U N I V E R S I T Y O F B E R G E N

Department of Clinical Science

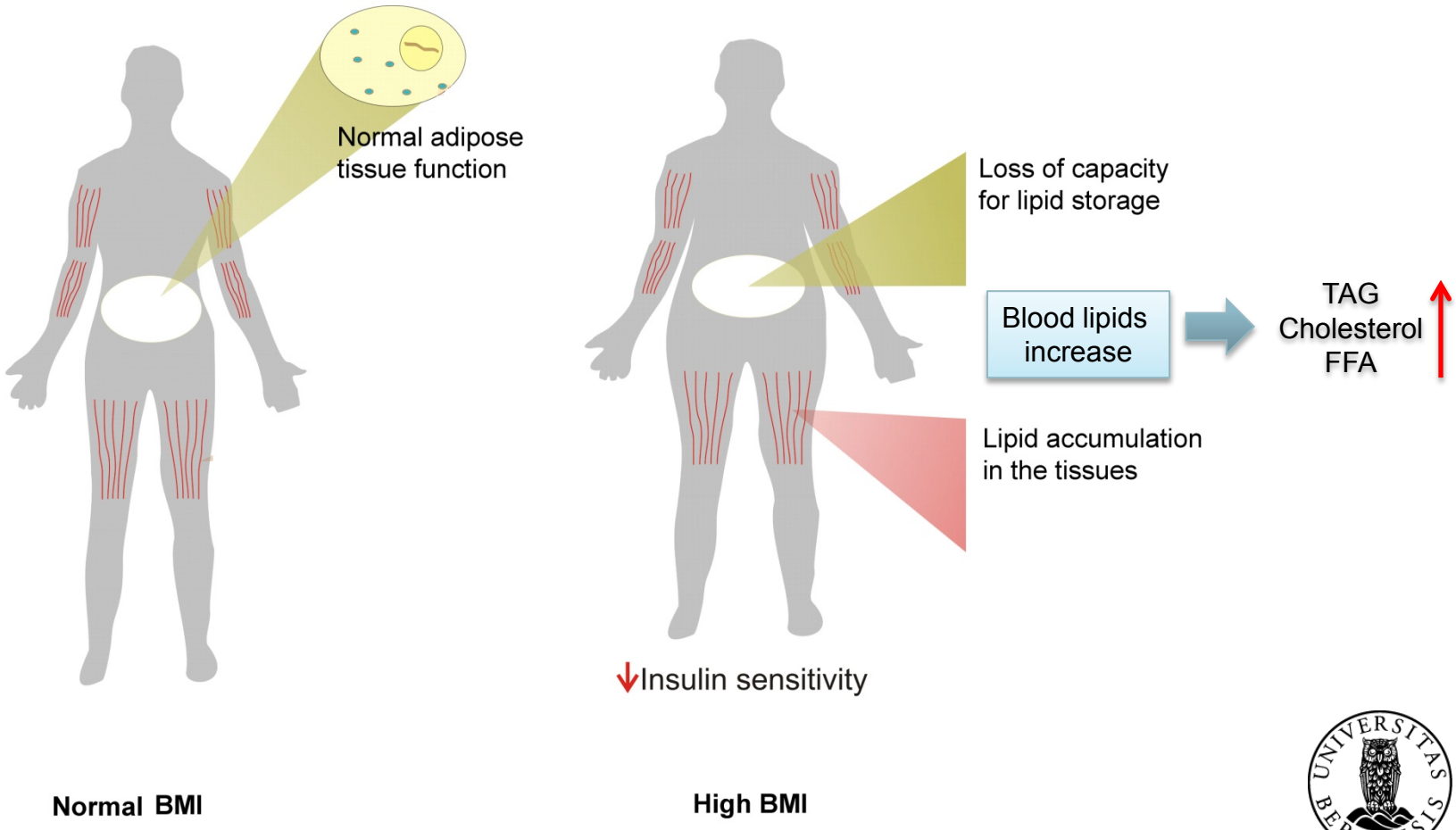
Phospholipids from Herring Roe
Pre-clinical and clinical data

