Associations between the hepatic lipidome and feed efficiency in Holstein dairy cows

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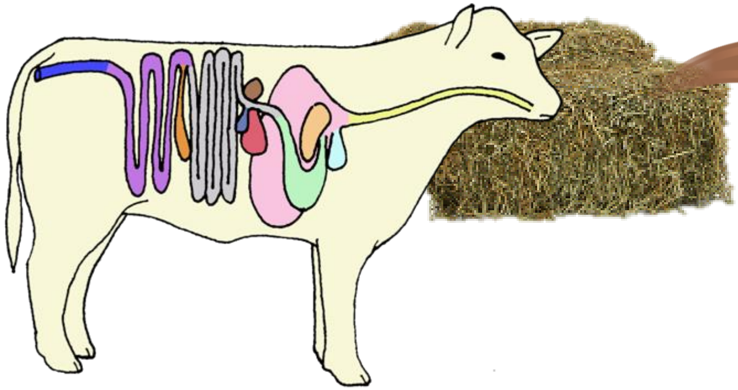
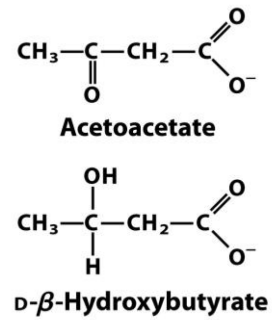
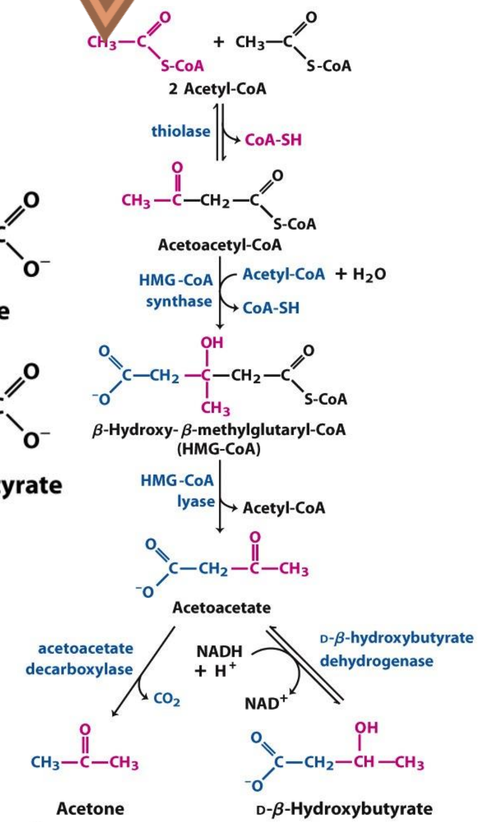
The Problem

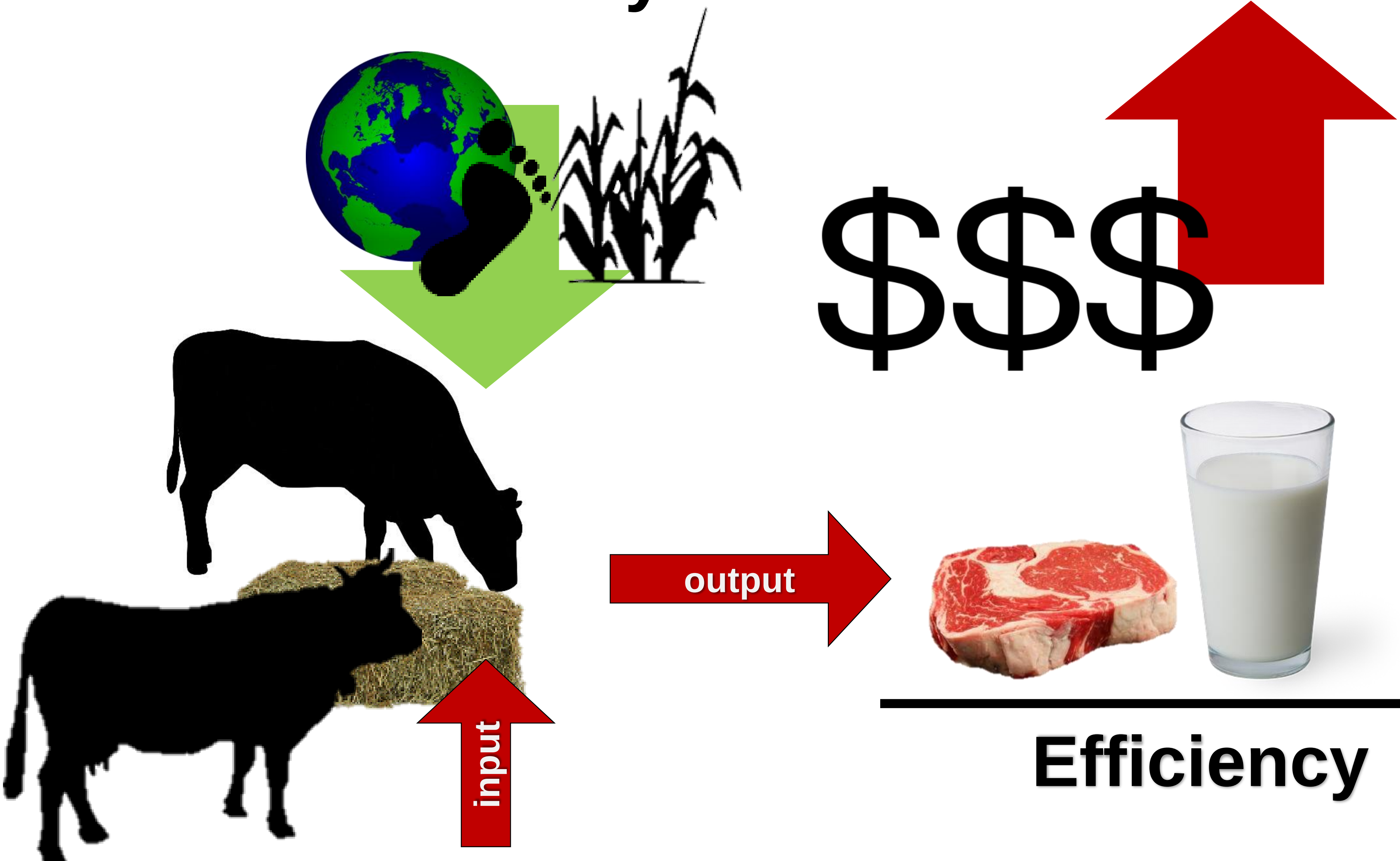






Transition

Metabolic efficiency



Important things to keep in mind about metabolic efficiency

- Economical vs. biological efficiency
- Metabolic efficiency: different measures, different outcomes…

