

Effects of rumen protected choline during transition phase on haematology of dairy cows

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Previous findings on choline in dairy cows

- Milk production and quality ⇒ generally, increased milk fat production
- Liver metabolism ⇒ improved prevention of fatty liver and ketosis
- Reproduction \Rightarrow (?)
- Haematology \Rightarrow (?)
- 2 types of role for choline:
 - Role of choline per se
 - Role as a methyl donor

Choline, metabolism, and haematology

Choline per se

PtdCho component

60 % of milk phospholipids contain choline → phospholipids are 0.5-1.0 % of total milk lipids → 105-210 mg/L of choline-containing phospholipids ⇒ choline is a limiting metabolite in lactating mammary tissue (Pinotti et al., 2002)

PtdCho essential component of VLDL and cannot be substituted by other phospholipids (Pinotti et al., 2002)

Increased synthesis of PtdCho → building of membranes during phagocytosis (García Gil et al., 1982; Biochem J)

Choline as methyl donor

- 2 principal methyl donors in animal metabolism
- choline → betaine
- methionine → S-adenosyl-methionine

Methyl groups may be derive from

- exogenous sources: Met, betaine, choline
- *de novo*: tetrahydrofolate (THF) system → folic acid and vitamin B12

Choline as methyl donor

Labile methyl groups: choline and Met are interrelated High producing dairy cow: Met is the 1st limiting a.a. Elevated requirement for Met for transmethylation reactions and milk protein synthesis ⇒ altered methyl group metabolism (Lobley et al., 1996) Ruminants → conservative methyl group metabolism

- elevated rate of *de novo* methyl group synthesis from the one-carbon pool
- low rate of methyl catabolism (low activity of choline oxidase)

Interchangeability → 28 % of Met is used for choline synthesis (Emmanuel and Kennelly, 1984) EAAP - 59th Annual Meeting VILNIUS (LITHUANIA) August 24th-27th 2008

Why haematology in nutrition?

Leukocyte

• progenitor cells (bone marrow): ketone bodies inhibit growth and differentiation $\rightarrow \frac{\Downarrow}{2}$ circulating <u>PMN</u> (Hoeben et al., 1999)

adipocytes in bone marrow? (Mikhail et al., 1997)

 development and proliferation: pH, BHBA (Donovan et al., 2003)

 functions: hormones (insulin, Okouchi et al., 2002), metabolites, vitamins, and trace minerals (Lacetera et al., 2002; Politis et al., 2004; Weiss and Hogan, 2007)

• BHBA $\rightarrow \Downarrow$ NEU extracellular traps and bactericidal activity (Grinberg et al., 2008)

Why haematology in nutrition?

Leukocyte

 <u>phagocytosis of apoptotic circulating PMNs</u>: liver and Kupffer cells status (Shi et al., 2001)

Erythrocyte

 nutritional status for vitamins and trace minerals

membrane fatty acids

Aim of this work

To examine the effects of supplementation of rumen protected choline (RPC) to transition cows on haematological features

Animals

- 22 Italian Friesian cows (parity ≥ 2) randomly assigned by
 - expected calving date
 - parity
 - previous milk yield
- to either be supplemented with rumen-protected choline (RPC) from d –21 relative to expected parturition until 35 DIM, or to consume basal diet only (CON)

Herd management and data recording

- cows milked with an automatic milking system
- milk yield was recorded at each milking

Item	Unit	Prepartum	Lactation
DM	% as fed	67.8	54.7
Estimated DMI	kg/d	11.3	20.1
Nutrients (on DM basis)			
NEL	Mcal/kg	1.38	1.64
ME	Mcal/d	24.3	49.2
СР	%	13.6	16.2
NDF	%	51.0	35.5
Met (estimated)	g/d	18	43

Treatments

- CON: TMR basal dry-cow diet until calving, followed by TMR basal lactation diet
- RPC: TMR basal dry-cow diet + 50 g of RPC per cow top dressed product (Sta-Chol[®], Ascor Chimici, Italy) until calving, followed by TMR basal lactation diet + 50 g of RPC per cow top dressed product just after TMR distribution
- RPC with 50% choline as choline chloride

Blood sampling

- before TMR distribution
- jugular blood samples: once just before the trial start, and then weekly until the 10th wk of lactation
- 5 mL evacuated sampling tubes, with K₃-EDTA as anticoagulant

Blood analysis

 automatic hematological analyzer (Cell Dyn 3700, Abbott Diagnostici, Italy)

Determined features:

- WBC; NEU; LYM; MONO; EOS; BASO
- RBC; HGB; HCT; MCV; MCH; MCHC
- PLT; MPV; PDW



Statistical analysis

- pre- and post-partum data separately analyzed
- randomized block design, mixed model
- main effects: choline supplementation (RPC vs. CON), week of trial (-3, -2, -1 and 1, 2, 3, 4, 5, 6, 7, 8, 9, and 10), and their interactions
- cow repeated in time

WBC count



Neutrophils count



Neutrophils percentage



Eosinophils count



Eosinophils percentage



Choline and neutrophils

- regulating n° of PMNs in the circulation and prompt removal of senescent PMNs → maintaining normal immune function and preventing tissue injury
- active phagocytosis of apoptotic circulating PMNs was essentially limited to the liver
- PMNs phagocytosed by Kupffer cells (Shi et al., 2001)
 ⇒ a role for choline in this activity?
- PtdCho to building PMN membrane during phagocytosis (García Gil et al., 1982) ⇒ a choline effect to improve PMN function in phagocytosis?





Plasma insulin concentration

Plasma NEFA

Abeni et al., 2007

Hypothesis

Choline and eosinophils

- Difficult to explain:
 - Data variance
 - Few available papers on this topic
- Regulation of eosinophil adhesion by lysophosphatidylcholine (Zhu et al., 2007) ⇒ improved adhesion and, as consequence, transmigration of EOS in tissues?
- Choline treatment (in mouse) significantly inhibited eosinophilic airway inflammation (Mehta et al., 2007) ⇒ a choline effect in reducing allergen-induced inflammation?

Lymphocytes count

Lymphocytes percentage

Conclusion

- Cows fed RPC had lower total leukocyte count (P<0.001), neutrophil count (P<0.05), lymphocyte count (P<0.001), and eosinophil count (P<0.001) than CON cows throughout their first 10 weeks of lactation
- Small differences between treatments on erythrocyte-related variables, both before and after calving
- Supplementation of transition cow diet with RPC affected the number of circulating leukocyte in early lactation, but further research is necessary to better understand the implications of these effects on transition cow health

Thank you for your attention