Bodil Bjørndal, PhD

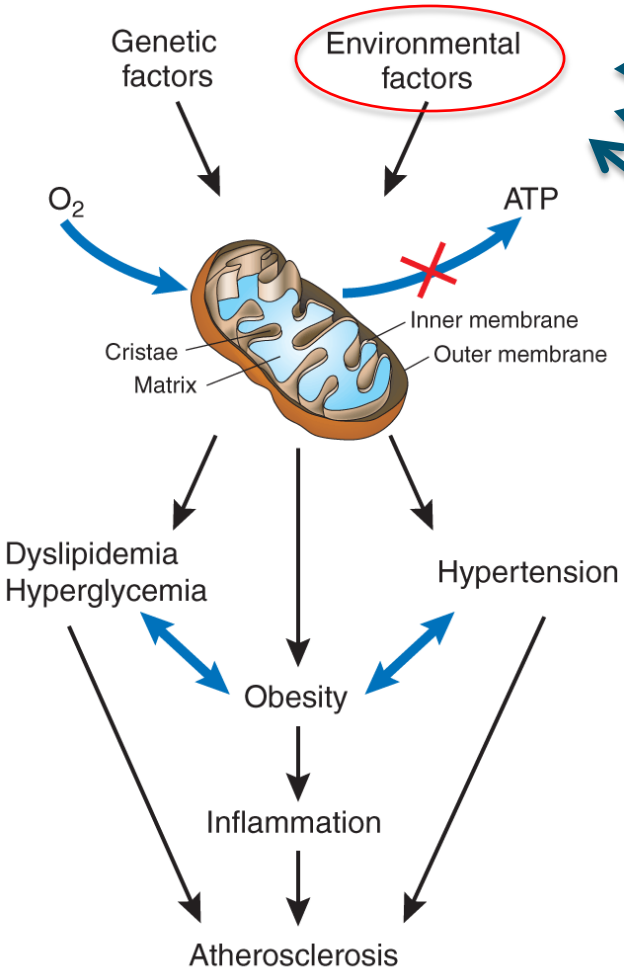
The Lipid Research Group

Department of Clinical Science, UiB

Obesity and the metabolic syndrome



Mitochondrial efficiency



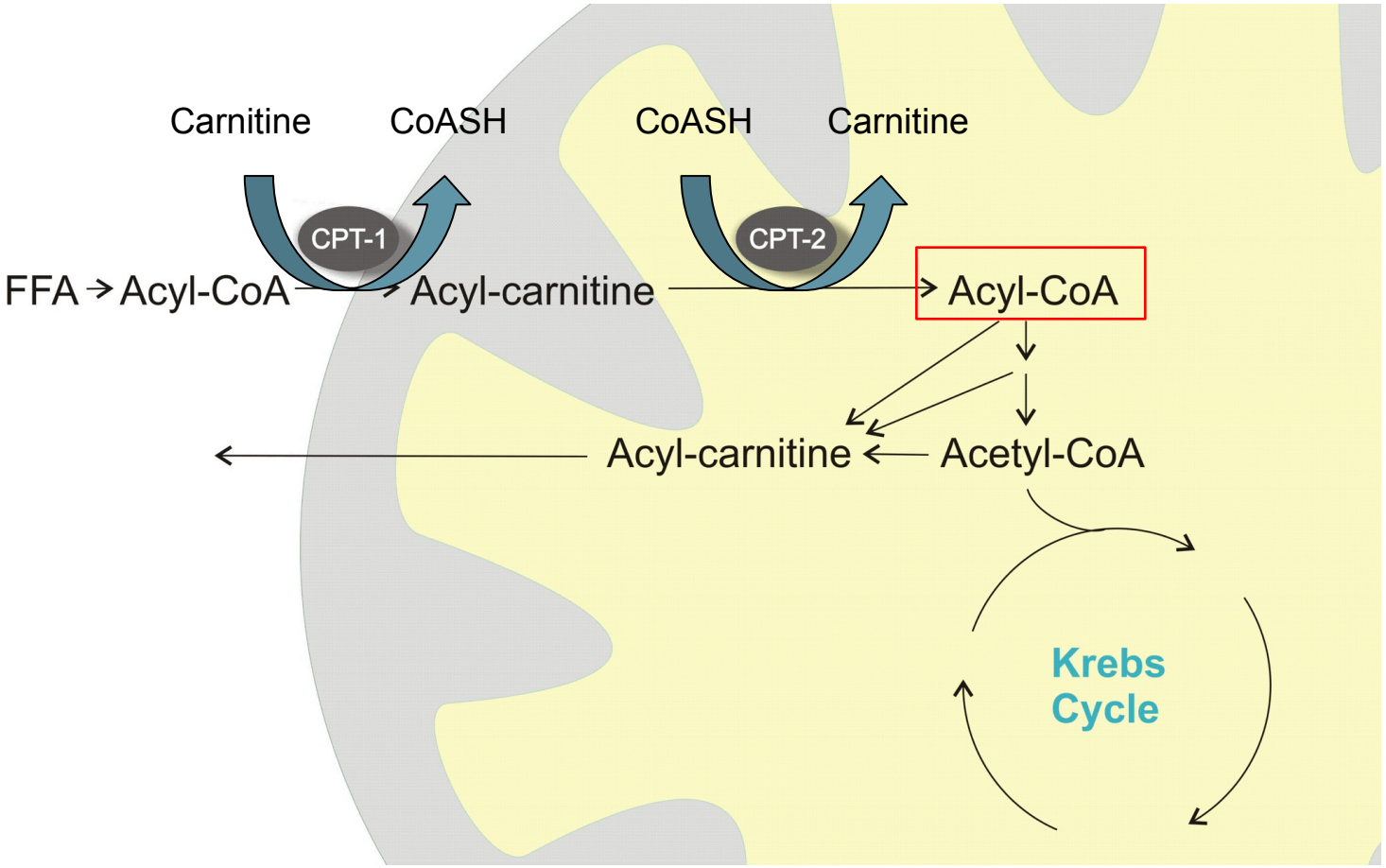
Diet / Bioactive compounds

Exercise

Drugs

Nature Medicine - ***12, 46 - 47 (2006)***
doi:10.1038/nm0106-46

Beta-oxidation and the carnitine shuttle



Ueland T, et al. ***Disturbed carnitine regulation in chronic heart failure - Increased plasma levels of palmitoyl-carnitine are associated with poor prognosis***. Int J Cardiol. 2013. **167**:1892-9.

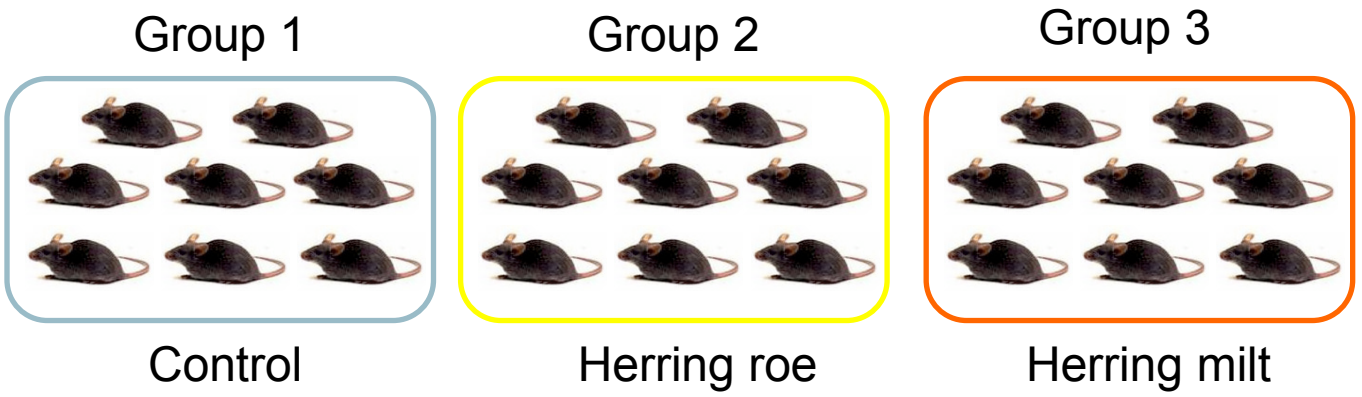
Marine ingredients and human health

- High fish intake – health benefits!

Can fish peptide fractions with specific bioactive properties be isolated?

Can lipid structure determine the effect of EPA/DHA-rich oils?

Studies in animals



Advantages:

- Defined diets
- Homogenous genetic background
- Minimal “lifestyle” influence
- Possible to study organ-organ interplay
- Transgenic mice models

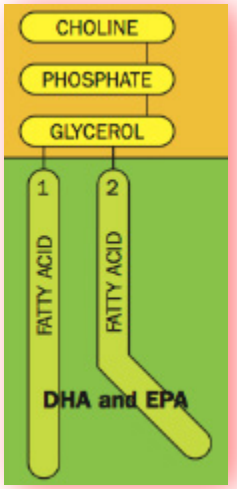
Disadvantages:

- Different lipoprotein composition rodents vs. humans
- Will the effects seen in animal studies result in actual health benefits in humans?

