Mjölkfett och det metabola syndrom: Vad betyder utfodringen?

Lars I Hellgren

Nutritional Immunology Group, Center for Biological Sequence Analyses, Technical University of Denmark

e-mail lih@bio.dtu.dk

DTU Biosys

Department of Systems Biology

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Agenda

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- **Background:**
  - Metabolic syndrome
  - The link between hepatic fat accumulation and metabolic syndrome
- **Our first study**
  - Dairy fat vs. high PUFA
- **Explaining the data**
  - Is there something protective in dairy fat?
- **Brief overview of the literature with our initial results**
- **Does the feeding regime affect the nutritious properties of dairy fat?**
  - "Tailored milk and Human Health" - A Danish National Research Initiative

Diseases of affluence

Metabolic syndrome

Obesity (in particular abdominal obesity)

- Insulin resistence
- Increased level of blood triacylglycerol
- Decreased levels of HDL (High density lipoprotein)
- High blood pressure

Inflammation

(de Luca & Olefsky (2008) ”Inflammation and Insulin resistance” , FEBS Letters **582:** 97-105)

Metabolic syndrome

Present model

Expression of inflammatory mediators (e.g. TNF-α, PAI-1 etc) in the adipose tissue

↓ Increased lipolysis

Release of non-esteried fatty acids and TNF- α

In the liver:

- Increased uptake of fatty acids
- Increased fatty acid synthesis
- Decreased fatty acid oxidation

→ **Hepatic lipid deposition**

Triacylglycerol
Ceramide
Diacylglycerol

↑ Increased concentrations

Ceramide and Diacylglycerol inhibit signalling from the insulin receptor

Rasouli *et al.* (2007) Diabetes Obesity & Metabolism **9:**1-10; Kotronen&Yki-Järvinen (2008) Arterio. Thromb. & Vasc. Biol. **28:**27-38; Holland & Summers (2008) Endocr. Rev. **29:**381-402

Dairy fat vs. Vegetable PUFAs

- **8 male Sprague-Dawley rats per group.**
- **Semisyntetic diet with 31E% fat for 14 weeks.**
  - Butter + 2% grapeseed oil
  - Grapeseed oil

Parameters:

- General physiological parameters in serum
- Glucose tolerance (OGTT after 9 and 13 weeks)
- Ectopic lipid deposition (TAG, NEFA and Ceramide content in liver, skeletal muscles and the heart)
- Fatty acid composition in PL and TAG in liver, skeletal muscles, adipose tissue and the heart

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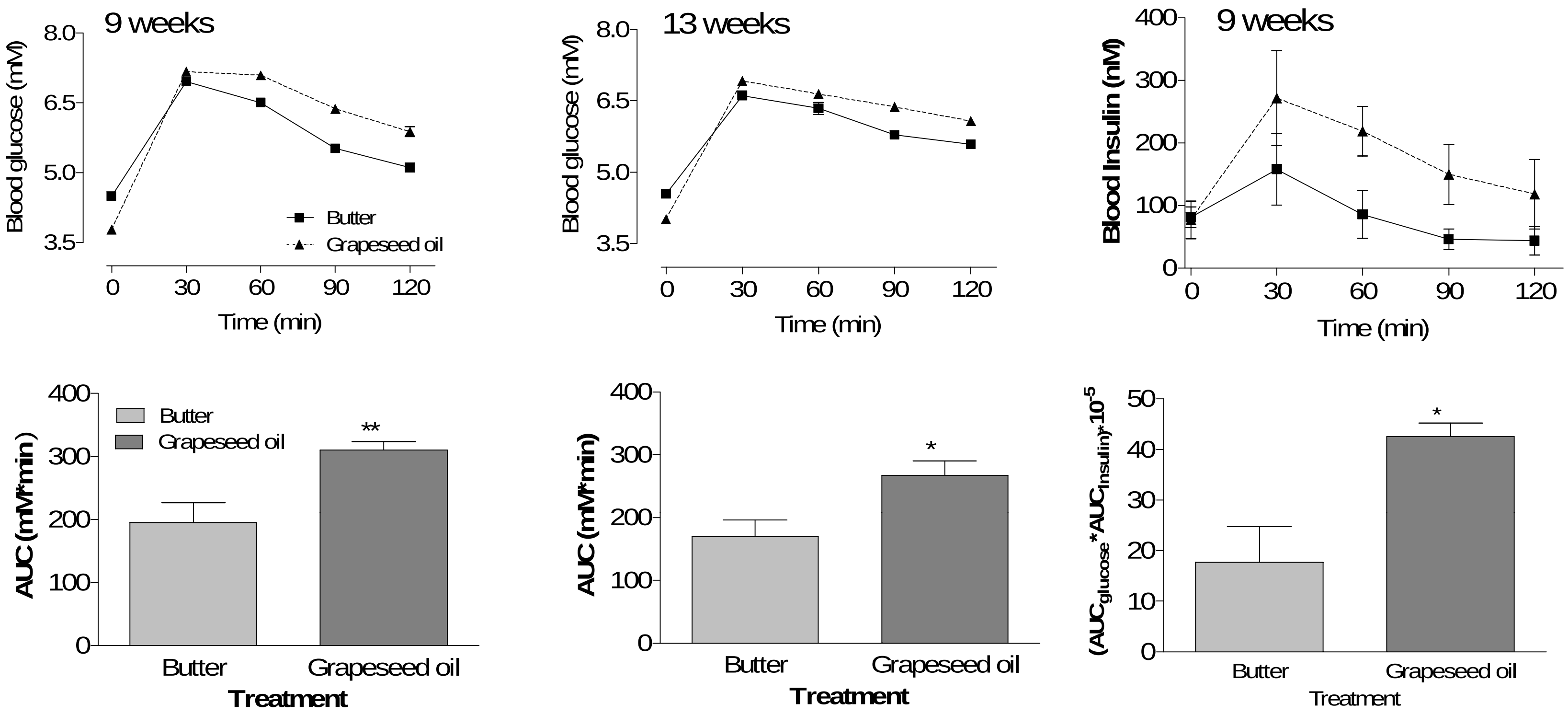
Results from OGTT

Oral glucose-tolerance test were performed after 9 and 13 weeks on the diets.



