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Darmmicrobiota en metabole gezondheid

Ellen Blaak

AOAC Lowlands Symposium Darmgezondheid, gezondheid zit in de darmen Breda, 21th Sept 2017

Professor in Physiology of fat metabolism, Department of Human Biology
NUTRIM School of Nutrition and Translational Research in Metabolism
Maastricht University Medical Centre+
The Netherlands

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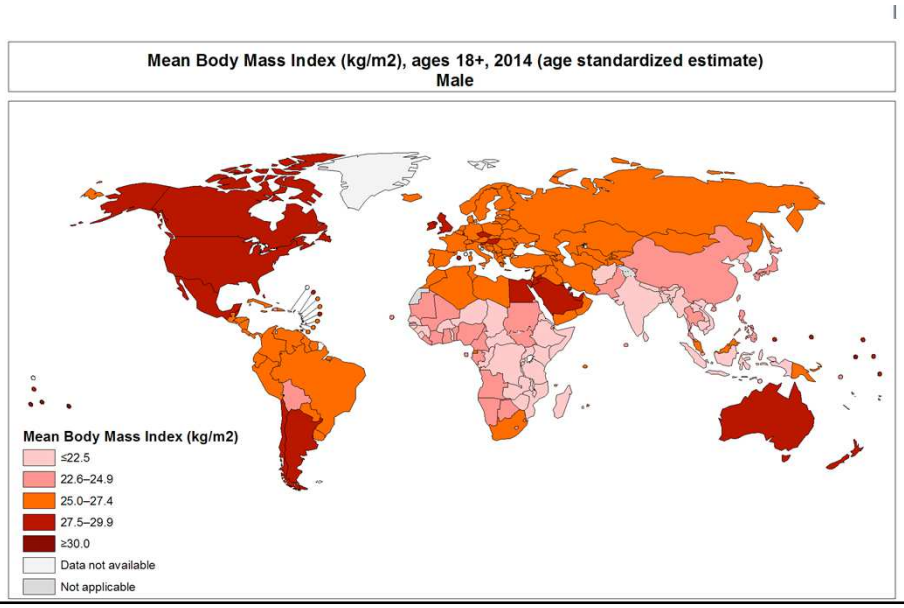
Overview van de presentatie

- Inleiding obesitas en microbiota
- Modulatie van microbiota en metabole gezondheid
  - Feces transplantatie
  - Antibiotica
- De rol van korte keten vetzuren (SCFA) in metabole gezondheid
- Modulatie van microbiota met voeding
- Conclusies

Obesitas – pandemie van 21ste eeuw



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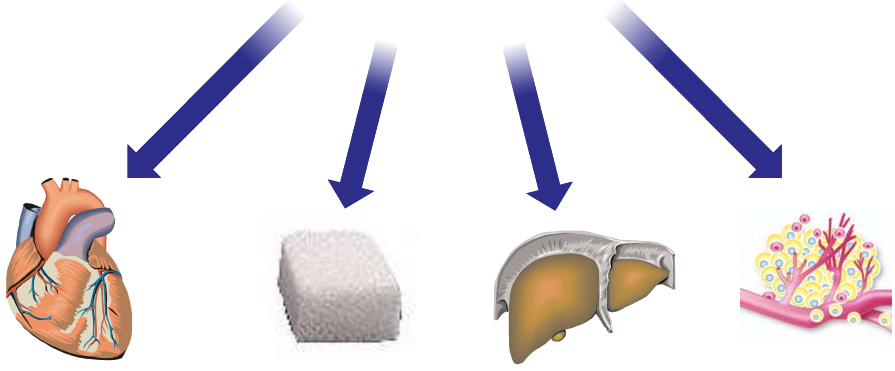
Obesitas: een belangrijke risicofactor voor chronische ziekten

OBESITY



Cardiovascular disease | Type 2 diabetes | Liver steatosis | Cancer

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Obesitas en diabetes en microbiota