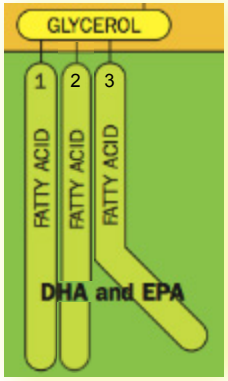
Marine phospholipids vs. fish oil

Herring roe

FISH OIL



Phospholipids
Polar, mixes with H_2O
Choline content



TAG
Non-polar, hydrophobic

Different structural forms can affect bioavailability, brain bioaccretion, incorporation into cells, susceptibility to peroxidation

Advantages of marine phospholipids

- Studies suggest that **PLs** is a more effective delivery form of n-3 PUFAs to body tissues than TAG
  - Liu et al. *J Lipid Res*. 2014, p531; Rossmeisl et al. *Plos ONE*. 2012, e38834; Wijendran et al. *Pediatr Res*. 2002, p265

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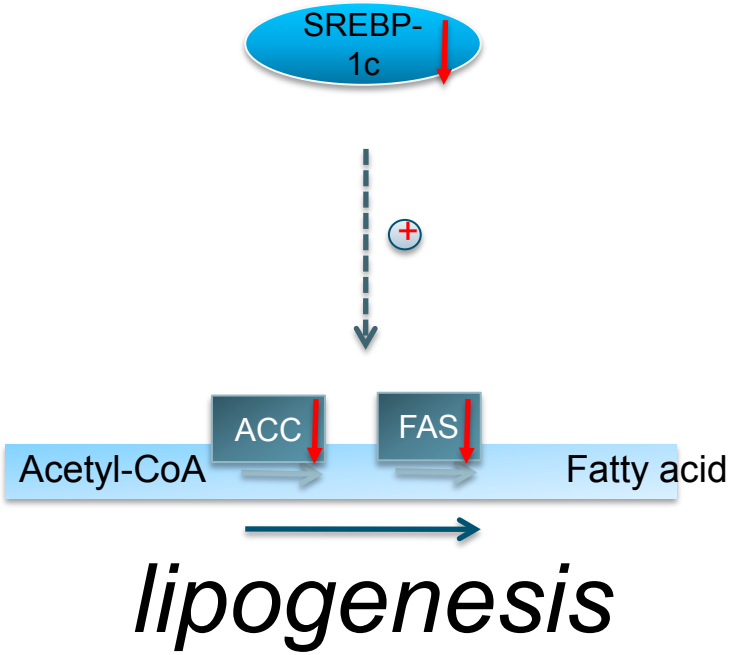
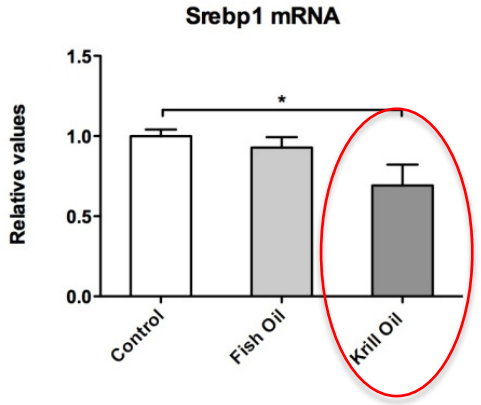
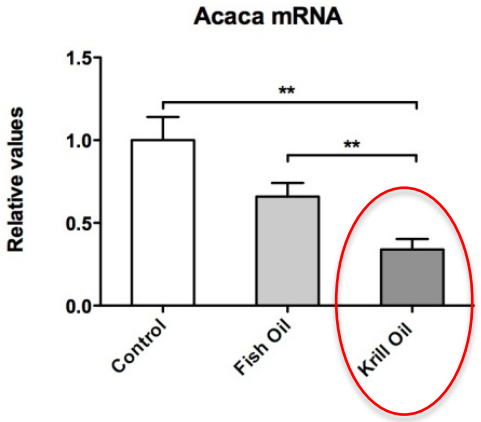
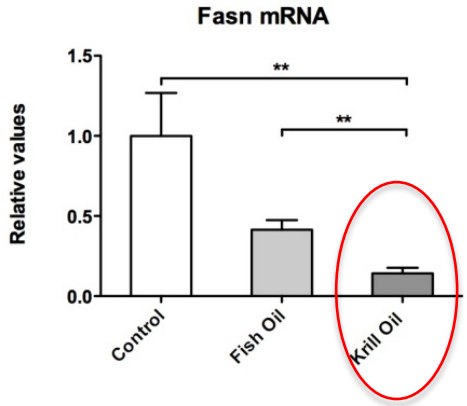
Fatty acid synthesis:









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  - Important for the syntesis of acetylcholine, PLs, transport of lipids and reductin of homocysteine.
  - Inadequate intake in several populations (Zeisel et al. Nutr Rev 2009, p615)

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Herring roe: Rich in PC and DHA (Bjørndal et al. *Lipids Health Dis*. 2013)

Eur J Nutr (2012) 51:741–753
DOI 10.1007/s00394-011-0254-8

ORIGINAL CONTRIBUTION

Dietary supplementation of herring roe and milt enhances hepatic fatty acid catabolism in female mice transgenic for hTNFα

Bodil Bjørndal · Lena Burri · Hege Wergedahl · Asbjørn Svardal · Pavol Bohov · Rolf K. Berge

Received: 13 July 2011 / Accepted: 26 September 2011 / Published online: 11 October 2011

Abstract
Purpose The beneficial effects of a seafood-rich diet are highly documented and can be attributed to both *n*-3 polyunsaturated fatty acids and other less studied nutritional components including protein and antioxidants. The reduced, hepatic TAG and plasma and hepatic cholesterol levels were increased by the herring diets. Both herring diets led to a substantial shift in the *n*-6:*n*-3 ratio in both liver and ovarian white adipose tissue. The herring diets also increased plasma carnitine and reduced the carnitine

Herring roe and milt in TNF-α transgenic mice

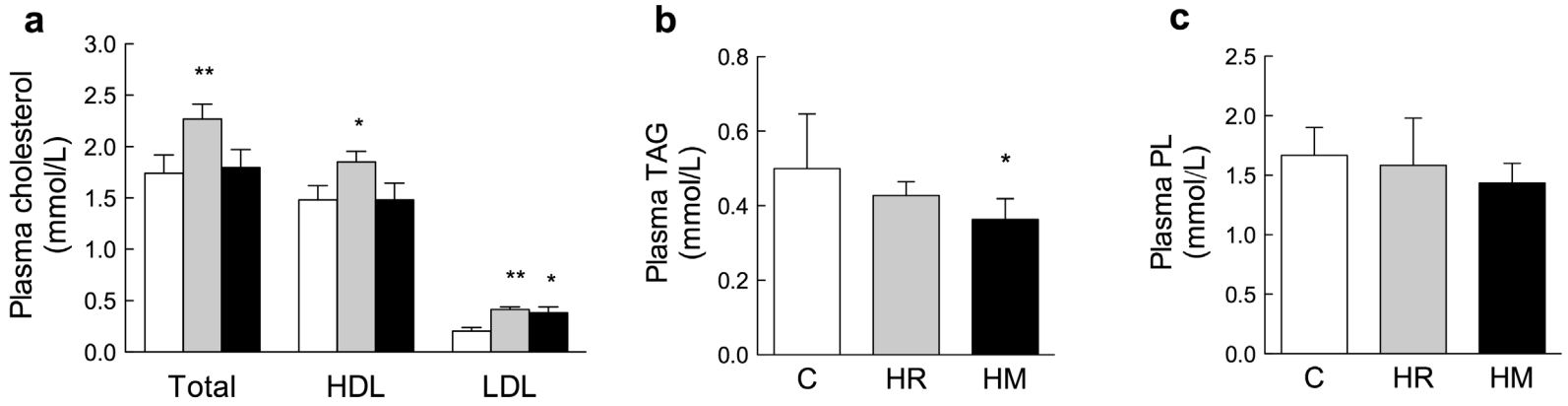
- Female TNF-α transgene C57BL/6 mice:
  - Control diet: 20% casein, 23% high-fat (lard)
  - Herring roe diet: 15% HR protein, 3.7% HR lipids
  - Herring milt diet: 15% HM protein, 1.3% HM lipids