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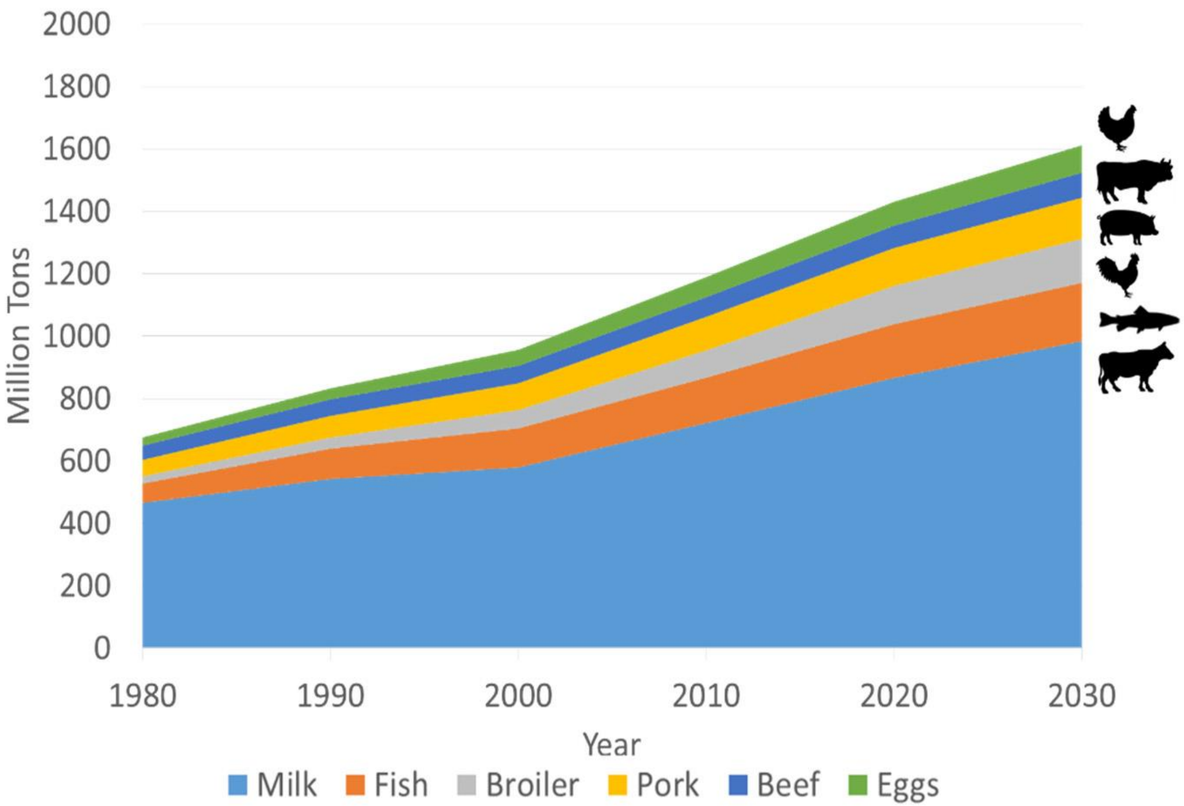
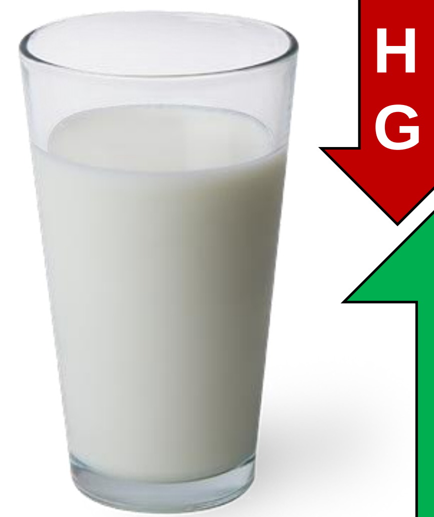
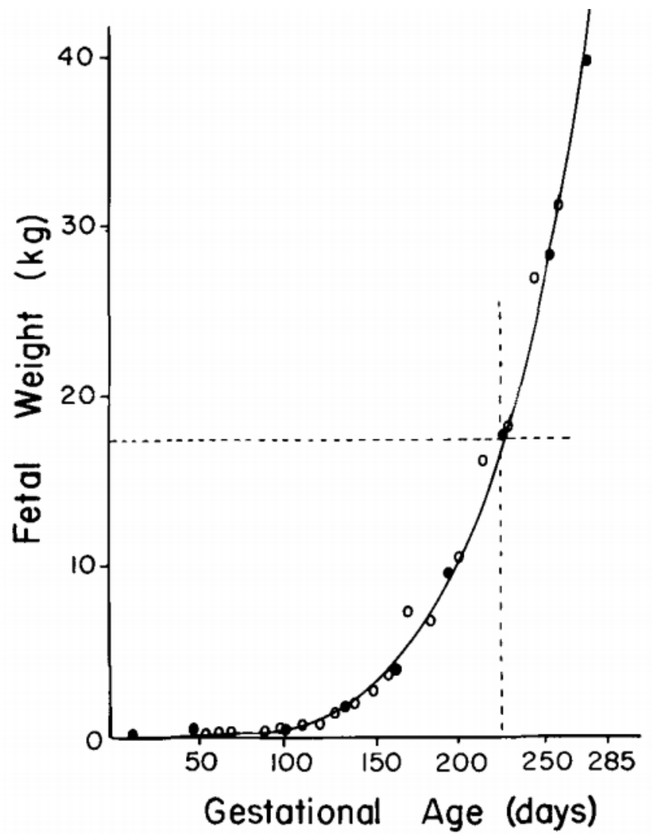
Improving metabolic efficiency as means to feed the growing population

