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Over-conditioned transition cows are notoriously poor performers and many blame excessive adipose mobilization on this phenomenon. Objectives were to evaluate the effects of feed restriction (FR) on basal and stimulated metabolism in fat and thin cows. Eleven multiparous Holstein cows (80 ± 8 DIM) were assigned to 1 of 2 treatments: (1) FAT (4.20 ± 0.25 BCS; 666 ± 48 kg BW; n = 6) and (2) THIN (2.85 ± 0.25 BCS; 578 ± 90 kg BW; n = 4). Cows were enrolled in 3 experimental periods (P): in P1 (3 d) baseline data were collected, in P2 (5 d) cows were FR to 60% based on P1 intake, and in P3 (3 d) cows were fed ad libitum to recover from FR. To evaluate whole-body nutrient partitioning, epinephrine challenges (EPI; 1.4 µg/kg BW) were performed on d 3 of P1, d 5 of P2, d 3 of P3. Data were analyzed using PROC MIXED of SAS. There were no treatment differences in DMI (27.7 kg/d) and milk yield (21.8 kg/d) during P1. DMI decreased during P2 and subsequently all cows similarly decreased milk yield ($P < 0.05$; 38% by d 5). Overall milk yield during P3 was not different during P2. There was an interaction ($P < 0.05$) on basal NEFA during FR as it acutely increased more in THIN cows (280 vs. 196%), but basal NEFA remained elevated in FAT cows while it slightly decreased as P2 progressed in THIN cows. NEFA acutely and similarly returned to basal levels in P3. Circulating insulin uniformly decreased during FR ($P < 0.05$; 89%). In P3 insulin acutely increased in FAT versus THIN cows (24.5- vs. 12.8-fold; $P < 0.01$) on d 1. Blood glucagon did not differ by period but was overall increased in THIN versus FAT cows (39%; $P < 0.05$). Blood BHB did not increase during P2 in either treatment but did similarly increase during P3 (19%; $P < 0.01$). In response to EPI, the glucose area under the curve (AUC) tended ($P = 0.10$) to decrease during P2 in THIN cows, but did not change in FAT cows. The EPI-induced NEFA AUC did not differ by BCS, but was increased (2.1-fold) during FR and it returned to basal levels in P3 ($P < 0.01$). Overweight cows do not appear to mobilize more adipose tissue during FR than THIN cows. Reasons why periparturient FAT cows are less successful are not likely related to energetics.

Key Words: fat, off-feed, lipid mobilization

1603 Methodology for breath profiling in dairy cows: A novel approach for metabolic assessment. M. A. Barrientos Blanco*[1], M. Z. Islam[1], R. Peng[1], S. E. Räisänen[1], F. Wahl[2], R. Zenobi[3], S. Giannoukos[3], and M. Niu[1], *[1]Animal Nutrition, Institute of Agricultural Sciences, ETH Zürich, Zürich, Switzerland, [2]Food Microbial Systems Research Division, Agroscope, Bern, Switzerland, [3]Department of Chemistry and Applied Biosciences, ETH Zürich, Zürich, Switzerland.*