- Wereldwijd bereikt obesitas een epidemische prevalentie
- De obesitas pandemie wordt niet verklaard door de meest voorkomende gen-omgevingsinteracties
- Darm microbiota geven producten die ons metabolisme kunnen beinvloeden en kunnen zo een rol spelen bij obesitas en diabetes
- Obesitas en Type 2 diabetes mellitus gaan gepaard met minder microbiële diversiteit en een andere microbiotasamenstelling
- Interventies die de darm microbiota manipuleren kunnen een gunstig effect hebben op diabesitas risico (resultaten niet consistent)

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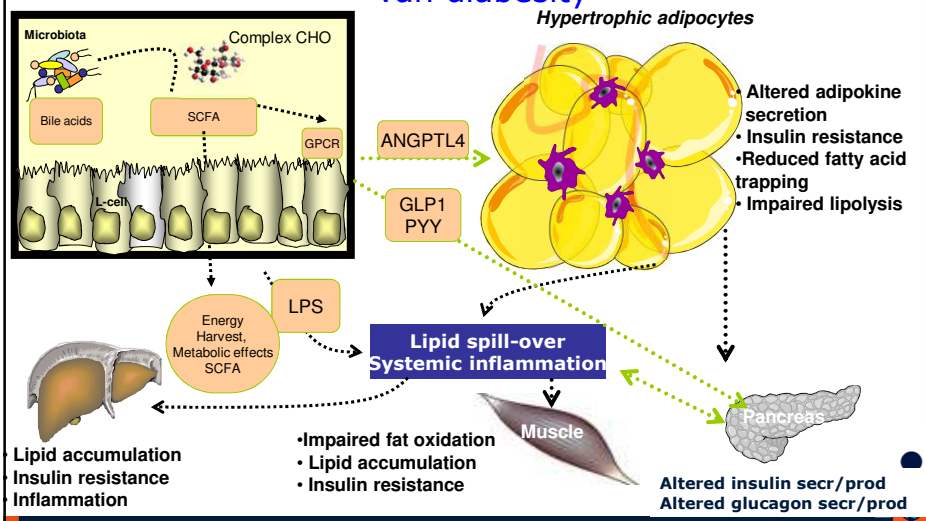
Darmbacteriën kunnen onze stofwisseling beinvloeden en zo een rol spelen in de ontwikkeling van diabesity



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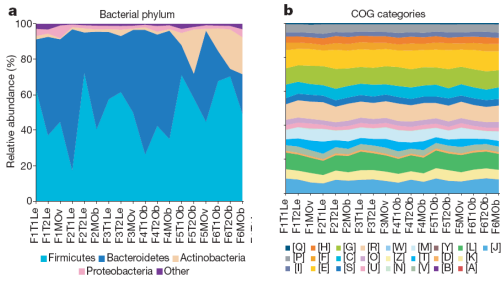
Microbiota is individual specific

- **Is there a normal microbiota?**

Qin *et al.* Science. 2010

- **57 species common >90% individuals**
- **~ 250.000 genes (at least 50% of the individuals)**

- **Compositional *vs.* functional differences**



Turnbaugh *et al.* Nature. 2009

Adapted from:
Miguel Gueimonde
CSIC

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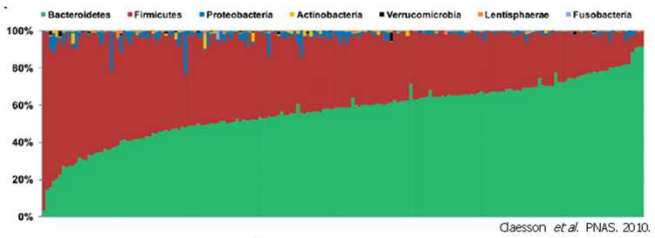
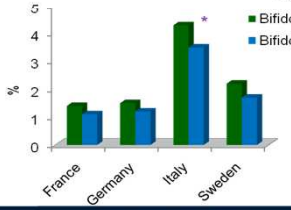
Microbiota is individual specific

- **Challenge:** definition of a normal/healthy microbiota
  - Inter-individual variation



Claesson *et al.* PNAS. 2010.

