FACULTY OF VETERINARY MEDICINE
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GHENT UNIVERSITY

INVITATION
PUBLIC DEFENSE
Title:
Curcumin and fish oil as natural anti-inflammatory compounds in Holstein neonatal calves: a metabolomics approach

Saeid Kameloroumieh

PROMOTERS
Prof. dr. Lynn Vanhaecke
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Curriculum Vitae

Saeid Kameloroumieh was born on January 4, 1988, in Iran. In 2007, he obtained his BSc in Animal Science at Ferdowsi University of Mashhad, Iran. Subsequently, in 2011 he performed research on nutrition in Holstein Dairy cows, to obtain his MSc degree from the Department of Animal Science at Ferdowsi University of Mashhad. He started his Ph.D. on the effect of natural compounds on calf feces metabolome at Ghent University in 2019. He is interested in improving livestock productivity, through nutrition and metabolomics research. He is the author of four scientific publications and was a speaker on two symposia while taking an active part in several national and international conferences.

Where?

The defense takes place on

Monday, November 8, 2021, at 5 p.m.

Auditorium 1
Faculty of Veterinary Medicine,
Ghent University, Campus Merelbeke
Salisburylaan 133, Merelbeke

A short reception will follow after the defense.

Registration

If you wish to attend the reception, please register before November 3, by e-mail at Saeid.Kameloroumieh@Ugent.be

Members of the Examination Board

Prof. dr. Siska Croubels
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Thesis summary

Calf mortality is a critical cause of economic loss in the dairy cow industry. In this regard, gram-negative bacteria are one of the most pathogenic agents which are common in calf diseases and have a significant effect on the health status of calves. Complementary and alternative medicines (CAM) may improve enteric disease resistance. Popular CAM therapies include amongst other herbs (e.g., cannabis, Chinese medicine, and curcumin), vitamins, probiotics, and fatty acids. Curcumin (CUR) is particularly promising because it has been ascribed antioxidant, anti-inflammatory, cancer chemo-preventive, and potential chemotherapeutic properties. In addition, feeding diets containing marine n-3 PUFA may be beneficial for calf health as well, as it alters the cytokine response and regulation of the inflammatory response, hence affecting the ability of the animal to respond to disease. Due to the speedy pathogenesis of gram-negative bacterial infections in calves, an early overcoming of the infection is critical since late treatment is typically not adequate to alter the outcome of the disease process. In this regard, metabolomics has surfaced in recent years as a promising methodology, reflecting the interactions of genetics with the environment, residing microbial ecosystem, and feeding regime, as such accurately representing the calf's phenotype. Indeed, metabolomics may support the identification of biomarkers that can be used for disease diagnosis or even risk prediction, as well as to unravel (patho)physiological pathways.

As the first part of our study, we aimed at investigating the effect of nanocurcumin (NCUR) and fish oil (FO) as natural anti-inflammatory compounds *vs.* dexamethasone (DEX) in response to a lipopolysaccharide (LPS) challenge in calves. To this extent, a total of 42 male Holstein calves were randomized into 7 groups: negative control (CON), positive control (LPS), fish oil 250 and 350 mg/kg BW per day + LPS, nanocurcumin 2 and 4 mg/kg BW per day + LPS, and 0.3 mg/kg BW dexamethasone + LPS. Except for the CON group, the other experimental groups were intravenously challenged once with 0.5 µg/kg BW ultrapure LPS from *E. coli* serotype 0111:B4. No difference between FO and NCUR *vs.* LPS in sickness behavior was observed. However, the DEX group recovered significantly faster than the other groups ($P < 0.01$). Finally, no effects of FO and NCUR on cytokines and acute-phase proteins could be observed and there was no significant difference between groups in dry matter intake and average daily weight gain during three days post LPS challenge. In conclusion, supplementation of FO and NCUR was not able to impact the acute phase response in calves, as levels of inflammatory cytokines, and acute-phase proteins, as well as sickness behavior, remained unchanged.

The second part of our study was dedicated to the identification of fecal metabolites that were significantly affected by LPS followed by assessing the impact of DEX, FO, and CUR treatments on the putatively identified gram-negative bacterial endotoxin biomarkers. To do this, fecal samples were acquired on a day before starting the experiment and a day after the LPS challenge. Ultra-high-performance liquid-chromatography coupled to high-resolution mass spectrometry (UHPLC-HRMS) was employed to map the metabolic fingerprints of feces from the various groups, before and after the LPS challenge. Based on the generated fingerprints, including 9,650 unique feature ions, significant separation according to the LPS group (day 0 *vs.* 9) was achieved through orthogonal partial least squares discriminant analysis, which allowed to retain 37 metabolites as bacterial endotoxin markers. Tentative identification of these markers indicated that the majority was belonging to the subclass of the carboxylic acid derivatives; amino acids, peptides, and analogs; and fatty amides, playing a role in the metabolism of steroids, histidine, glutamate, and folate. In conclusion, this study allowed to reveal various metabolite biomarkers that were significantly altered upon LPS-challenge and thus potentially involved in acute phase response. Moreover, dexamethasone, nanocurcumin, and fish oil had a positive effect on around 64, 29, and 27% of the revealed gram-negative bacterial endotoxin biomarkers, respectively.

In general, these findings may suggest that the effect of NCUR and FO on NF-κB transcription in ruminants is different compared to other animals. Thus, proof of concept is needed in cell culture to assess the potential protective effects of NCUR and FO on NF-κB transcription in calves. Since NCUR and FO had a significant effect on some gram-negative bacterial endotoxin biomarkers (generally around 30%), it is interesting to consider whether their combination can have a synergistic effect and lead to better calf performance during the disease. In addition, evaluating the effects of co-administration of other anti-inflammatory herbs such as green tea and black pepper with CUR in calves' diets could be of interest for future experiments. Finally, because most gram-negative bacterial endotoxin biomarkers belonged to the carboxylic acids and fatty amides class/subclasses, lipidomics experiments can be of particular importance in assessing metabolic pathways affected by gram-negative bacterial endotoxins as well as identifying new biomarkers.