  Animals were fed for 2 weeks (n = 6)

Illustration: The Jackson Laboratory

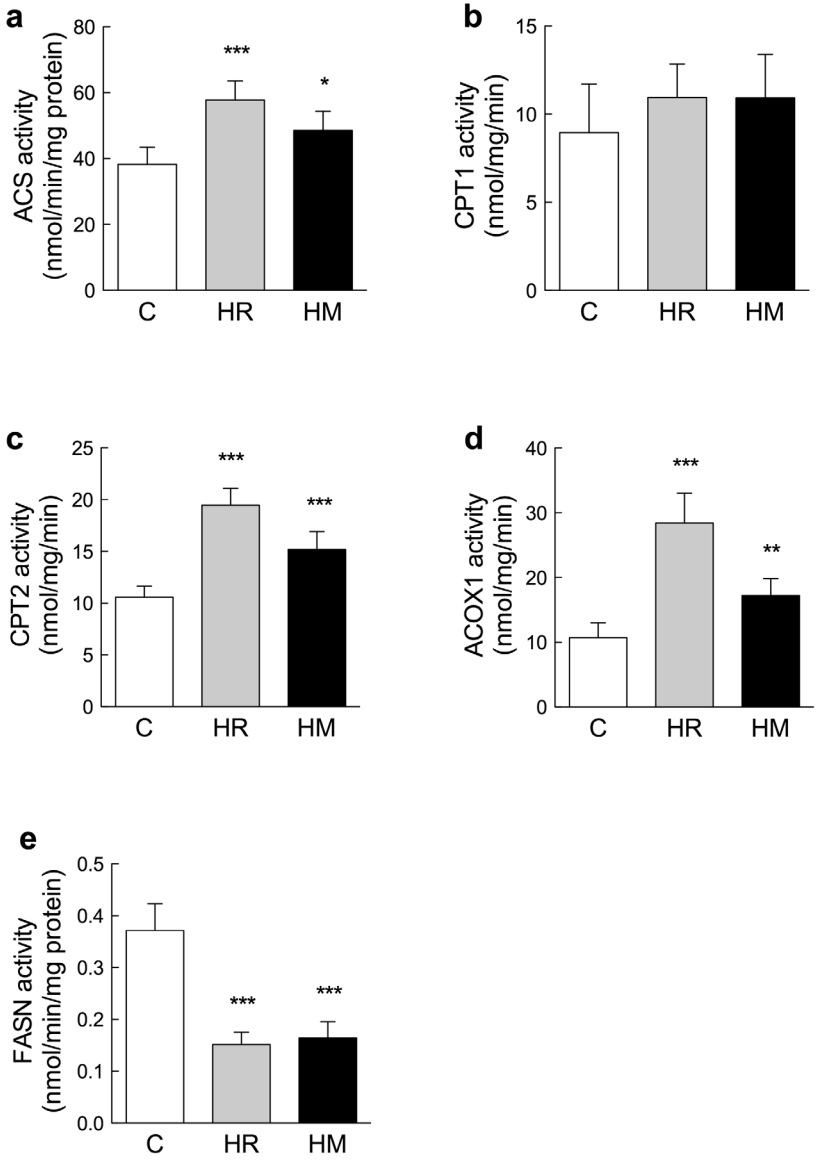
RESULTS: Plasma lipid levels

- TAG was only significantly reduced by the herring milt diet



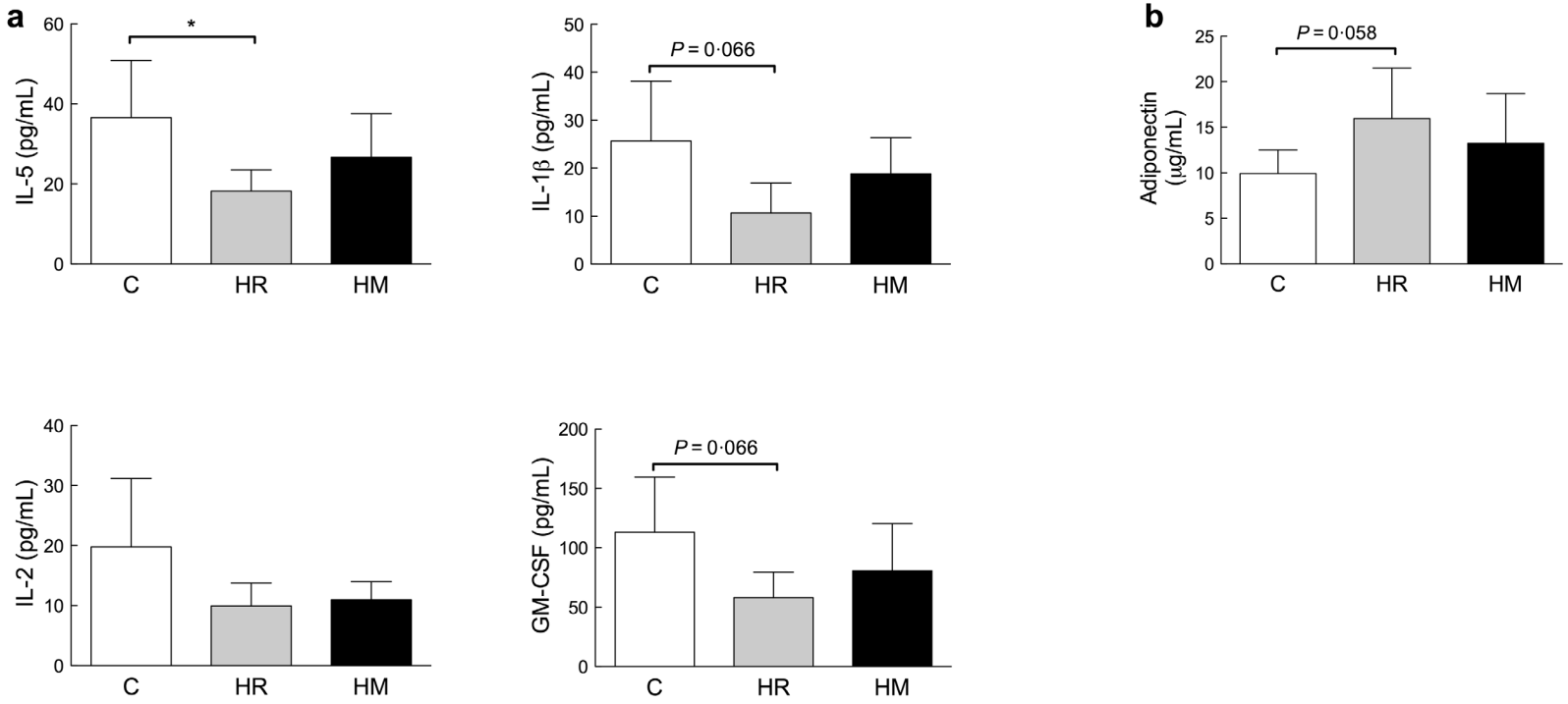
Hepatic lipid metabolism

- Lipid oxidation increased in:
  - Mitochondria (CPT2)
  - Peroxisomes (ACOX1)
- Lipogenesis reduced (FASN)