  - Geographical differences



Mueller et al. AEM. 2006.

Adapted from:
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Human gut microbes associated with obesity

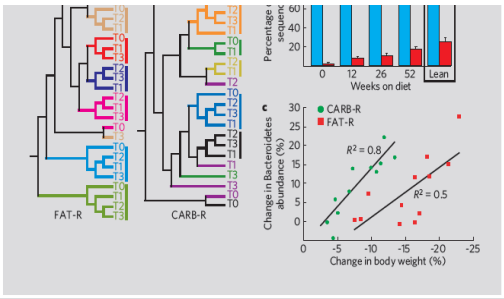
Ruth E. Ley, Peter J. Turnbaugh, Samuel Klein Jeffrey I. Gordon

Beinvloed door voeding,populatie, ethniciteit, leeftijd, geslacht, glucometabole status

that this proportion increases with weight loss on two types of low-calorie diet. Our findings indicate that obesity has a microbial component, which might have potential therapeutic implications.

Trillions of microbes live in the human gut, helping to break down otherwise indigestible foods[1]. Transplanting the gut microbiota from normal mice into germ-free recipients increases their body fat without any increase in food consumption[2], raising the possibility that the composition of the microbial community in the gut affects the amount of energy extracted from the diet[2].

The relative abundance of the two predominant bacterial divisions (deep evolutionary lineages or superkingdoms) in mice differs between lean and obese animals: mice that



NATURE|Vol 444|21/28 December 2006

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Feces transplantatie van muis naar muis

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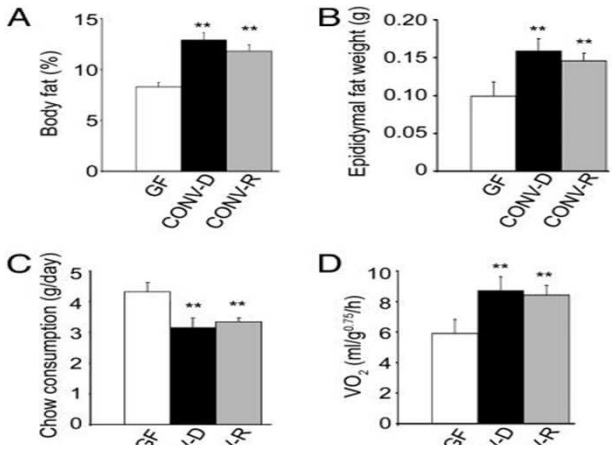
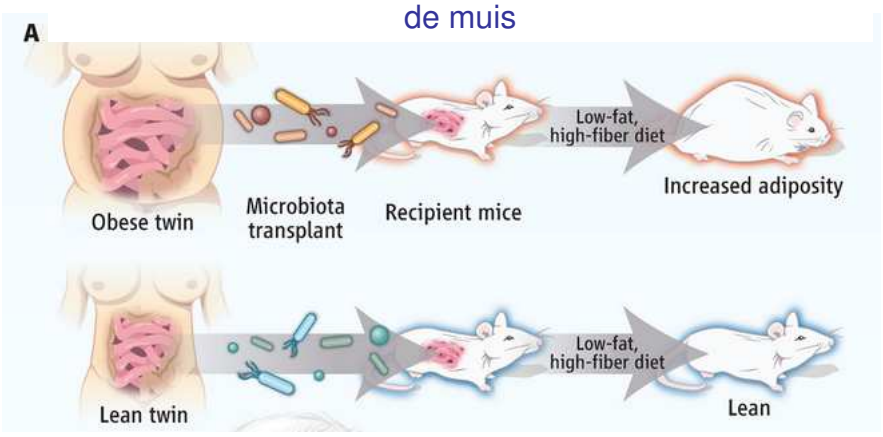
Van muis naar muis: Darm Flora draagt bij aan obesitas en diabetes

