

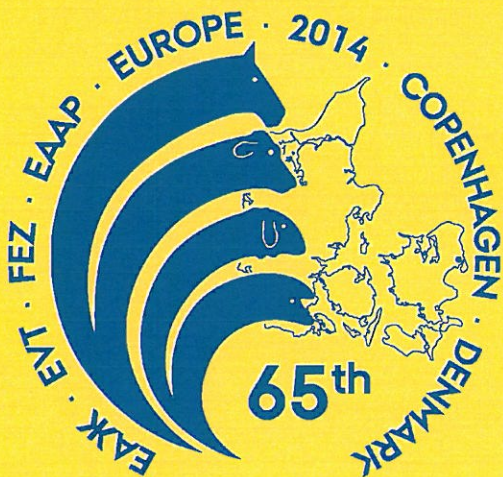
**Vitamin A and E circulate in different cattle plasma fractions***L. Hymøller and S.K. Jensen**Aarhus University, Animal Science, Blichersallé 20, 8830 Tjele, Denmark; lone.hymoller@agrsci.dk*

Analysis of fat soluble vitamins in plasma fractions of protein, chylomicrons, and lipoproteins can give information about their plasma transport and physiological function. Vitamin A (A) and vitamin E (E) are known to circulate in different plasma fractions; hence they were used to verify a simple plasma fractionation method for cattle plasma. Blood was collected in Na-EDTA coated tubes from 3 Holstein bull calves, which for 3 weeks prior to sampling were fed 3 kg/d of concentrate. The concentrate contained 3 µg/kg A and 120 mg/kg E. Plasma was isolated by centrifugation at 1,500×g for 10 min. Chylomicrons were floated by ultracentrifugation of 6 ml plasma at 100,000×g for 1 h in a Beckman-Coulter 70.1 fixed angle rotor. Fractions of lipoproteins (LPF) and protein were separated by adding 1.5 ml OptiPrep™ (Axis-Shield, Norway), layering Hepes buffered saline 0.85%, pH 7.4 on top, and applying ultracentrifugation at 300,000×g for 18 h. Fractions were harvested top-down with Pasteur pipette and analysed for A and E by HPLC. Average amounts of A and E found in each plasma fraction, in % of the total amount of A and E in all fractions, were: Protein: A (54%) and E (7%). Chylomicrons: A (13%) and E (15%). LPF1 (VLDL): A (1%) and E (2%). LPF2 (LDL/HDL): A (5%) and E (62%). LPF3 (HDL): A (27%) and E (15%). A is transported in chylomicrons from its site of uptake in the intestines but circulates in plasma bound to specific binding proteins, which are responsible for its biological action in the body. Likewise E is transported from the intestines to the liver in chylomicrons. Binding proteins specific to E is known to exist, however E is found in the LDL/HDL fraction of plasma. This indicates that cattle plasma was separated into relevant plasma fractions using the presented ultracentrifugation method. In conclusion, by fractionating cattle plasma by ultracentrifugation it was shown that A mainly circulated in heavier plasma fractions i.e. protein and HDL, whereas most E was found in fractions representing LDL/HDL.

**In different worlds: transport of dietary vitamin D<sub>2</sub> and vitamin D<sub>3</sub> in cattle plasma fractions***L. Hymøller and S.K. Jensen**Aarhus University, Animal Science, Blichersallé 20, 8800 Tjele, Denmark; lone.hymoller@agrsci.dk*

It is often stated that vitamin D<sub>2</sub> (D<sub>2</sub>) and D<sub>3</sub> (D<sub>3</sub>) have similar physiological effects but studies in cattle showed, that D<sub>2</sub> is less efficient than D<sub>3</sub> at securing vitamin D status, measured as liver derived 25-(OH)-D<sub>2</sub> (25OHD<sub>2</sub>) and 25-(OH)-D<sub>3</sub> (25OHD<sub>3</sub>). The aim was to investigate if binding to different plasma fractions could explain the physiological inefficiency of D<sub>2</sub> compared to D<sub>3</sub> in cattle. 3 Holstein bull calves were fed 75 µg/d D<sub>3</sub> and had ad lib access to hay, naturally containing D<sub>2</sub>. Blood was collected weekly for 11 weeks and plasma isolated by centrifugation. Plasma was fractionated by ultracentrifugation and fractions of protein, chylomicron, and lipoprotein (LPF) analysed for content of 25OHD<sub>2</sub>, 25OHD<sub>3</sub>, D<sub>2</sub>, and D<sub>3</sub>. Contents of 25OHD<sub>2</sub> vs 25OHD<sub>3</sub> and of D<sub>2</sub> vs D<sub>3</sub> in each fraction were compared as average of all samples by Student's t-test. Average percentages of each metabolite in each fraction were: Protein: 25OHD<sub>2</sub> (8%) vs 25OHD<sub>3</sub> (75%; P<0.001) and D<sub>2</sub> (10%) vs D<sub>3</sub> (16%; P+0.03). Chylomicron: 25OHD<sub>2</sub> (39%) vs 25OHD<sub>3</sub> (17%; P<0.01) and D<sub>2</sub> (0%) vs D<sub>3</sub> (6%; P<0.01). LPF1 (VLDL): 25OHD<sub>2</sub> (0%) vs 25OHD<sub>3</sub> (0%) and D<sub>2</sub> (52%) vs D<sub>3</sub> (33%; P+0.07). LPF2 (LDL/HDL): 25OHD<sub>2</sub> (9%) vs 25OHD<sub>3</sub> (2%; P+0.07) and D<sub>2</sub> (4%) vs D<sub>3</sub> (31%; P<0.01). LPF3 (HDL): 25OHD<sub>2</sub> (34%) vs 25OHD<sub>3</sub> (6%; P<0.001) and D<sub>2</sub> (0%) vs D<sub>3</sub> (15%; P<0.01). D<sub>2</sub> and D<sub>3</sub> were only transported evenly distributed in protein, whereas the % of D<sub>2</sub> and D<sub>3</sub> found in other fractions differed. If this means that D<sub>2</sub> is transported in more volatile plasma fractions than D<sub>3</sub>, then D<sub>2</sub> could be more prone to degradation and excretion, contributing to its lower physiological efficiency. 25OHD<sub>3</sub> was mainly transported in protein whereas 25OHD<sub>2</sub> was transported in chylomicron and LDL/HDL. Associating with designated binding proteins is vital to the function of 25OHD<sub>3</sub>. Hence transportation in other plasma fractions than protein could explain a compromised physiological function of 25OHD<sub>2</sub> compared to 25OHD<sub>3</sub>. In conclusion, D<sub>2</sub> and 25OHD<sub>2</sub> are transported in different plasma fractions than D<sub>3</sub> and 25OHD<sub>3</sub>, respectively, which could offer an explanation to a lower physiological efficiency of D<sub>2</sub> than D<sub>3</sub> in cattle.

# **Book of Abstracts of the 65<sup>th</sup> Annual Meeting of the European Federation of Animal Science**



**Book of abstracts No. 20 (2014)  
Copenhagen, Denmark  
25 - 29 August 2014**

# Book of Abstracts of the 65<sup>th</sup> Annual Meeting of the European Federation of Animal Science

Copenhagen, Denmark, 25-29 August, 2014



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**Wageningen Academic  
Publishers**

**EAN: 9789086862481**  
**e-EAN: 9789086867998**  
**ISBN: 978-90-8686-248-1**  
**e-ISBN: 978-90-8686-799-8**  
**DOI: 10.3920/978-90-8686-799-8**

**ISSN 1382-6077**

**First published, 2014**

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**The Netherlands, 2014**

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