Rats were given 1.33g glukose/g body-weight at t=0. n=8, * $p<0.05$; ** $p<0.01$

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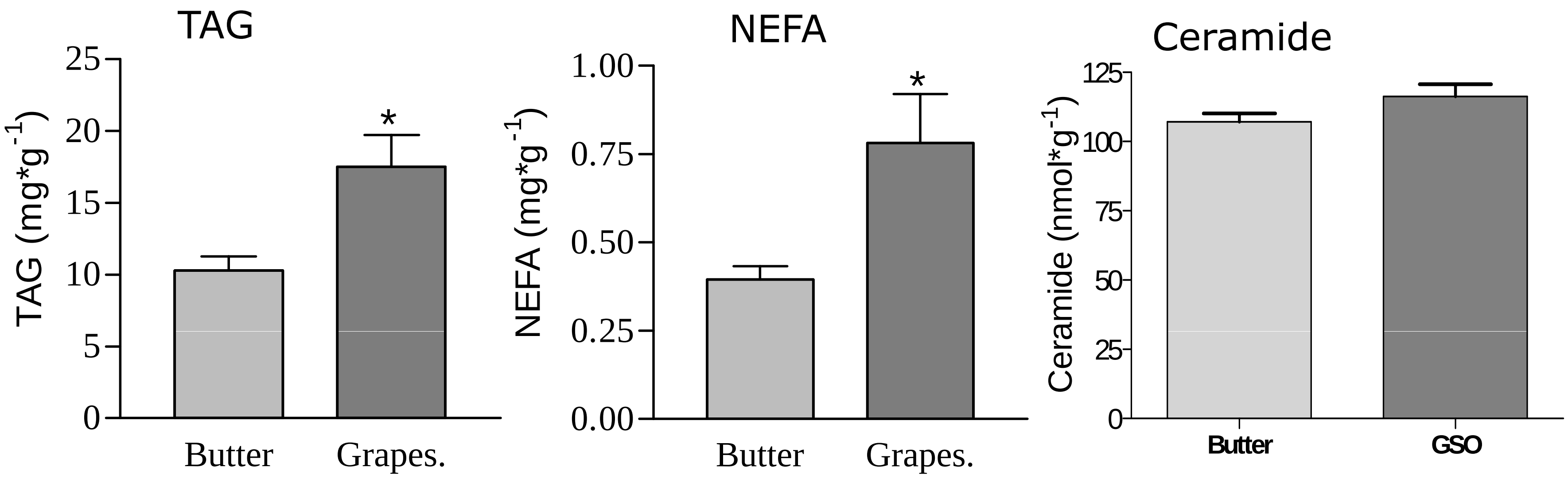
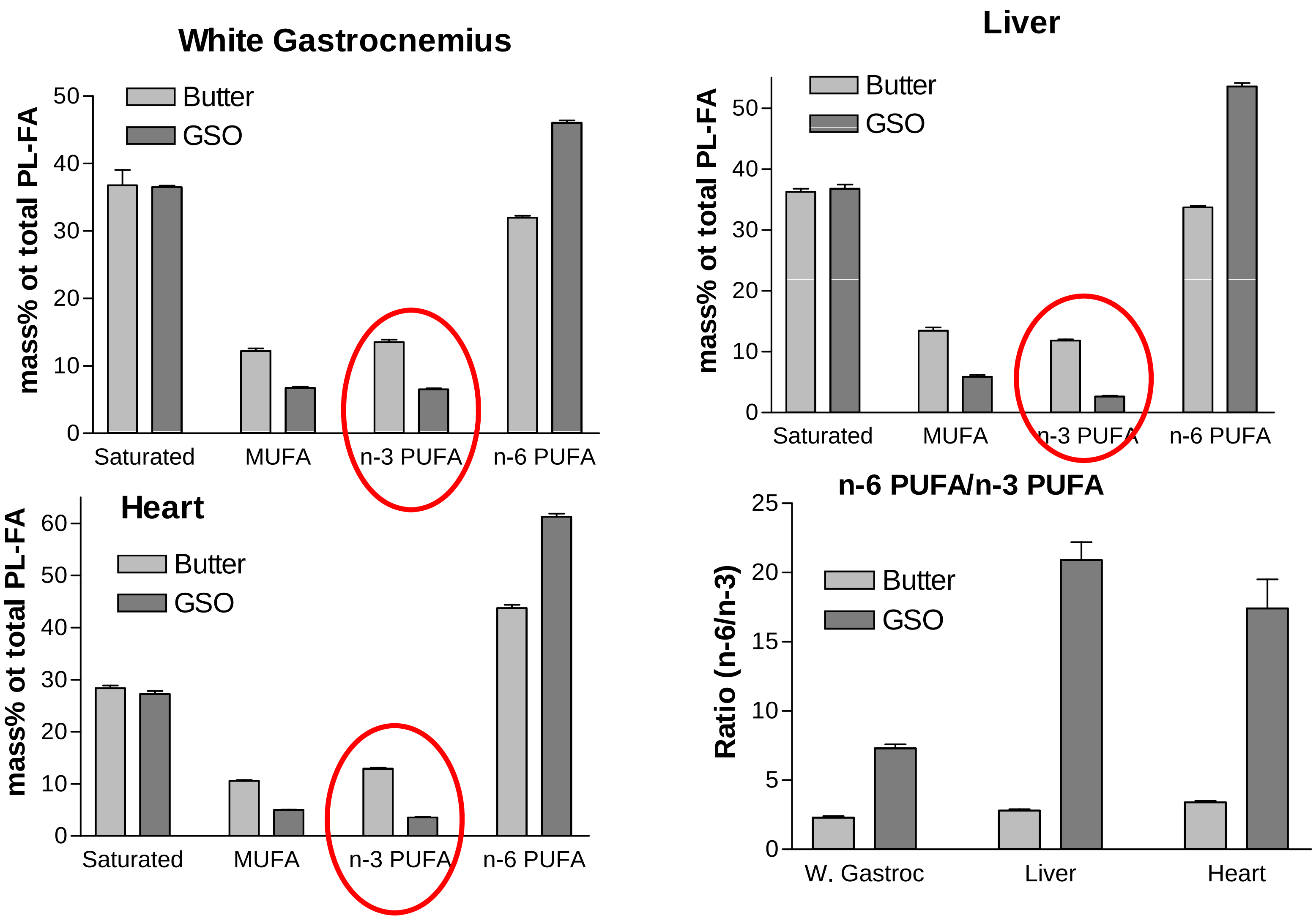
Hepatic lipid deposition