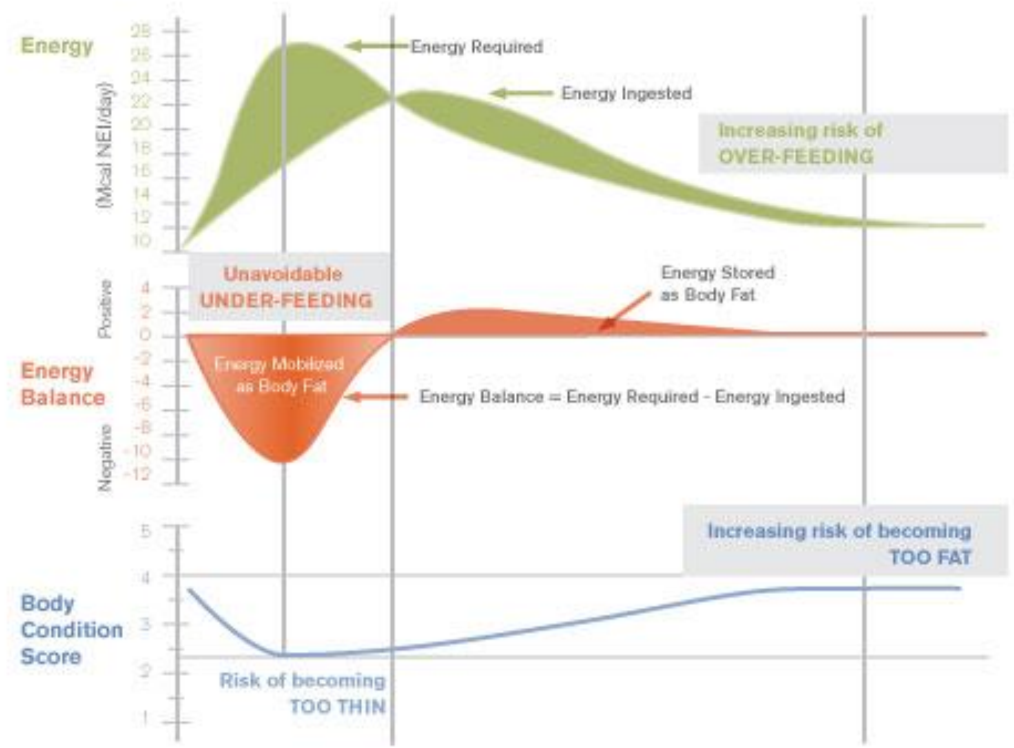




Alexandratos and Bruinsma, 2012; Capper et al., 2009

Transition period in dairy cows





Bauman and Currie, 1980

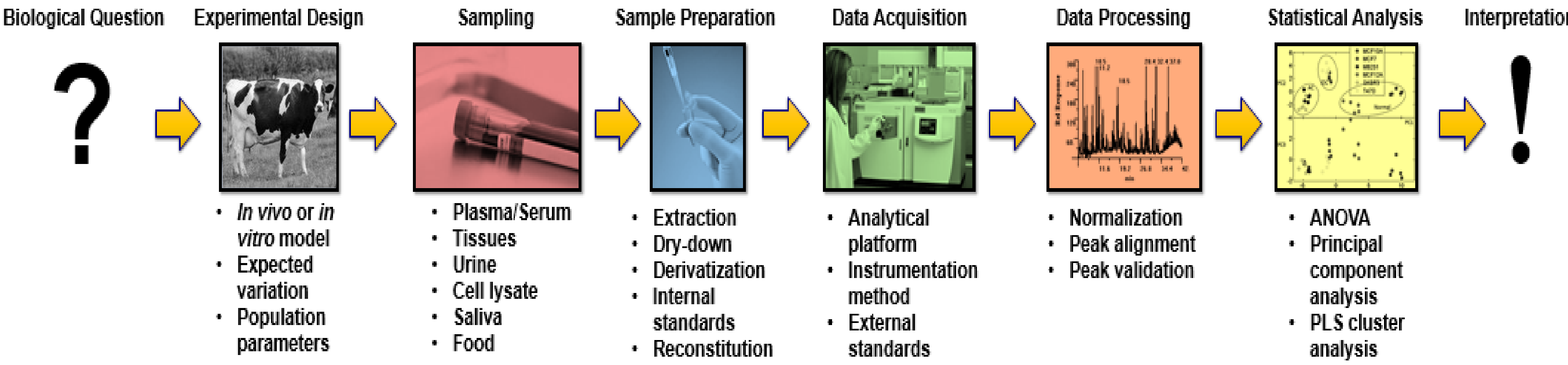
Nutrient partitioning

- Homeostasis – Maintenance of physiological equilibrium, *i.e.* constant conditions in the internal environment
- Homeorhesis – Orchestrated changes for the priorities of a physiological state, *i.e.* coordination of metabolism in various tissues to support a physiological state

Table II. Possible Homeorhetic Hormones and Glucose-Related Tissue Responses in Pregnancy and Lactation

State	Hormone	Putative action	Tissue/response
Mid pregnancy	Progesterone	↑ insulin sensitivity	↑ adipose glucose uptake ↑ adipose lipogenesis
Late pregnancy	Placental lactogen Estrogens	↓ insulin sensitivity and responsiveness	↓ glucose uptake by adipose and muscle ↓ adipose lipogenesis ↑ muscle glycolysis and lactate release
Lactogenesis, early lactation	Prolactin Estrogen Cortisol Somatotropin	↓ insulin sensitivity and responsiveness	↑ liver gluconeogenesis ↓ glucose uptake by adipose and muscle ↓ adipose lipogenesis ↓ protein synthesis } muscle ↑ protein degradation } muscle ↑ amino acid release } muscle

Lipidomics has transformed our understanding



To date we have measured 1,469 lipids in the cow

- Lipids include:
  Sphingolipids: ceramides, monohexosylceramides, lactosylceramides, and sphingomyelins
  Fatty acylglycerols: diacylglycerols, triacylglycerols, and monoalkyldiacylglycerols
  Glycerophospholipids: phosphatidylcholine, phosphatidylethanolamines, phosphatidylglycerols, phosphatidylserines, and lyso-phospholipids