(Bäckhed et al, 2004, 2007)



Microbiota vrije muizen zijn beschermd tegen een verstoorde glucose stofwisseling en insuline resistentie

Van mens naar muis: Darmmicrobiota van tweelingen met/zonder overgewicht moduleren de stofwisseling van de muis



V K Ridaura et al. Science 2013;341:1241214

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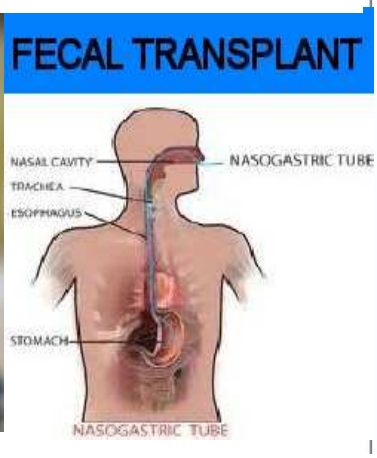
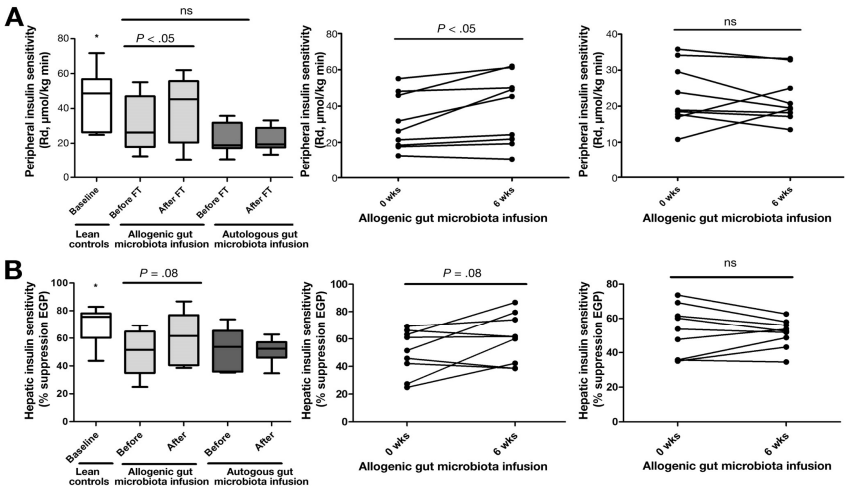
Van mens naar mens: feces transplantatie en stofwisseling



Transplantatie van Intestinele Microbiota van donoren met normaal gewicht verhoogt Insuline Gevoeligheid bij mensen met het Metabool Syndroom



Vrieze et al, 2012

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Transplantatie van microbiota tussen

- normale en microbiota vrije muizen,
- mensen en muizen
- mensen en mensen

heeft een rol van intestinale microbiota bij de ontwikkeling van obesitas en insuline resistentie aangetoond

Effecten bij de mens zijn klein!

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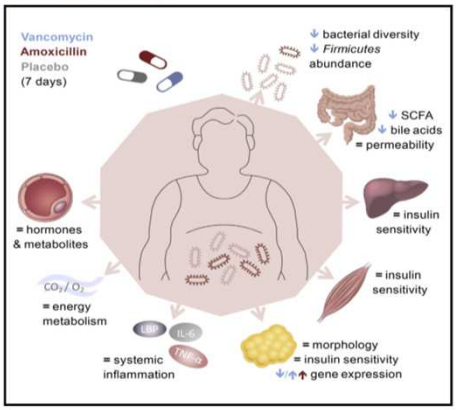
Cell Metabolism