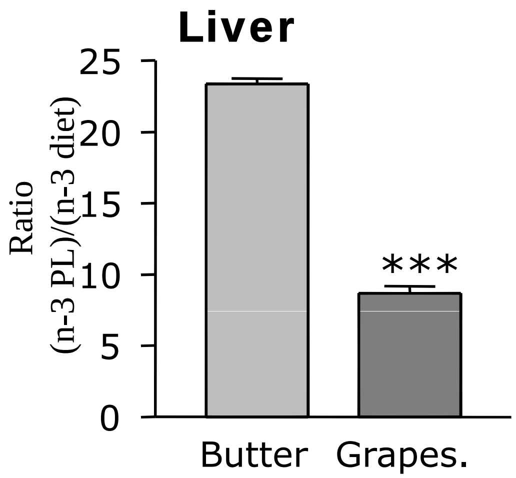


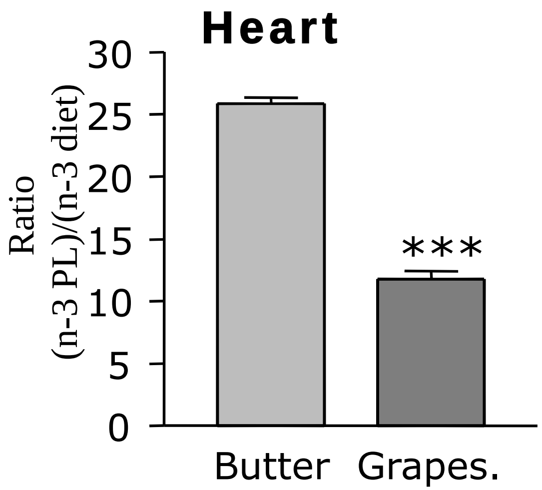
Intake of butter as the dominating fatty acid source has a positive impact on glucose tolerance and ectopic fat depositions, than a highly polyunsaturated vegetable oil!

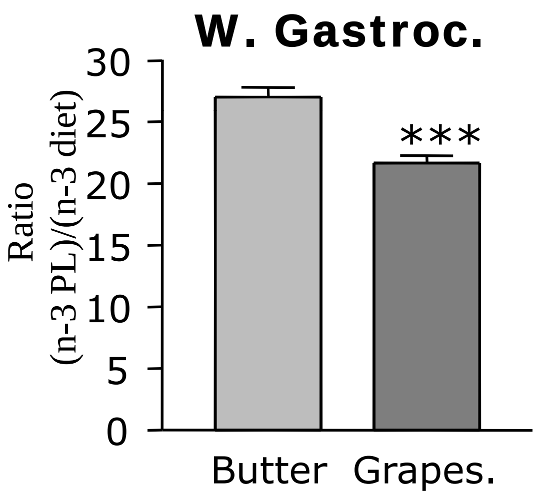
Is there something protective in dairy fat?