- Plasma/serum, liver, adipose, skeletal muscle, cultured cells, and isolated lipoproteins (VLDL, LDL, and HDL)

Hypothesis

- Considering the energetic links between nutrient partitioning and hepatic function during the transition period it is reasonable to hypothesize their relationship with metabolic efficiency in dairy cows

Objectives

- To evaluate performance, steatosis and the hepatic lipidome in dairy cows diverging in feed efficiency

Material and Methods

Animals and Experimental Condition

- DoVan Farms, Berlin, PA
- 23 multiparous gestating and lactating Holstein cows

  BW: 646 ± 86 kg,

  BCS: 3.4 ± 0.54,

  MY: 37.52 ± 3.84 kg

  Parity: 3.59 ± 1.64
- Corn silage-based diet

Ingredients, % of DM	Pre-partum	Lactation
Corn silage	29.00	33.20
Grass haylage	21.70	8.61
Grass hay	25.00	1.95
Dry ground corn	2.72	20.80
Prepartum mix[1]	12.80	-
Lactation mix A[2]	-	17.80
Aminoplus ®	5.82	5.72
Cottonseed with lint	-	4.35
Sugar cane syrup	-	3.40
Lactation mix B[3]	-	4.19
Close-up supplement[4]	3.01	-
Nutrient Composition	**Pre-partum**	**Lactation**
DM, %	51.30	51.70
CP, % of DM	13.40	15.50
NDF, % of DM	47.90	31.10
ADF, % of DM	30.50	19.80
Crude fat, % of DM	3.30	4.80
Ash, % of DM	8.30	7.90

Productive Performance

- 40-day evaluation period
  - Weekly BW, BCS
  - Daily MY (thrice daily)
- Liver biopsy
  - Pre-partum (d = - 12)
  - Post-partum (d = 10)

Productive Performance

- Residual Feed Intake (Koch et al., 1963; Xi et al., 2016)

$$RFI = DMI_{actual} - DMI_{predicted}$$

$$DMI_{predicted} = 7.68 + (0.16*parity) + (0.37*pcADG) - (0.02*pcBW^{0.75}) + (0.21*ECM)$$

$r^2 = 0.33$

- High-RFI = Inefficient > 0.5 SD mean (n = 10)
- Low-RFI = Efficient < 0.5 SD mean (n = 13)

Sample Analyses

- Liver lipid content

Hexane Method (Hara and Radin, 1978; Starke et al., 2010)

- Time-of-flight mass spectrometry (TOFMS)

Statistical Analyses

MetaboAnalyst (http://www.metaboanalyst.ca/)

- Log transformation, auto-scale

  Scores Plot

  VIP Scores

SAS (version 9.4): GLM procedure

- LS Means: productive performance and liver lipid of RFI groups
- Partial correlations (MANOVA/PRINTE statement)

- Significance: $P \leq 0.05$
- Trend towards significance: $0.10 \geq P > 0.05$

Results and Discussion

Feed efficiency and milk production

Productive performance traits	Inefficient	Efficient	*P*-value
Dry matter intake (kg/d)	**15.28**	**13.11**	**0.01**
Pregnancy corrected BW (kg)	642.56	649.57	0.86
Residual feed intake (kg/d)	**1.20**	**-0.77**	**0.01**
Parity (n)	3.22	3.85	0.39
Calf BW (kg)	39.64	40.37	0.67
Milk yield (kg/d)	39.15	37.15	0.19
ECM (kg/d)	39.08	38.27	0.67
Fat (%)	3.92	4.10	0.42
Protein (%)	3.41	3.54	0.37
Lactose (%)	4.62	4.67	0.45

Feed efficiency and hepatic lipid content in the pre-partum period

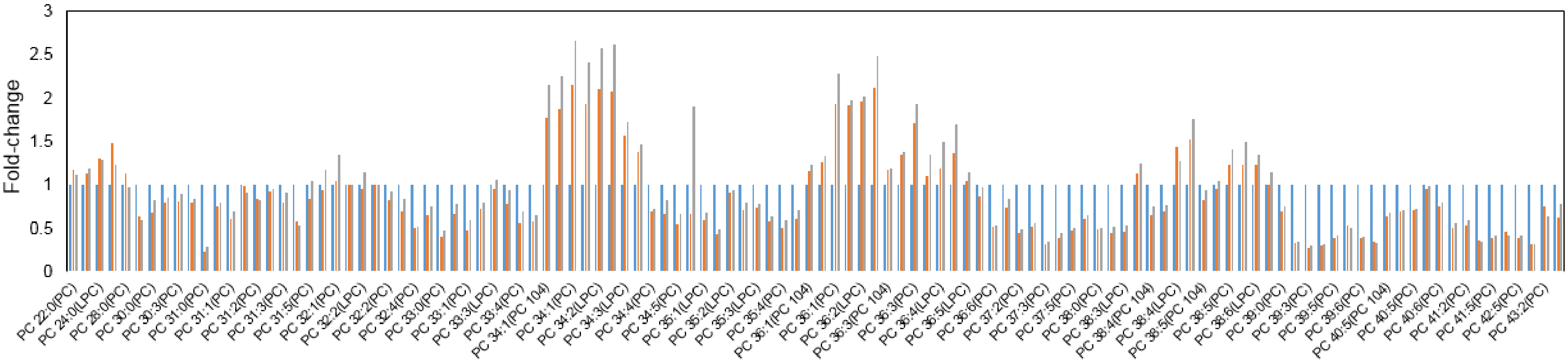
Lipid classes (cps)	Inefficient	Efficient	*P*-value
Sphingolipids			
Ceramide	**24.93**	**22.74**	**0.10**
Monohexosylceramide	**0.35**	**0.38**	**0.09**
Sphingomyelin	103.59	114.99	0.17
Fatty Acylglycerols			
Monoalkyldiacylglycerol	133.64	522.86	0.20
Diacylglycerol	4.81	6.19	0.12
Triacylglycerol	13.68	38.72	0.29
Glycerophospholipids			
Phosphatidylcholine	35.95	38.80	0.25
Phosphatidylethanolamine	26.52	29.44	0.11