Clinical and Translational Report

Effects of Gut Microbiota Manipulation by Antibiotics on Host Metabolism in Obese Humans: A Randomized Double-Blind Placebo-Controlled Trial

Graphical Abstract



Authors

Dorien Reijnders, Gijs H. Goossens, Gerben D.A. Hermes, ..., Erwin G. Zoetendal, Cornelis H.C. Dejong, Ellen E. Blaak

Correspondence

e.blaak@maastrichtuniversity.nl

In Brief

Reijnders et al. show that a 7-day antibiotic treatment (amoxicillin/vancomycin) has no clinically relevant impact on host metabolism in obese humans, both directly after treatment cessation and at 8-week follow-up, despite profound changes in gut microbiota, short-chain fatty acid, and bile acid concentrations induced by vancomycin.

Highlights

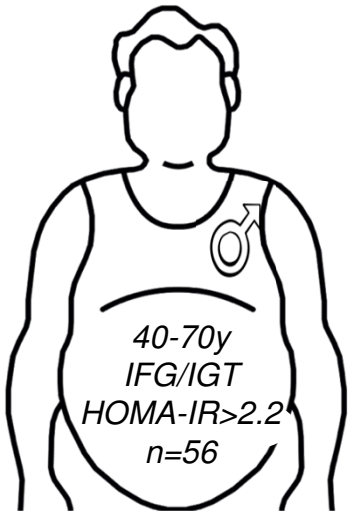
Accession Numbers

Antibiotica interventie

Treatment

Placebo
Amoxicillin
Vancomycin

1500 mg/day
7 days

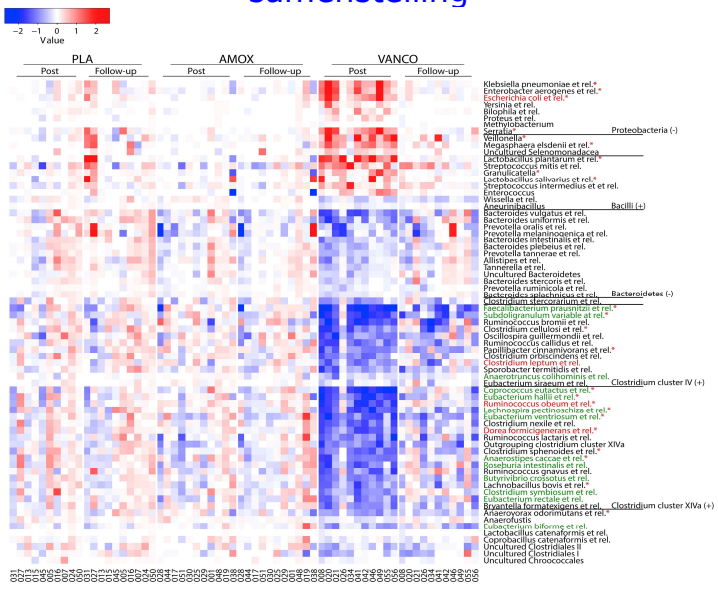


Timing

wk 1 pre
wk 2
2d wash out
wk 3 post
wk 8 follow-up

Reijnders et al, Cell metabolism, 2016

Duidelijke veranderingen in microbiota samenstelling



Vanco:
- Increased gram negative Proteobacteria
- less gram-positive bacteria
- decreased diversity
- After 8 weeks similarity with baseline still lower, diversity slightly lower