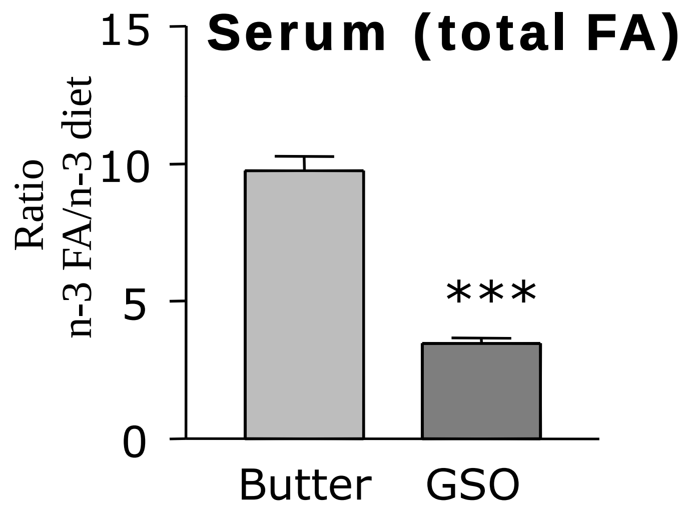


Efficiency in transformation of α-linoleic acid

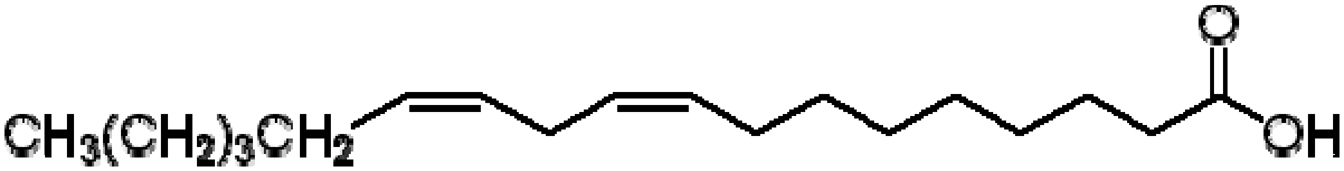




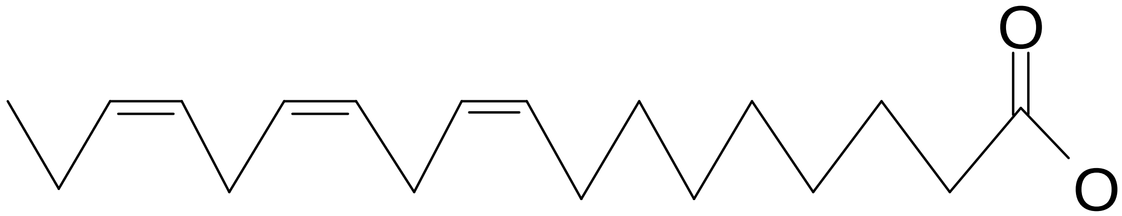




Is dairy PUFA relevant in a normal diet?

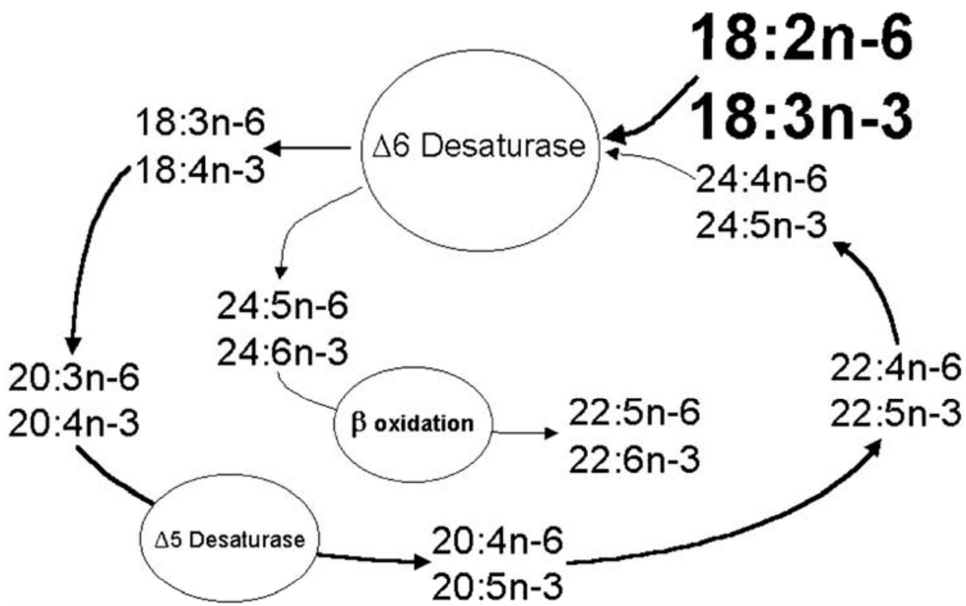


Linoleic acid (*cis*-9, *cis*-12, -Octadecadienoic acid)



α-linolenic acid (cis-9, *cis*-12, *cis*-15-Octadecatrienoic acid

Is dairy PUFA relevant in a normal diet?



Blank, C. et al. J. Lipid Res. 2002;43:1537-1543

Linoleic and α-linolenic compete for the same enzyme systems in elongation to the long-chained DHA and EPA.

Intake of α-linolenic from milk in normal Danish diet; ~150 mg.

Possible with strategic feeding (and same milk-fat intake) ~ 450 mg

Total intake of PUFA in the Danish population ~**12 g/day** (*Danskernes Kostvaner 2000-2002, www.food.dtu.d*k)

Is dairy PUFA relevant in a normal diet?

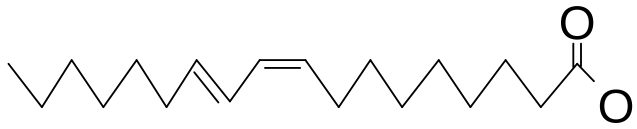
No, probably not!

What about other bioactive fatty acids?