Feed efficiency and hepatic lipid content in the post-partum period

Lipid classes (cps)	Inefficient	Efficient	*P*-value
Sphingolipids			
Ceramide	22.76	21.74	0.17
Monohexosylceramide	0.50	0.58	0.20
Sphingomyelin	133.81	133.05	0.96
Fatty Acylglycerols			
Monoalkyldiacylglycerol	2503.70	3321.86	0.31
Diacylglycerol	12.23	20.78	0.23
Triacylglycerol	195.32	307.95	0.25
Glycerophospholipids			
Phosphatidylcholine	43.10	44.53	0.81
Phosphatidylethanolamine	37.46	36.97	0.91

Changes in hepatic lipid moieties are not uniform

- PC example



Plasma; Saed Samii et al., 2017 (ADSA Abstract)

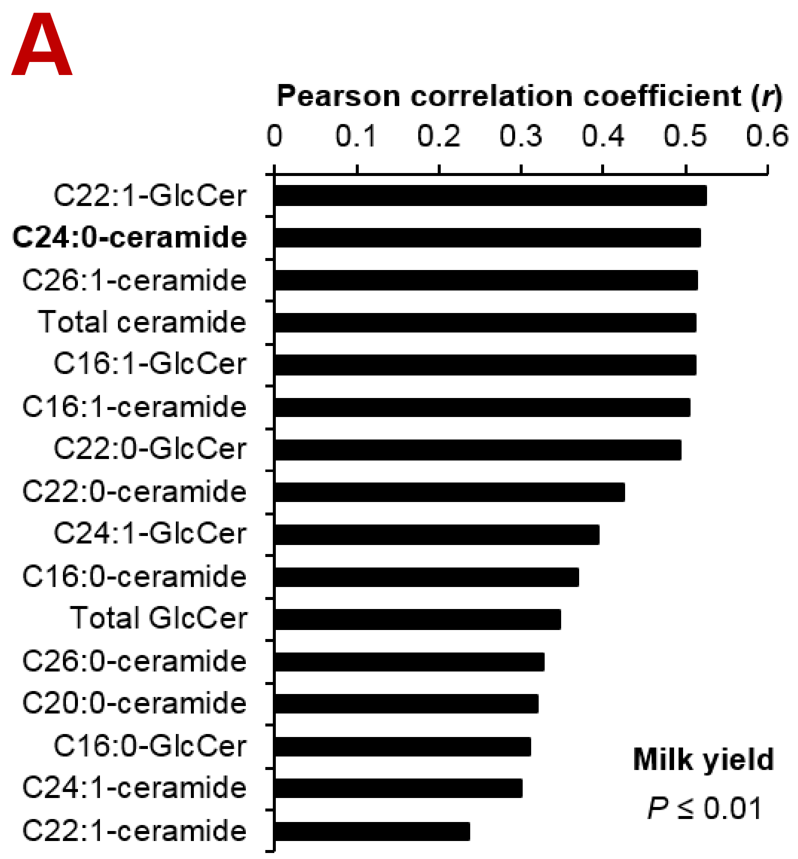
Cornell CALS College of Agriculture and Life Sciences

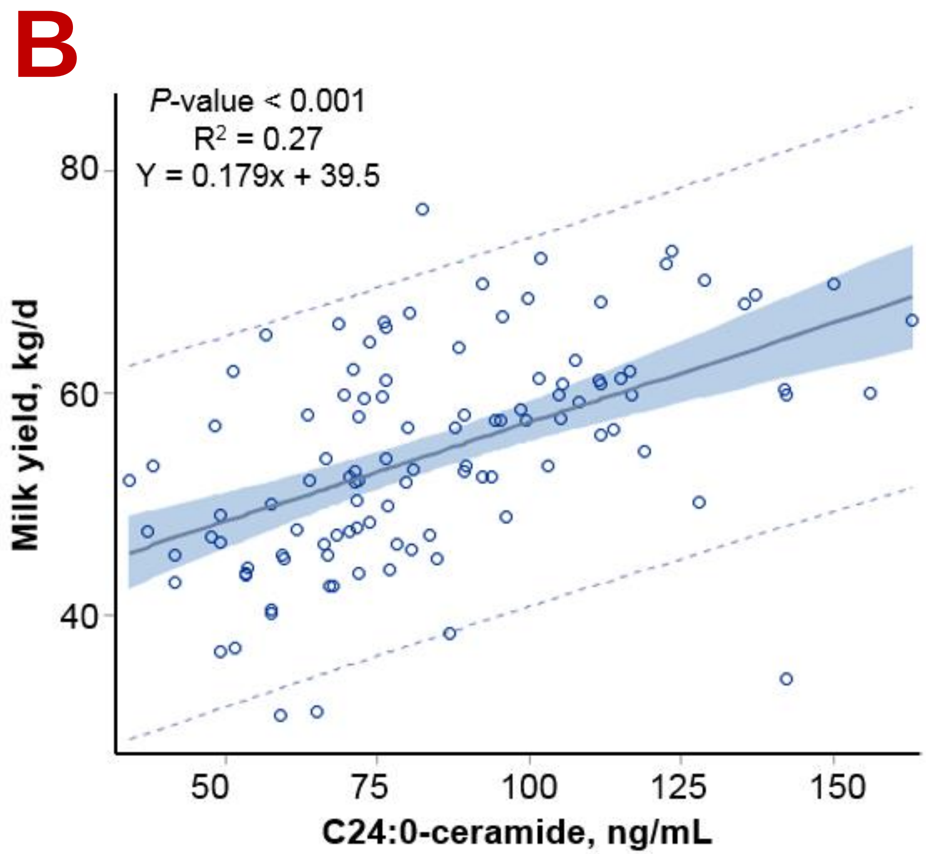
Partial correlations in the pre-partum period