Plasma cytokine levels

- Herring roe reduced inflammation in TNFalpha mice



Bjørndal *et al. Lipids in Health and Disease* 2014, **13**:82
http://www.lipidworld.com/content/13/1/82

LIPIDS IN HEALTH AND DISEASE

RESEARCH Open Access

Phospholipids from herring roe improve plasma lipids and glucose tolerance in healthy, young adults

Bodil Bjørndal[1*], Elin Strand[1], Jennifer Gjerde[1,2], Pavol Bohov[1], Asbjørn Svardal[1], Bernd WK Diehl[4], Sheila M Innis[3], Alvin Berger[5,6] and Rolf K Berge[1,7]

Abstract

Background: Herring roe is an underutilized source of n-3 polyunsaturated fatty acids (PUFAs) for human consumption with high phospholipid (PL) content. Studies have shown that PL may improve bioavailability of n-3 PUFAs. Arctic Nutrition's herring roe product MOPL™30 is a PL: docosahexaenoic acid (DHA)-rich fish oil mixture, with a DHA:eicosapentaenoic acid (EPA) ratio of about 3:1, which is also rich in choline. In this pilot study, we determined if MOPL30 could favorably affect plasma lipid parameters and glucose tolerance in healthy young adults.

Methods: Twenty female and one male adults, between 22 and 26 years of age, participated in the study. Participants took encapsulated MOPL30, 2.4 g/d EPA + DHA, for 14 days, and completed a three-day weighed food record before and during the capsule intake. Plasma lipids and their fatty acid (FA) composition, plasma and red blood cell (RBC) phosphatidylcholine (PC) FA composition, acylcarnitines, choline, betaine and insulin were measured before and after supplementation (n = 21), and one and four weeks after discontinuation of supplementation (n = 14). An oral glucose tolerance test was performed before and after supplementation.

MOPL30 supplement: phospholipids from herring roe

Benefits of Herring-products:

- Immature roe from Spring-spawning Norwegian herring is an underutilized source of omega-3 for humans
- Arctic Nutrition's herring roe MOPL30™ product has about 30% phospholipids and DHA:EPA ratio of 3:1
- A majority of phosphatidyl-choline

- MOPL30: 511 mg fill wt capsules
  - 56 mg EPA
  - 158 mg DHA
  - 12 mg n3 DPA

ARCTIC NUTRITION

UNIVERSITAS BERGENSIS

uib.no

Adapted from prof. Alvin Berger

Study justification

- Studied healthy, young subjects (normal BMI) with high habitual fish consumption (=Norwegians) to determine if MOPL30 still increases plasma omega 3 and affects other parameters
- Evaluated glucose parameters
- Wash-out effects are rarely examined
- Used dose (≈2 g) consistent with TAG lowering

Adapted from prof. Alvin Berger

Participants

- Age 20-26 years
- Average BMI: 21.2 ± 2.8
- 20 women and 1 man

- Omega-3 supplements and roe-products were excluded from the diet 3 wks before baseline samples (habitual fish consumption permitted)

Study design

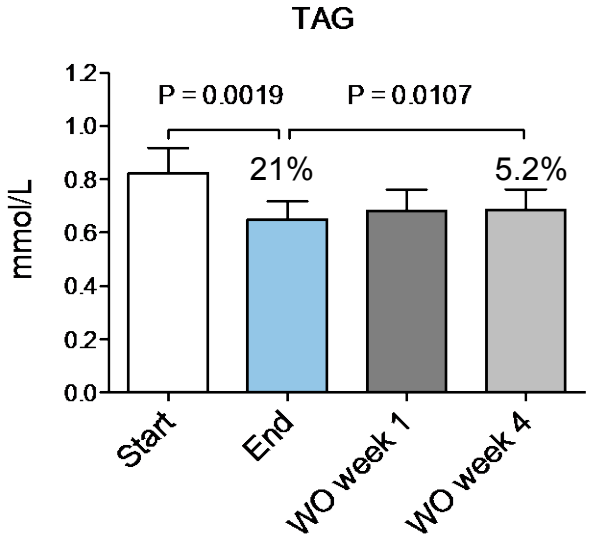
- 21 healthy individuals were given MOPL30 supplement for **14 days**
- **EPA (613 mg) + DHA (1737 mg)** per day
  - 4 capsules breakfast (8-9)
  - 4 capsules lunch (12)
  - 3 capsules dinner (16-17)
- Blood samples taken, and oral glucose tolerance test (OGTT) performed (n = 21 subjects) at **baseline** and **end of study**
- Fasting plasma glucose, insulin, and lipids was measured

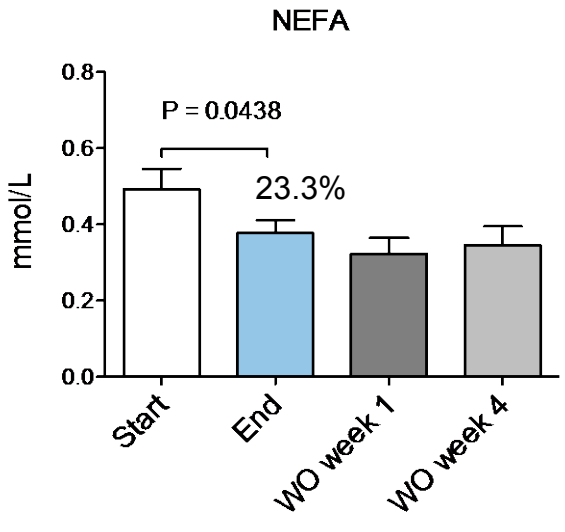
Study design – wash out effect

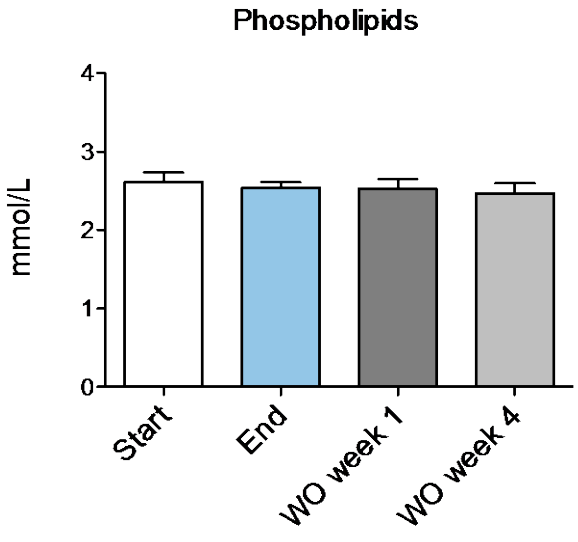
- EDTA-plasma collected **1 wk** and **4 wks** after final day of capsule intake (n=14)
  - Fasting plasma lipids, glucose and insulin were measured