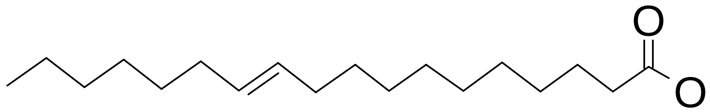
Fatty acids in milk and the metabolic syndrome

PUFA (linoleic & α-linolenic acid; C18:2 (9^c,12^c) & C18:3 ($^{all\ cis}$9, 12, 15)

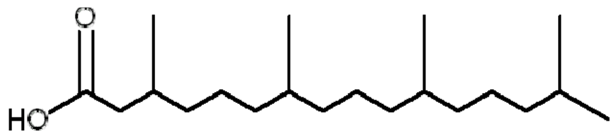
Conjugated Linoleic acid (C18:2 (9^c, 11^t))



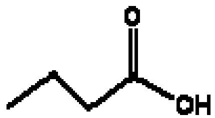
***trans*-Vaccenic acid (**C18:1 (11^t))



Branched chained fatty acids (phytanic acid)

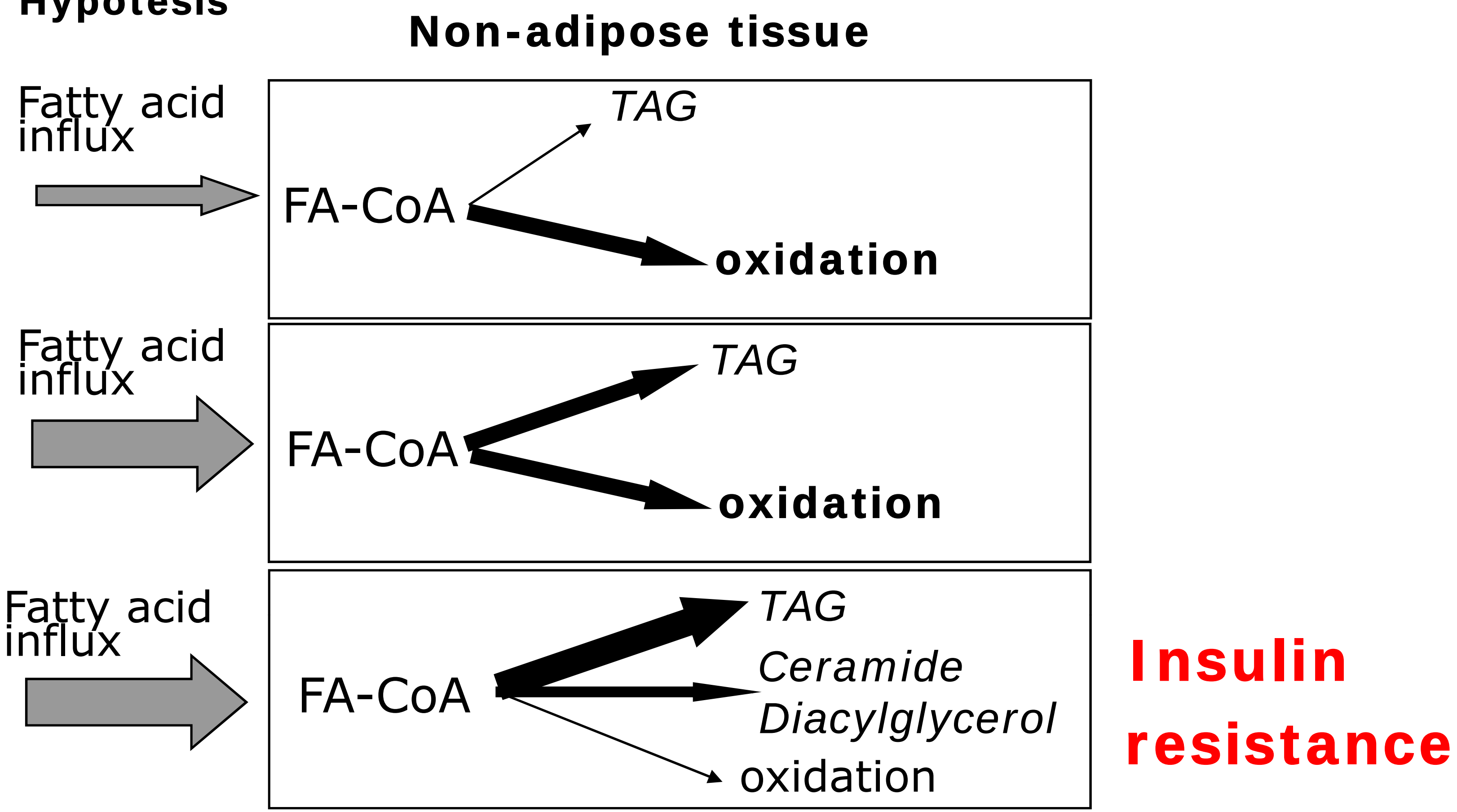


Short-chained fatty acids (butyric acid; C4:0)



Insulin resistance and lipid accumulation

Hypotesis



Is it possible to alter the rate of fatty acid oxidation through the diet?

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Control of fatty acid oxidation

- The balance between glucose and fatty acid oxidation is controlled via the Peroxisome Proliferating Activator Receptor (PPAR)- system.
- PPAR-α activated by fatty acids, form dimer with retinoid-X-receptor

