rational vaccine design in times of emerging diseases: the critical choices of immunological correlates of protection, vaccine antigen and immunomodulation. Pharmaceutics. 13:10.33390 5. Palomares, et al. 2016. Effects of injectable trace minerals on humoral and cell-mediated immune responses to *Bovine* viral diarrhea virus, Bovine herpes virus 1 and Bovine respiratory syncytial virus following administration of a modified-live virus vaccine in dairy calves. Vet Immun. and Immunopath. 178:88-98.