Amox:
- no diversity and/or composition differences

Reijnders et al, Cell metabolism, 2016

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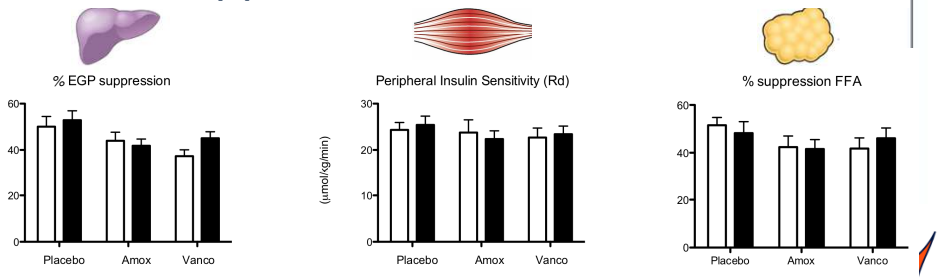
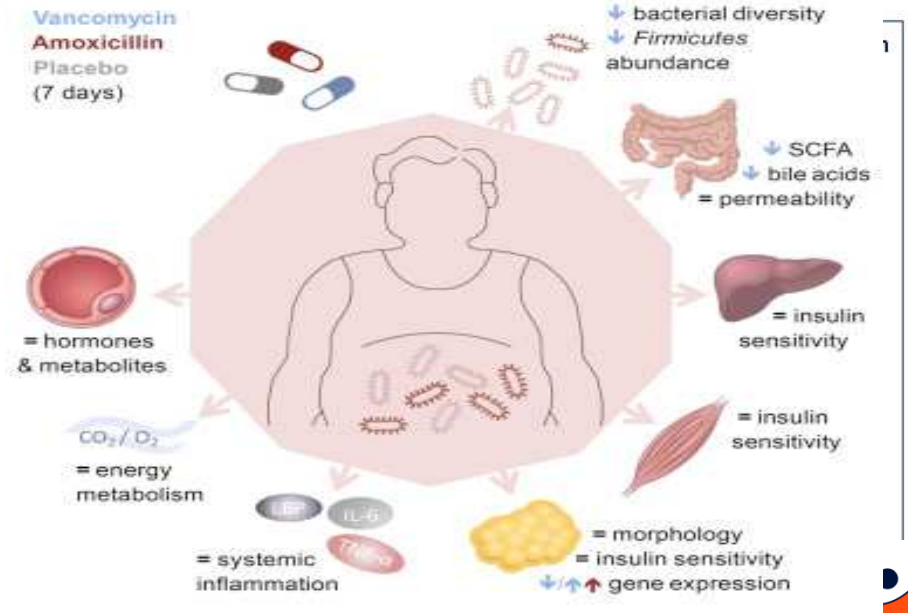
Geen effect op insuline gevoeligheid

- *Two-step hyperinsulinemic-euglycemic clamp*
  - ***E**ndogenous **G**lucose **P**roduction*

Geen effect op insuline gevoeligheid 8 wk na het stoppen van de interventie



Reijnders et al, Cell metabolism, 2016



Reijnders et al, Cell metabolism, 2016

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Perspectief

- Modulatie van de microbiota door een 7-daagse antibiotica kuur heeft geen effect op de stofwisseling, ook niet 8 wk na het stoppen van de interventie
- In tegenspraak met veel dierstudies!

- Meer frequent antibiotica gebruik, lange termijn dieet effecten?
- Speelt het metabole fenotype een rol?

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Functie microbiota: saccharolytische en proteolytische activiteit

- Saccharolysis leads primarily to SCFA and gasses
  - SCFA: acetate, propionate, butyrate
  - lactate -> only accumulates when there is a fast fermentation
  - CO_2, CH_4, H_2

- Proteolysis in addition leads to toxic metabolites
  - BCFA: iso-butyrate, iso-valerate
  - ammonia
  - phenolics: phenol, indol, *p*-cresol, skatol
  - H_2S, CH_3SH, etc

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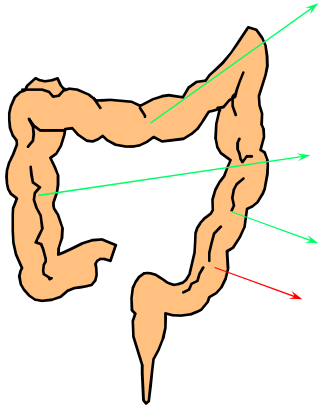
Plaats van fermentatie