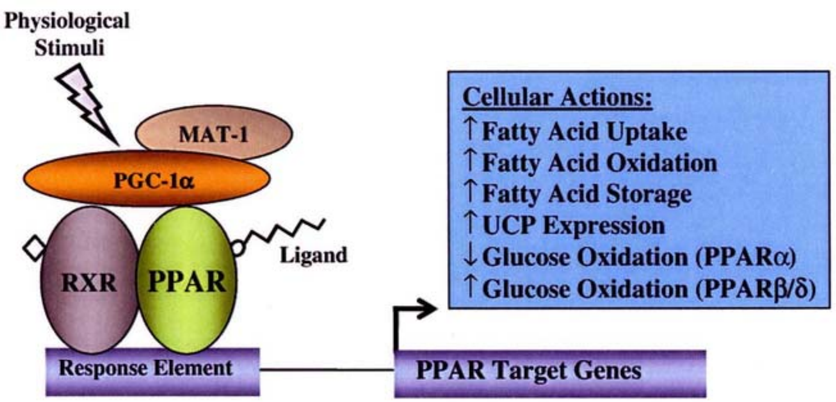
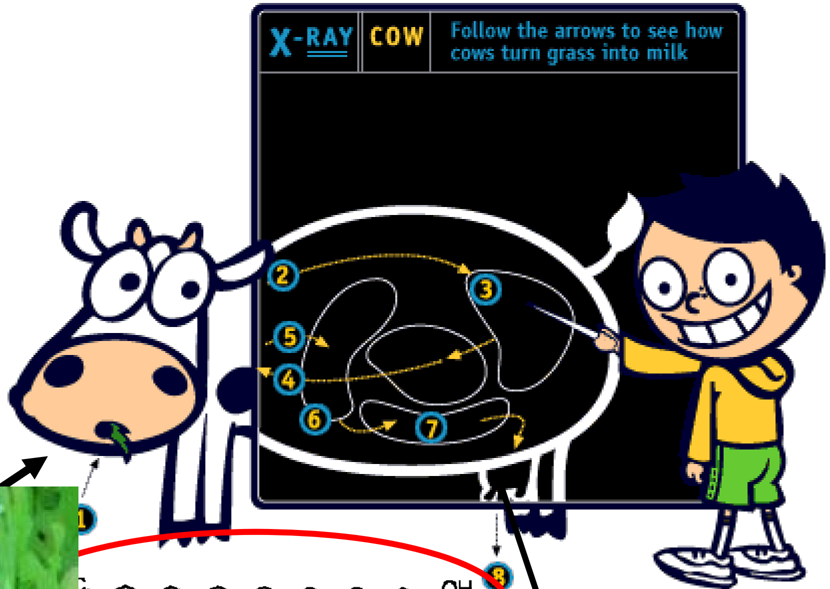
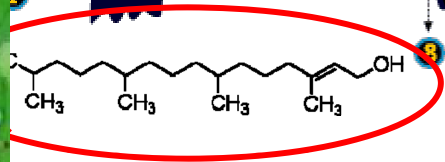


Fig. 1. The PPAR family of transcription factors forms dimers with RXRs and, upon binding cognate DNA response elements on target gene promoter regions, activates the transcription of target genes. The transcriptional activity of the PPARs is influenced by binding of endogenous ligands and protein coactivators such as PGC-1 and its regulator MAT-1. Some of the major actions of PPARs in the heart are shown in the box. Note that some of the actions are PPAR subtype-specific. UCP, uncoupling protein.

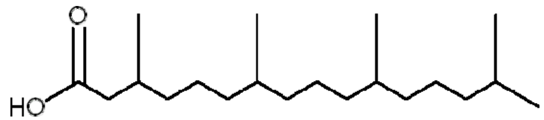
Picture from Madrazo &Kelly (2008) J. Mol. Cell. Cardiol **44:**968-75

Branched chained fatty acids – Overlooked bioactive fatty acids?





Phytol



Phytanic acid

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Effets of phytanic acid

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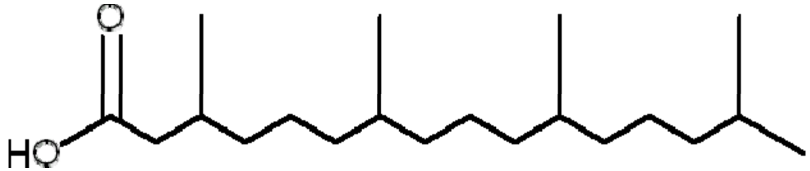


Phytanic acid

- Activates both PPAR-α and RXR (Lemotte&Keidel, 1996; Elinghaus & Wolfrum, 1999)

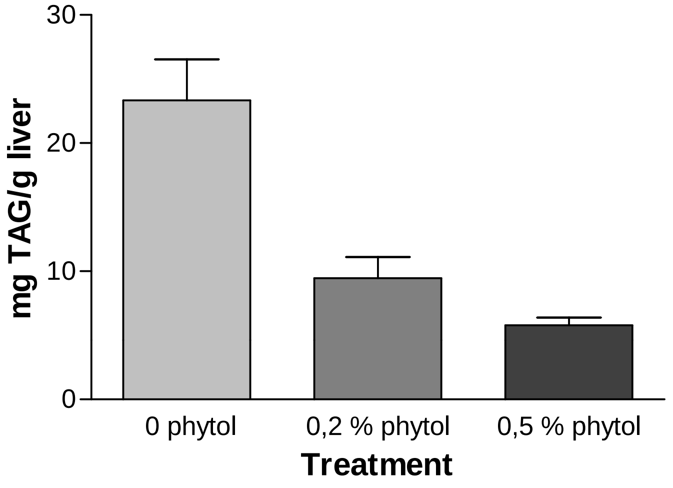
- Induces increased heaptic fatty acid oxidation (Hashimoto *et al.* 2006)

- Induce glucose uptake in hepatocyte and primary porcine muscle cells also at low μM concentrations (Heim *et al*, 2002; Ngum Che, Hellgren & Young, unpublished)

