



UNIVERSITÀ DEGLI STUDI DI MILANO

DIPARTIMENTO DI SCIENZE VETERINARIE
PER LA SALUTE, LA PRODUZIONE ANIMALE
E LA SICUREZZA ALIMENTARE



STSM proposal: Dr Carlotta Giromini

A metabolomic investigation of choline supplementation using ^1H NMR spectroscopy of bovine blood plasma

Choline is considered to be a vitamin-like compound that is essential for mammals when the supply in methionine and folates is not adequate (Zeisel, 2000). Functions of choline in the organism can be divided into two areas: choline per se, and choline as a source of methyl groups (Pinotti *et al.*, 2008; Pinotti, 2012). Choline per se is a constituent of all cell membranes in the form of phosphatidylcholine, lysophosphatidylcholine, choline plasmalogen or sphingomyelin and it plays a crucial role in lipid metabolism. Choline is also an important source of labile methyl groups for the biosynthesis of other methylated compounds.

During my Ph.D. I have investigated the *in vitro* and *in vivo* role of choline and methionine with different studies. Due to their physiological health-promoting effects, they are commonly used in animal nutrition, being part of the normal diet. Choline and methionine have been identified as required nutrients and play various biological functions on cells, including regulation of growth and proliferation. One of the main outcomes of my thesis was that under a condition of stress, choline and methionine have an important role in enhancing cell viability and in counteracting oxidative stress *in vitro* (Rebucci *et al.*, 2013).

Choline has been proposed as limiting nutrient for lactating dairy cow. Accordingly, at the University of Milan we performed a study aimed to evaluate the effect of Rumen-Protected Choline (RPC) supplementation in early lactating dairy cow receiving hay-based diet. For this purpose 14 Italian Holstein multiparous cows, in the first month of lactation (28 DIM), were divided into two experimental groups: control group (CTR) receiving no choline, and RPC group receiving 20 g/day of choline in rumen protected form (Balchem, New Hampton, NY).

Throughout the 9 weeks, treatment did not affect the DMI (22.7 vs. 23.5 kg/d in CTR and RPC respectively) and milk yield (28.2 vs. 29.4 kg/d). Throughout the first 3 weeks of the experiment choline treatment did not affect the metabolic profile of experimental cows, except for plasma glucose concentrations, which was higher in RPC cows than in CTR cows (3.26 vs. 3.00 mmol/L). Plasma methionine content was not influenced by the treatment. By contrast lysine concentration was higher in RPC cows than in CTR cows (43.6 vs. 35.8 $\mu\text{mol/mL}$) (Pinotti *et al.*, 2014).



During the STSM the plan is to further investigate the effect of choline supplementation by metabolomics studies. In particular, the main aim will be to identify possible metabolite markers linked to the effect of choline supplementation using ^1H nuclear magnetic resonance (NMR). The principal component analysis (PCA) and partial least square–discriminant analysis (PLS-DA), assessment will be done to identify those possible metabolite markers.

Plasma metabolomics may provide a deeper understanding of the effect of choline supplementation in dairy cow feed with hay-based diet. Studies of metabolic processes by NMR on biological fluids can be considered an emerging and promising science with a level of information that spans the traditional approach for elucidating the metabolic plasma profile.

The Universitat Autònoma de Barcelona (UAB) is the ideal place to carry out the proposed research study. This is because the UAB possess 8 Bruker NMR spectrometers for analysis or study of any sample and Dr. Ahmed A.K. Salama has an experience in the NMR analysis as proven by his publication record. Additionally, and more importantly, the STSM will be an opportunity to establish a collaboration between the University of Milan and the UAB.

Therefore, Dr. Giromini will greatly benefit from the great expertise of the Ruminant Research Group of the UAB both on the NMR analysis and on both animal physiology and nutrition. Furthermore, the experience acquired will be used in future work at Dr. Giromini home university.

The aim of the STSM is the collaboration between early stage researchers in order to produce when possible joint publications according to COST action aims. Due to the importance of collaborative work within DairyCare, the two research units (i.e. Universitat Autònoma de Barcelona and the University of Milan) are willing to work together for the production of a high quality academic publication.

References

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