Plasma lipids: TAG, NEFA, PL

- TAG and NEFA reduced after 2 wks MOPL30-
- TAG increased 4 wks after trial vs end, but still lower vs start



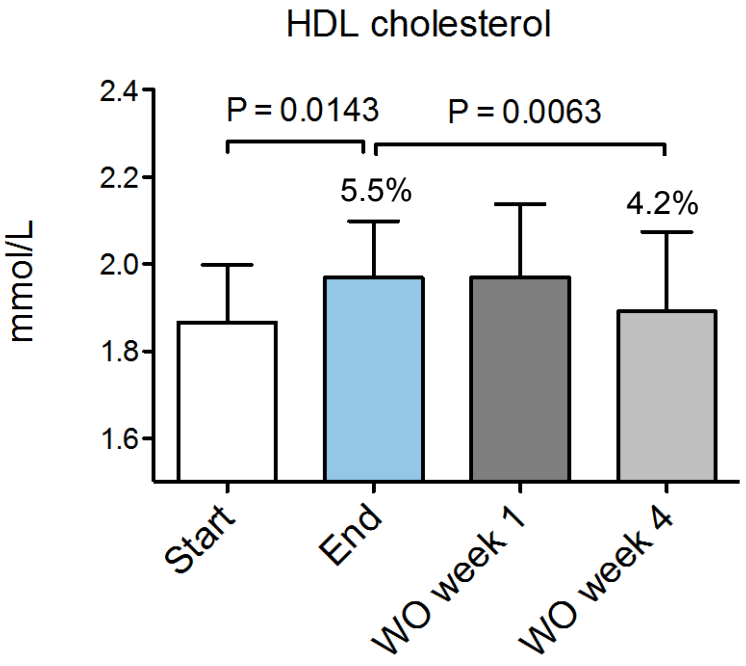


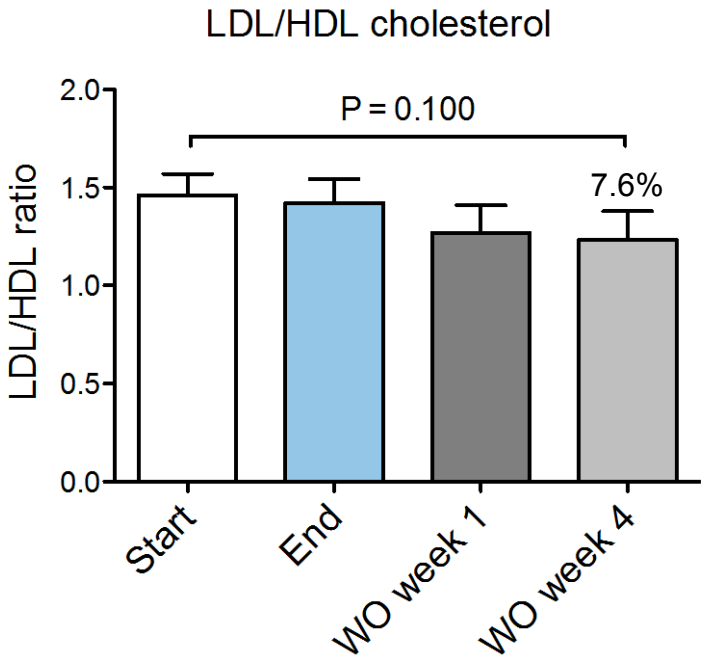


$\bar{X}$+SEM shown. Non-parametric pairwise t-test for start vs end (n=21 subjects); or end vs wash out (WO) wk 1 and 4 (n=14) D'agostine and Pearsons omnibus normality test, Prism GraphPad

HDL cholesterol and LDL/HDL ratio

- HDL increased after 2 wks MOPL30
- HDL returned to start levels after 4 wks WO
- LDL/HDL cholesterol ratio unchanged





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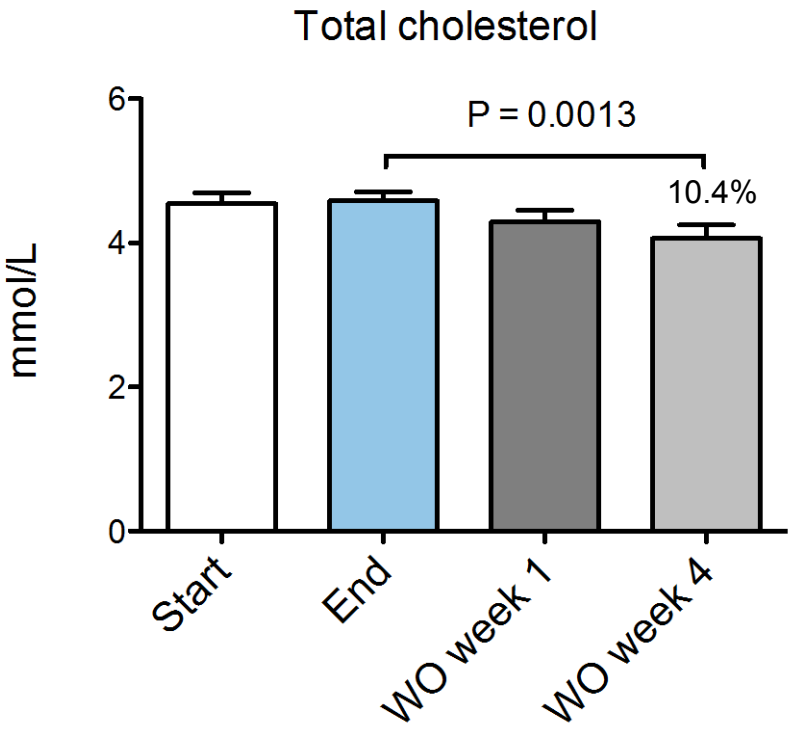
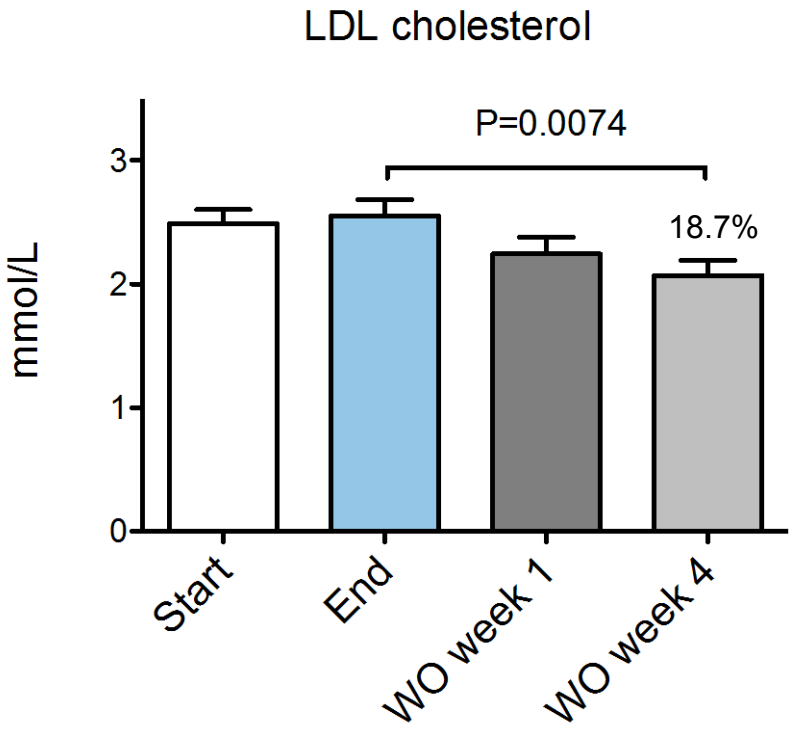
Plasma lipids: total and LDL-cholesterol