Frequent eructation in ruminant animals results in a blend of exhaled ruminal and breath volatile organic compounds (VOC). This physiological distinction might limit the applicability of breathomics in describing the metabolic phenotype of cows. The primary focus of this study was to differentiate the origin of exhaled VOC, with the aim to utilize breathomics for the assessment of dairy cow metabolism. Eighteen multiparous lactating Holstein cows (203 ± 60 DIM; 32.7 ± 4.95 kg/d milk yield) were enrolled in the study. Cows were fed a similar diet. Exhaled VOC in breath (Br; blood-borne metabolites) and exhalome (Ex; a mixture of ruminal eructation and Br) were sampled separately using a head chamber (GreenFeed System®; GF) 8 × to represent every 3 h of a day. Methane (CH_4), originating solely from ruminal fermentation, was used as the marker to differentiate breath from eructation events. Using a previously established method by our laboratory, GF real-time readings were used to collect eructation CH_4 peak events as Ex samples. A threshold of <100 ppm CH_4 was set to sample Br. Both samples were analyzed using secondary electrospray ionization-high resolution mass spectrometry. Data were analyzed using mixed model with a random effect of cow and origin × time of day interactions. Logarithmic transformation on Br over Ex mean ratio and fold change (FC) were calculated to differentiate the origin of metabolites. Putative metabolite annotation and most enriched pathways were conducted using MetaboAnalyst. Identified as Ex ($P < 0.01$), ruminal VFA acetate, propionate, and butyrate were differentiated with FC >0.5. Energy metabolites such as pyruvate and TCA cycle precursors citrate, cis-aconitate, succinate, fumarate, and malate were detected in Br ($P < 0.01$), and differentiated with FC >0.2. While variations in VFA concentrations over time were linked to feeding events, TCA cycle precursors seemed to vary relative to milking time. Our study not only established a method to sample Br versus Ex but also provided compelling evidence of the potential to implement breathomics as a noninvasive tool for conducting metabolic assessment in ruminant research.

Key Words: dairy cow, metabolomics, breath

1604 Links between volatile organic compounds from exhaled breath and metabolites of blood and ruminal fluid of lactating dairy cows. J. Eichinger*[1,3], A.-M. Reiche[1], P. Fuchsmann[2], K. Huber[3], and F. Dohme-Meier[1], *[1]Agroscope, Fribourg, Switzerland, [2]Agroscope, Bern, Switzerland, [3]University of Hohenheim, Baden-Wuerttemberg, Germany.*

Exhaled breath sampling is a low-invasive intervention and an interesting matrix as it contains information about the organism's metabolism in form of volatile organic compounds (VOC). However, the exact physiological information of exhaled VOC about a cow's metabolism is mainly unknown. Therefore, we performed an untargeted analysis of exhaled VOC and investigated their relationship with blood and ruminal fluid metabolites. Thirty-two lactating healthy Holstein cows (83.27 ± 31.40 d in milk) were fed either hay (n = 16) or silage (n = 16); all cows were supplemented with concentrate feed. The study lasted 12 weeks with sampling of exhaled breath, blood, and ruminal fluid every 4 weeks. The VOC were analyzed by dynamic headspace vacuum-in-tube extraction GC-MS. Concentrations of glucose, nonesterified fatty acids (NEFA), β-hydroxybutyrate (BHB), cholesterol, and urea in blood were analyzed by a BT1500 autoanalyzer, short-chain fatty acids (SCFA) by HPLC, and ammonia in ruminal fluid colorimetrically. Pearson correlations between exhaled VOC and blood and ruminal fluid metabolites were calculated separately for each sampling time point. Significant ($P < 0.05$) diet-independent correlations with r > 0.35 were selected and the implicated VOC were identified (NIST 17, Gaithersburg, MD). Five exhaled VOC (4-hydroxy-4-methylpentan-2-one, tetradecanal, 3-penten-2-one, γ-hydroxybutyrate, dodecanal) correlated with blood metabolites (BHB and cholesterol, BHB and NEFA, NEFA, glucose and urea, glucose, respectively) and 6 (2-ethylhexanal, 4-hydroxy-4-methylpentan-2-one, γ-hydroxybutyrate, 3-penten-2-one, 2-ethylhexan-1-ol, p-cymene) correlated with ruminal fluid metabolites (acetate and butyrate, acetate, acetate and valerate, butyrate, butyrate, ammonia, respectively). The correlated VOC are e.g., part of the fatty alcohol cycle and the GABA shunt. This suggests that exhaled breath of cows contains information about the fatty acid, amino acid, and therefore energy metabolism. The usability of exhaled breath sampling as low-invasive intervention to assess other metabolic states needs further investigation.

Key Words: noninvasive, metabolism, volatilome