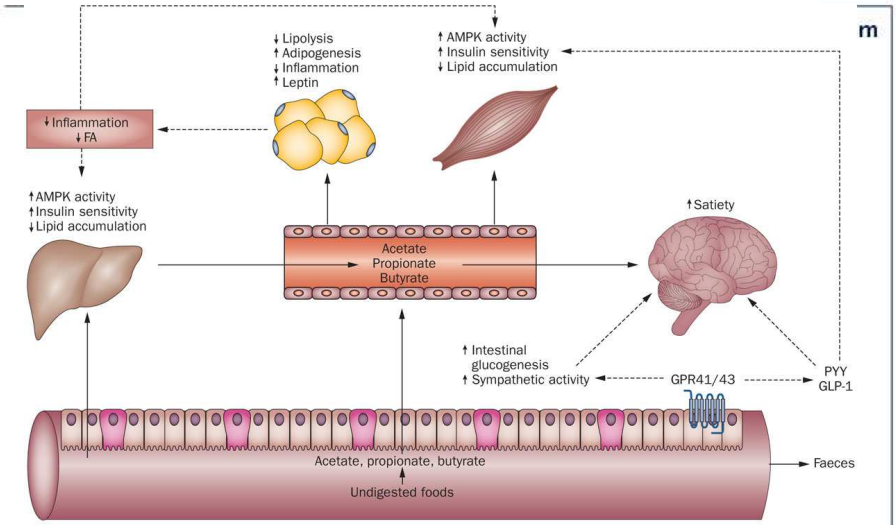
transverse colon: combination of saccharolytic and proteolytic fermentation

proximal colon: primarily saccharolytic fermentation

distal colon: primarily proteolytic fermentation

distal colon: colon cancer and IBD

Functionality of microbiota: SCFA



Nature Reviews | Endocrinology

Canfora, E. E. *et al.* (2015) Short-chain fatty acids in control of body weight and insulin sensitivity *Nature Reviews Endocrinology* doi:10.1038/nrendo.2015.128

Major SCFA

Acetate

Propionate

Butyrate

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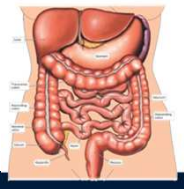
Distal and proximal colonic acetate infusion and metabolic profile

Hypothesis:Colonic administration of SCFA has beneficial effects on human substrate and energy metabolism

Question:

Where to administer? Distal / Proximal?



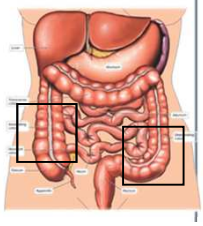
Distal and proximal colonic acetate infusion and metabolic profile

Aim:

To investigate differential effects of proximal and distal colonic infusions with sodium acetate on human fat oxidation, energy expenditure and circulating metabolic markers

Study design

- Double blind, placebo controlled, randomized crossover study
- Six healthy overweight males (BMI 25 – 34.9 kg/m^2)
  - Aged 20 – 50 years;
  - Weight stable for at least 3 months (± 2 kg)
  - No use of antibiotics, pre- or probiotics

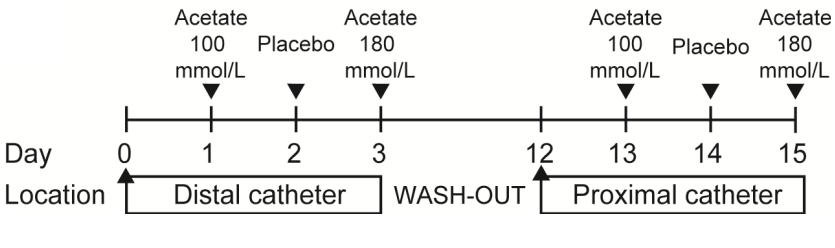


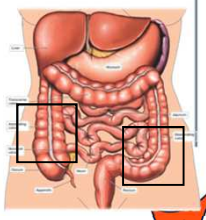
Intervention

1. Sodium acetate 100mmol/L (12mmol in 120mL water)
2. Sodium acetate 180mmol/L (21.6mmol in 120mL water)
3. Placebo (0.9% NaCl) in 120mL