- No effect on total- and LDL after 2 wks
- Total- and LDL reduced- 4 wk wash out vs end
- Note: MOPL30 has 2.2% (w/w) cholesterol or 124 mg/d in 11 capsules (1/3 of requirement)





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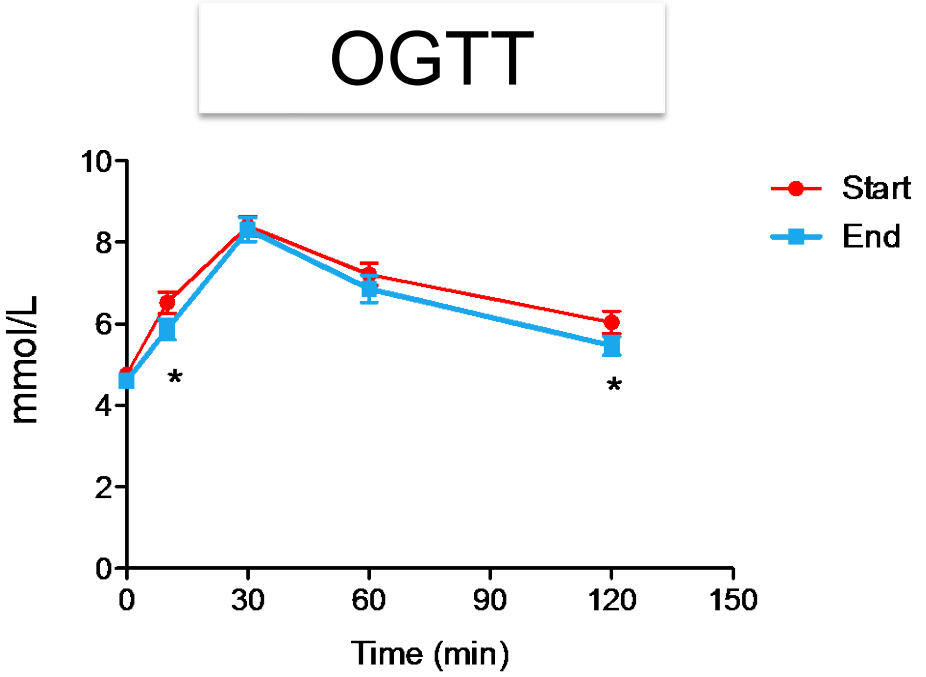
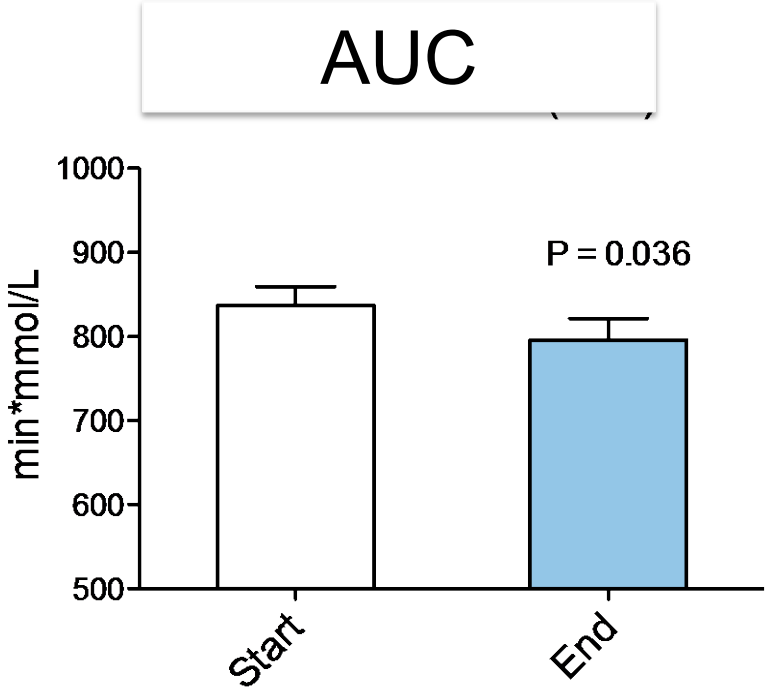
Oral glucose tolerance test (OGTT)

- Reduced plasma glucose level 10 min and 2 h after glucose intake, and reduced area under the curve (AUC) after 2 wks MOPL30