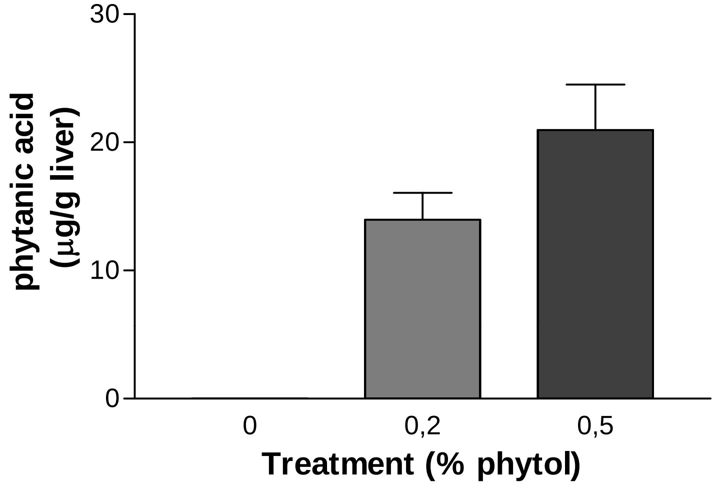
Effects; Initial studies

Experimental design: Mice (n=3) were fed a standard diet containing 0, 0.2 or 0.5 mass% phytol in diet for three weeks.

Hepatic TAG accumulation



Phytanic acid in liver



Hellgren, unpublished

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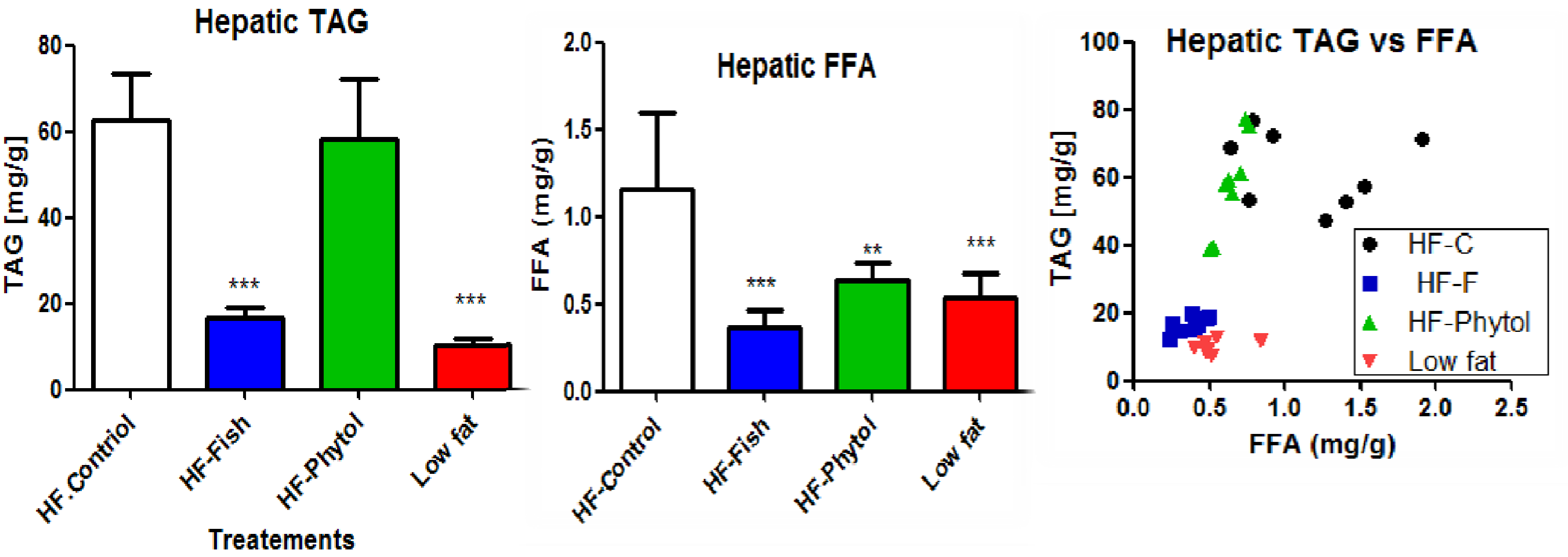
Effects in a high energy diet

Female C57bl/6 mice were fed 60E% fat and given 15% sucrose in drinking water for 5 weeks.

Fat: **HF- Control:** High oleic sunflower oil (78 % oleic acid, 12 % linoleic acid), **HF-fish** 50% fish oil/50% high oleic sunflower, **HF-phytol** = HF-control + **0.5% of fat as phytol**. **Low fat** = 7E% fat, no sucrose in drinking water

Fat in fish oil diet contained 8.4 % EPA and 5.7 % DHA



Hellgren, unpublished

What about the feeding regime ?

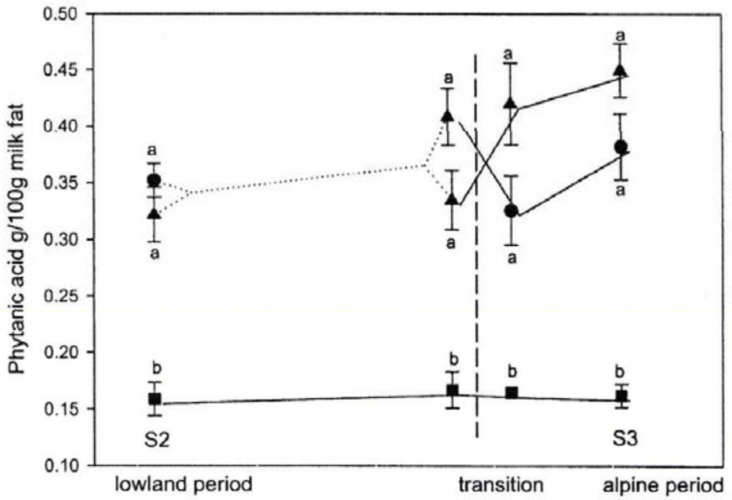


FIG. 6. Phytanic acid concentration in milk fat. S2, lowland period; S3, alpine period. ● Pasture group; ▲ indoor grass group; ■ control group; n = 6 per group. Dotted lines indicate that half of the cows were switched between pasture and indoor group. Dashed line marks the day of transport of grass-fed cows to the alpine location. Error bars represent SE. Different superscripts indicate significant differences at $P < 0.05$.