Lipid classes	DMI	RFI	Parity	Calf BW	Milk yield	ECM	Fat	Protein	Lactose
Sphingolipids									
Ceramide	-0.07	0.19	-0.34	-0.17	**0.48***	-0.35	0.09	-0.03	-0.06
Monohexosylceramide	-0.32	-0.28	0.17	0.06	-0.35	-0.21	0.07	0.24	-0.01
Fatty Acylglycerols									
Monoalkyldiacylglycerol	-0.23	-0.26	0.27	0.2	-0.33	-0.07	0.24	0.40*	-0.29
Diacylglycerol	-0.32	-0.36	0.32	0.15	-0.29	-0.04	0.27	0.3	-0.24
Triacylglycerol	-0.2	-0.2	0.22	0.2	-0.35	-0.13	0.17	0.39*	-0.29
Glycerophospholipids									
Phosphatidylcholine	**-0.54***	**-0.40***	-0.23	0.24	-0.31	-0.40*	-0.21	0.01	0.11
Phosphatidylethanolamine	**-0.45***	**-0.43***	0.06	0.23	-0.1	-0.12	-0.11	0.19	-0.01

***$P \leq 0.01$; **$P \leq 0.05$; *$0.05 < P \leq 0.10$

Circulating ceramide is consistently positively related to milk yield





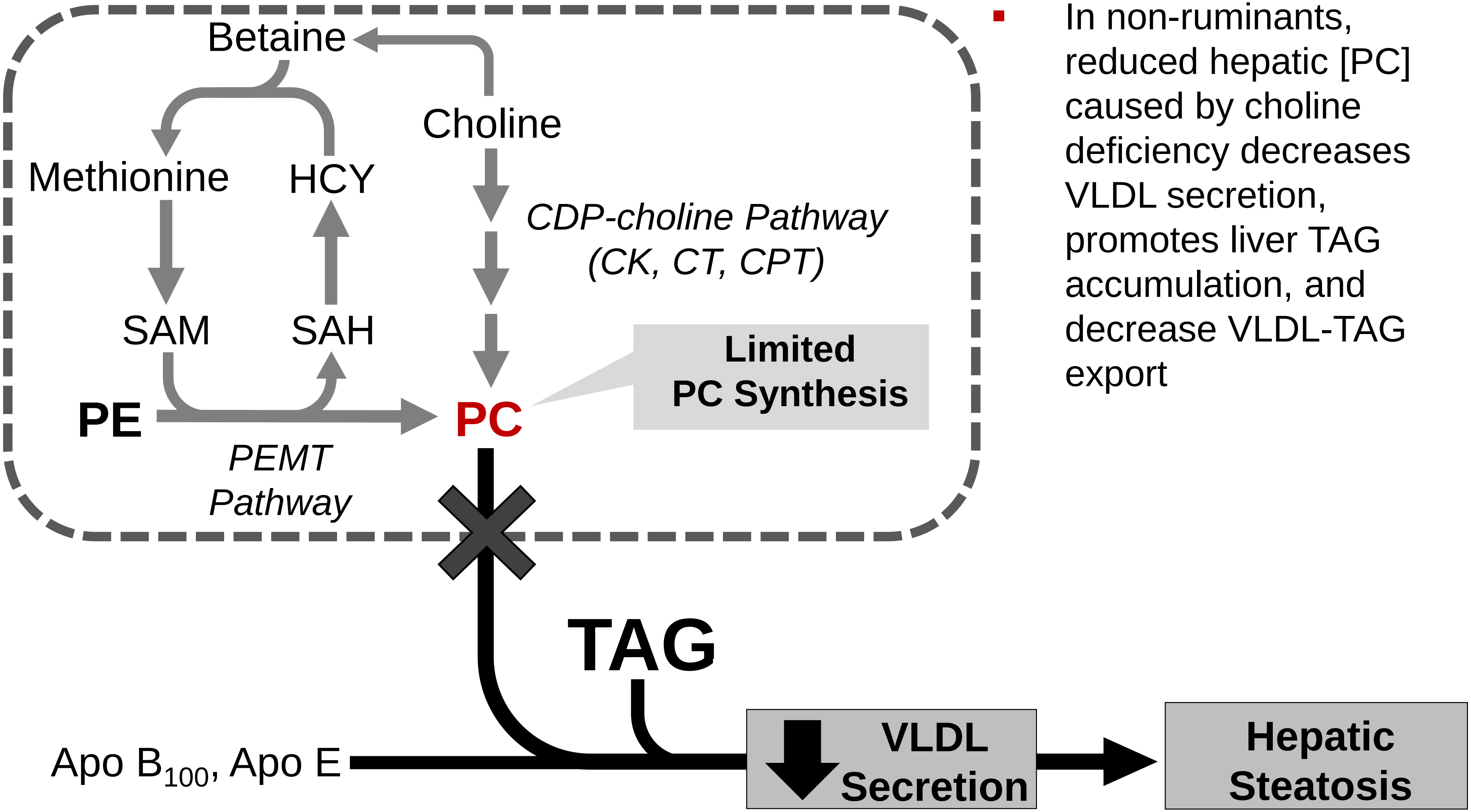
Early lactation; Plasma; N = 37; Rico et al., 2016, 2017 (ADSA abstract); Davis et al., 2017 (ADSA abstract)

Partial correlations in the pre-partum period

Lipid classes	DMI	RFI	Parity	Calf BW	Milk yield	ECM	Fat	Protein	Lactose
Sphingolipids									
Ceramide	-0.07	0.19	-0.34	-0.17	**0.48*** **	-0.35	0.09	-0.03	-0.06
Monohexosylceramide	-0.32	-0.28	0.17	0.06	-0.35	-0.21	0.07	0.24	-0.01
Fatty Acylglycerols									
Monoalkyldiacylglycerol	-0.23	-0.26	0.27	0.2	-0.33	-0.07	0.24	0.40*	-0.29
Diacylglycerol	-0.32	-0.36	0.32	0.15	-0.29	-0.04	0.27	0.3	-0.24
Triacylglycerol	-0.2	-0.2	0.22	0.2	-0.35	-0.13	0.17	0.39*	-0.29
Glycerophospholipids									
Phosphatidylcholine	**-0.54*** **	**-0.40* **	-0.23	0.24	-0.31	-0.40*	-0.21	0.01	0.11
Phosphatidylethanolamine	**-0.45*** **	**-0.43*** **	0.06	0.23	-0.1	-0.12	-0.11	0.19	-0.01