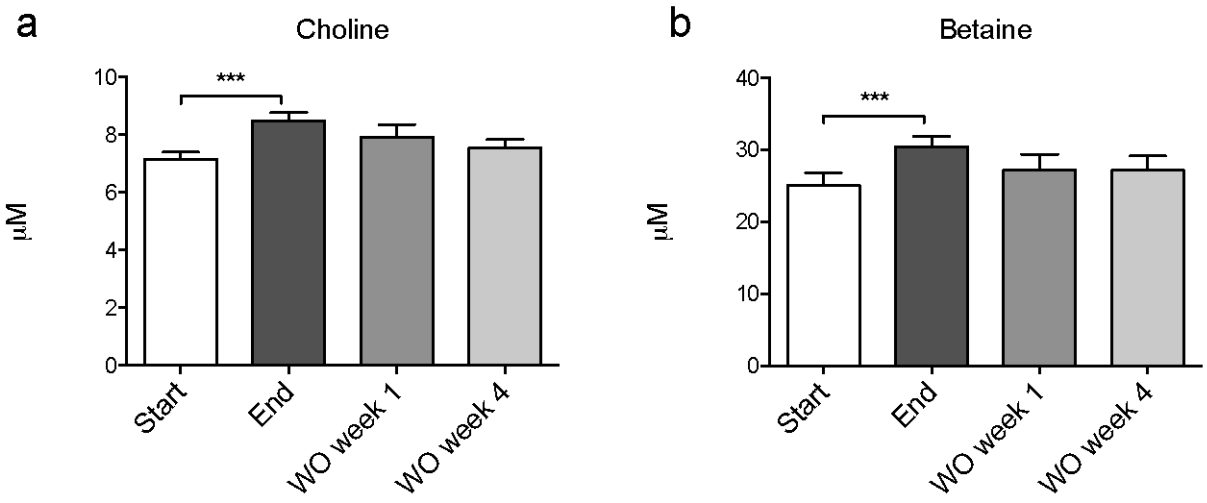




$\bar{X}$+SEM shownn (n = 20) Pairwise t-test. *p<0.05

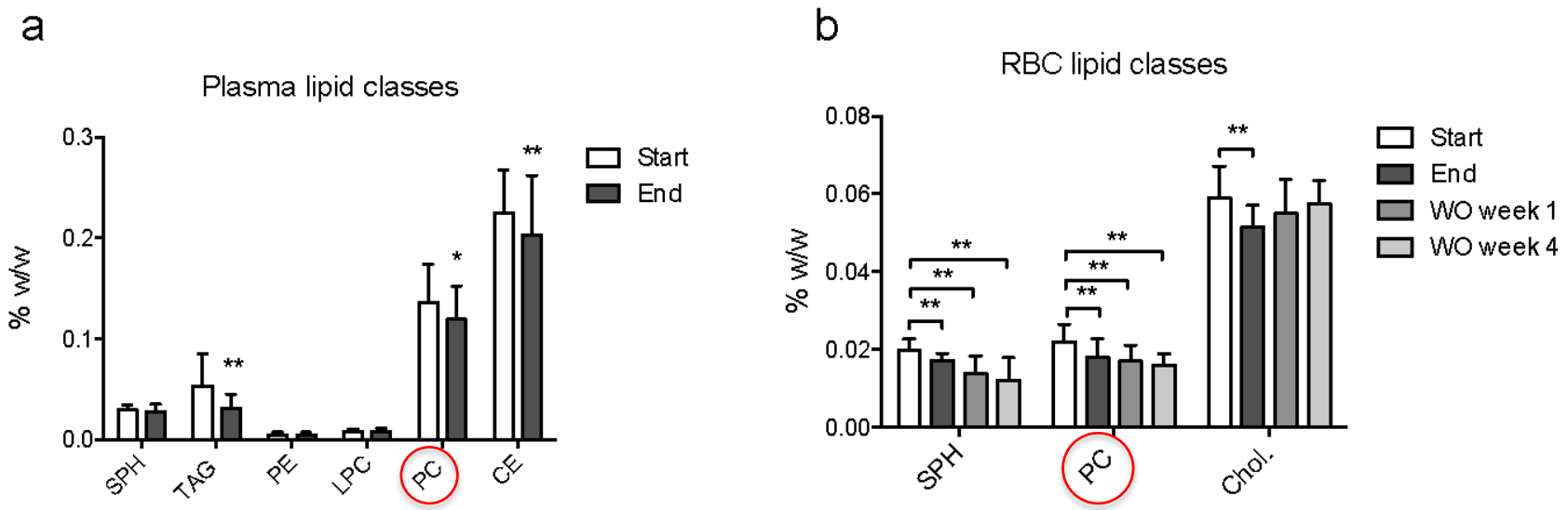
Plasma choline and betaine levels

- 2 wks MOPL30 increased plasma choline and betaine



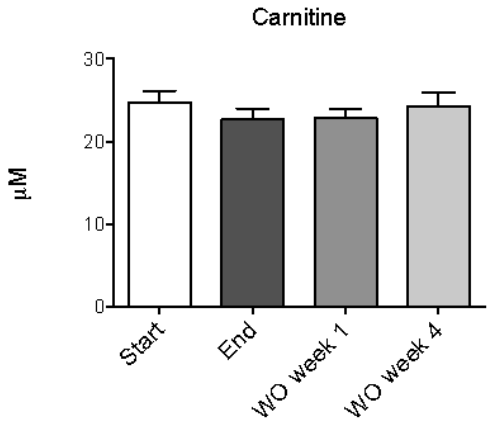
Plasma and RBC lipid classes (shotgun lipidomics)

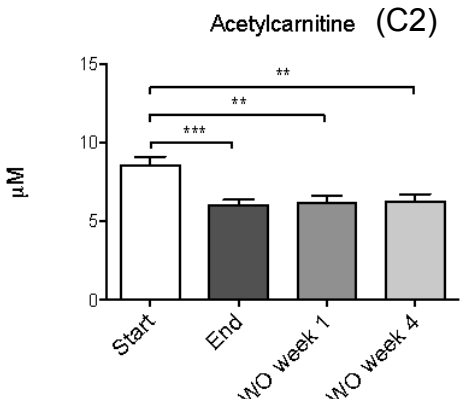
- Interestingly, PC was not increased in plasma and RBC

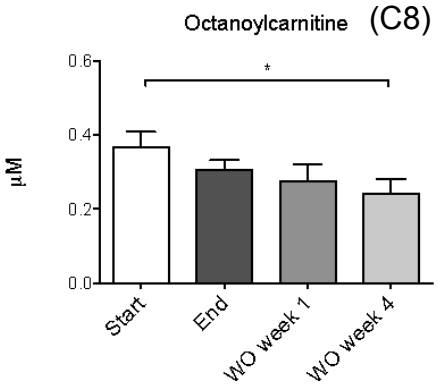


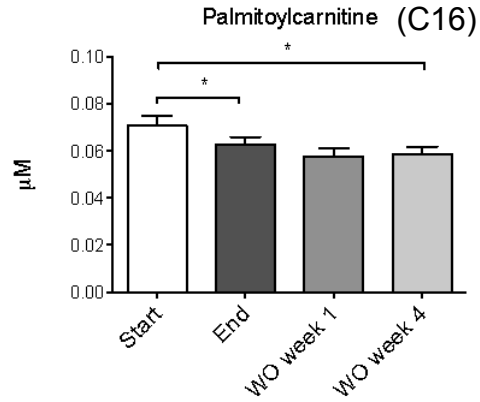
Plasma carnitine and acylcarnitines

- While carnitine itself was unchanged, acylcarnitines were reduced in plasma by MOPL30









High levels linked to poor prognosis in CVD patients

CONCLUSIONS: baseline to end

- 2 g MOPL30/d reduced plasma **TAG** and **NEFA**, and increased **HDL-C** in young individuals with a diet rich in omega-3.
- In other short-term studies in healthy people consuming high levels of omega-3, fish oil-TAG did not lead to favorable changes in lipoproteins
- Reduced levels of CVD risk-associated **acylcarnitines**
- Changes to AUC in OGTT suggested improved **insulin response**
- Free **choline** and **betaine** were increased, but not PC
- **EPA** increased more than **DHA** in the PC fraction

CONCLUSIONS-Wash out

- Decreases in **TAG** and increases **HDL** were reversed 4 wks after discontinuation of MOPL30
- Total **cholesterol** and **LDL** were unchanged from baseline – end of MOPL30 but reduced 4 wks after end of supplement (vs End) - a delayed response to MOPL30?
- A prolonged effect on plasma acylcarnitine levels?

Acknowledgement

Lipid Research Group:
Prof. Rolf K. Berge
Dr. Bodil Bjørndal
Prof. emeritus Jon Skorve

Kari Mortensen
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Prof. Bernt Diehl

Aker Biomarine / Dr. Lena Burri

Prof. Asbjørn Svardal

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