Leiber *et al (*2005) Lipids **40**:191-202

Phytanic acid concentration in milk is strongly dependent on the amount of chlorophyll containing feed.

Ensilage vs. Grass ???

Do short-chained fatty acids (SCFA) have systemic effects ?

There are specific receptors for short-chained fatty acids:

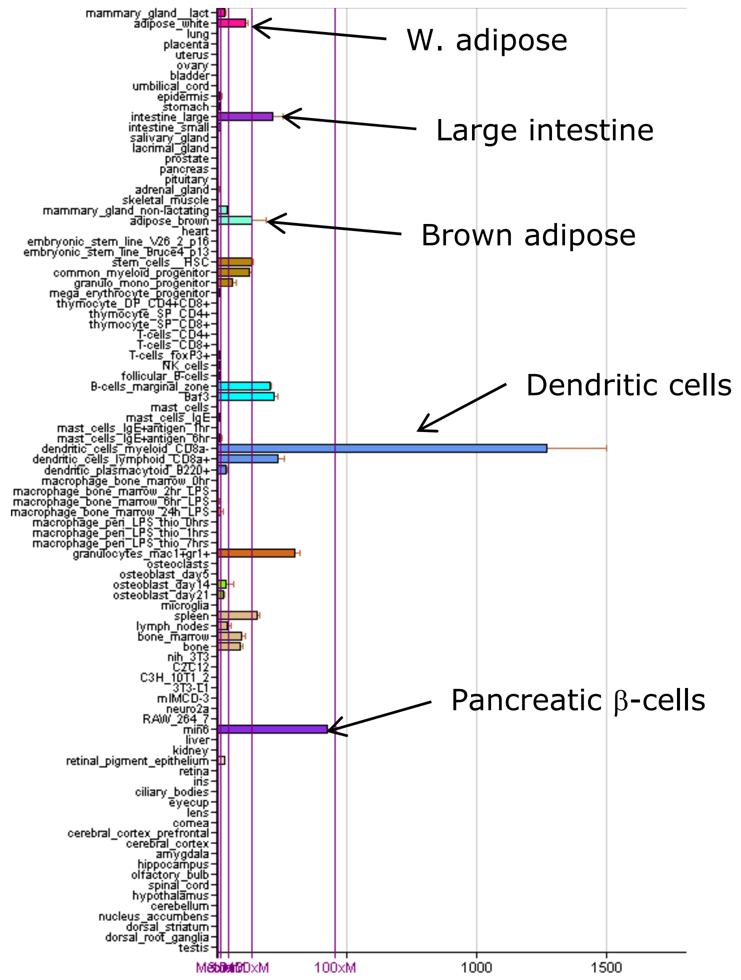
FFAR2 (GPC 41)

FFAR3 (GPC43)

FFAR2 is produced in adipose tissue and in the insulin-producing β-cells

→ SCFA most likely have a regulatory impact here!

FFAR2 expression



Tissue expression libraries from www.biogps.gnf.org

Described effects of SCFA via FFAR2 and FFAR3:

- Increased leptin release from adipocytes (Xiong *et al. 2004)*
- Induce adipogenesis (Hong *et al.* 2005)
- Induce expression of PPAR-α and reduce expression of NFkB in different cell types *in vitro (*Zapolska-Downar *et al 2004)*
- Intake of SCFA increas PPAR-a and reduce NFkB expression in both white adipose and the liver *in vivo* in mice (Pedersen & Hellgren, unpublished)

Bild 23 har utgått

Kontakta författaren Lars Hellgren
lih@bio.dtu.dk

Bild 24 har utgått

Kontakta författaren Lars Hellgren
lih@bio.dtu.dk

Short-chain fatty acids and feed?

- Lindmark Månsson (2008) (Food Nutr. Res. DOI: 10.3402/fnr.v52i0.1821)
  - No seasonal variation in milk delivered to Swedish Dairies
  - Variation in total mol% C4:0 and C6:0 from ~13 to 16 mol%

- Increased starch feeding, increase the amount of SCFA (see for example Bargo *et al 2005)*

- ***Is it possible to achieve both high PUFA/phytanic acid and high SCFA?***

Tailored milk and human health

- A national research initiative (Strategic Research Network) comprising of the projects:
  - *Milk with designated health effects* (Kristen Sejrsen, Århus Univ, Foulum)
  - *Green feed improves nutritional properties of the milk fat fraction* (Lars I. Hellgren, Tech. Univ. Denmark)
  - *Milk protein, obesity and the metabolic syndrome* (Christian Mølgaard, Copenhagen Univ-LIFE)
  - *Colostrum for gut protection and recovery* (Per Sangild, Copenhagen Univ-LIFE)
  - *Sund og velsmagende mælk baseret på græsmarksafgrøde* (Jacob Holm Nielsen, Århus Univ, Foulum)
  - *Cholesterol transport and milk protein* (Torben Ellebæk Pedersen, Århus Univ.)

Major funding from: The Danish Strategic Research Council, Danish Dairy Research Foundation, Danish Cattle Association, Thise Mejeri