***$P \leq 0.01$; **$P \leq 0.05$; *$0.05 < P \leq 0.10$

Glycerophospholipids and fat export through the liver



- In non-ruminants, reduced hepatic [PC] caused by choline deficiency decreases VLDL secretion, promotes liver TAG accumulation, and decrease VLDL-TAG export

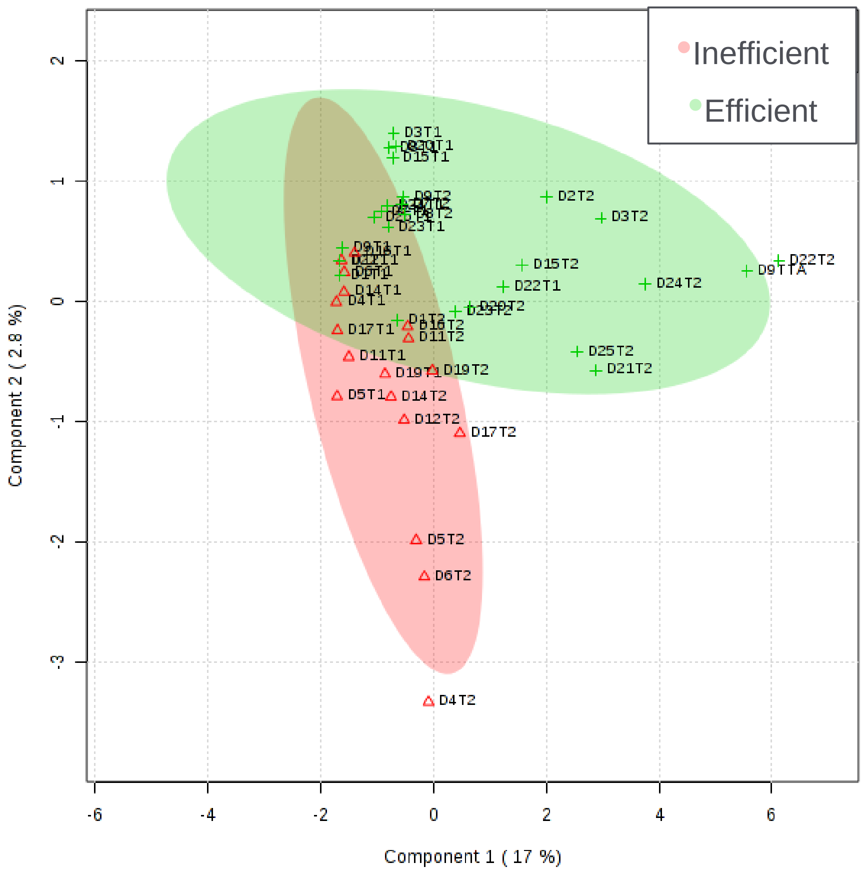
Cole et al., 2012

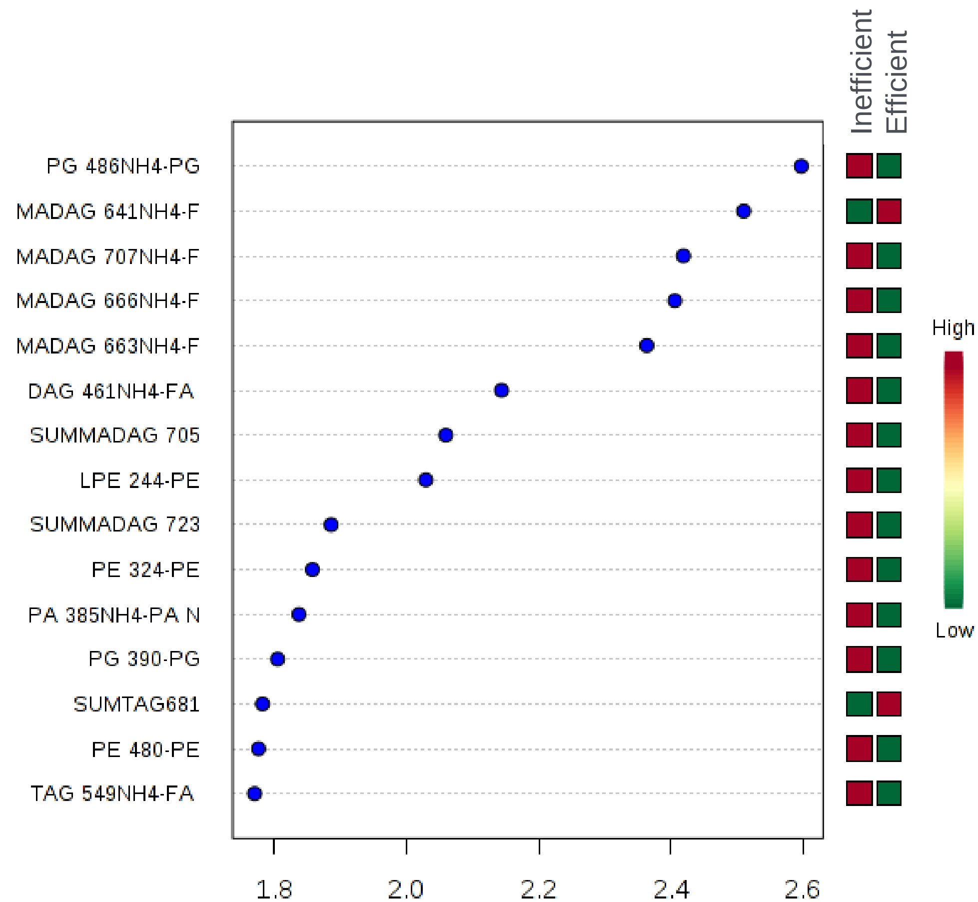
Partial correlations in the post-partum period

Lipid Classes	DMI	RFI	Parity	Calf BW	Milk yield	ECM	Fat	Protein	Lactose
Sphingolipids									
Ceramide	0.09	0.08	-0.08	0.13	-0.21	-0.03	0.09	0.38*	0.12
Monohexosylceramide	-0.27	-0.23	0.40*	0.15	-0.18	-0.11	0.06	0.16	-0.02
Fatty Acylglycerols									
Monoalkyldiacylglycerol	-0.25	-0.26	0.3	0.1	0.09	0.04	-0.03	-0.08	0.14
Diacylglycerol	-0.22	-0.2	0.31	0.2	-0.37	-0.19	0.09	**0.43*** **	-0.21
Triacylglycerol	-0.25	-0.25	0.33	0.11	0.08	0.02	-0.05	-0.07	0.2
Glycerophospholipids									
Phosphatidylcholine	-0.1	-0.06	-0.15	0.06	0.32	0.07	-0.21	-0.37*	**0.42*** **
Phosphatidylethanolamine	-0.05	-0.05	-0.14	-0.05	0.36*	0.12	-0.2	-0.41*	0.37*

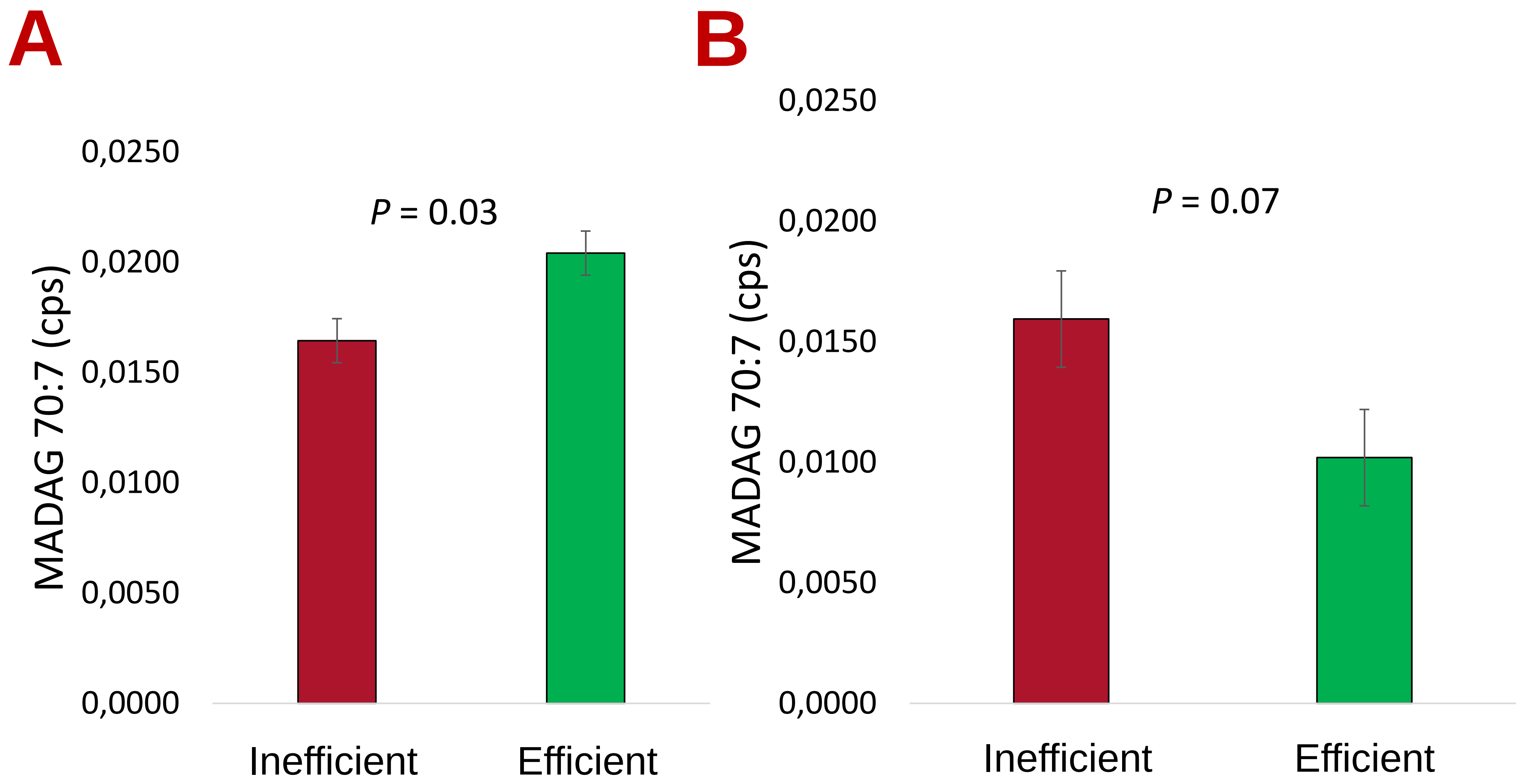
***$P \leq 0.01$; **$P \leq 0.05$; *$0.05 < P \leq 0.10$

Scores plots and VIP scores

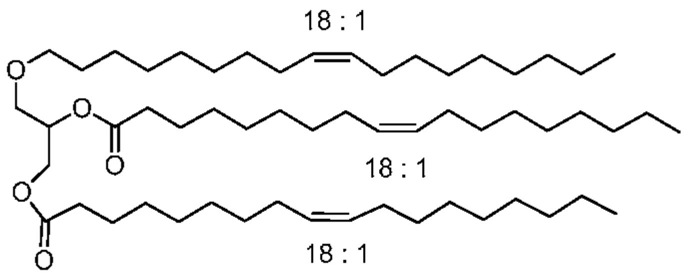




Timing of fat mobilization



Timing of fat mobilization



MADAG

Diacylglycerol
(e.g. C16:0/C18:1 DAG)

↓

Triacylglycerol
(e.g. C16:0/C18:1/C18:0 TAG)

Storage

Erion and Shulman , 2010; Newgard, 2012;
Summers, 2015; Ma et al., 2017

Summary of Findings

1. Distinct hepatic lipidomic profiles were identified in the present study
2. The negative associations between PCs and PEs with RFI might support improved fat export through the liver
3. Efficient animals displayed a pattern of elevated MADAGs in pre-partum and lowered values post-partum

Implications

- Considering our results, timing of fat mobilization might be of great importance in the context of feed efficiency. The differences in the hepatic lipidomic profile of cows with divergent efficiencies highlight new opportunities in the development of efficiency biomarkers related to lipid metabolism

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Questions?

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