Van der Beek et al, Clin Sci 2017

Intervention protocol





Van der Beek et al, Clin Sci 2017

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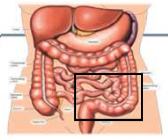
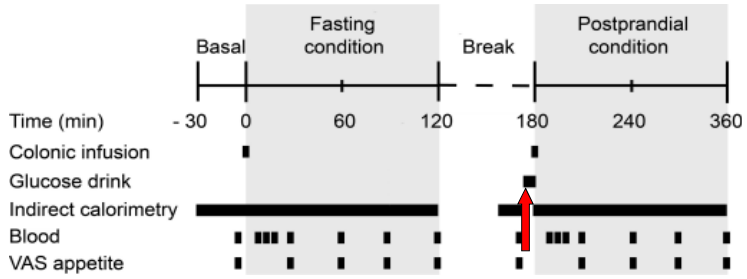
Test day protocol





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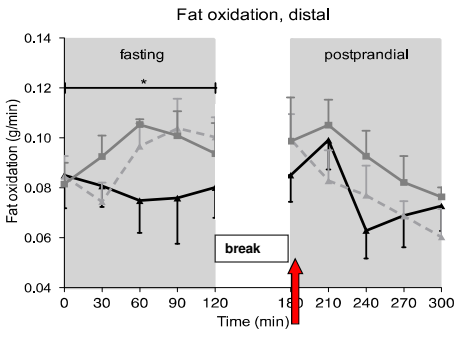
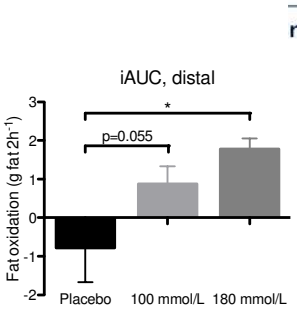
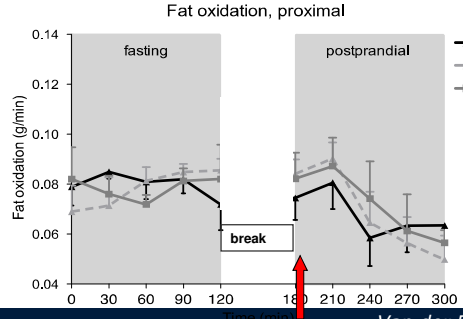
Van der Beek et al, Clin Sci 2017

Fat oxidation







ANOVA * p<0.05 placebo vs 180mM

Ingestion oral glucose solution

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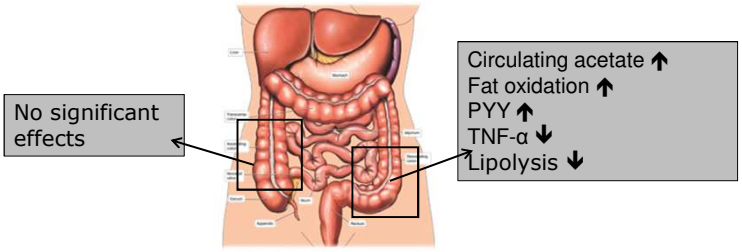
Van der Beek et al, Clin Sci 2017

Distal, not proximal, colonic acetate infusion improves metabolic profile



→ *Increasing colonic and systemic acetate beneficially affect the metabolic profile*

→ *Validated distal colonic infusion as a good model to study SCFA effects on metabolism*

Van der Beek et al, Clin Sci 2017

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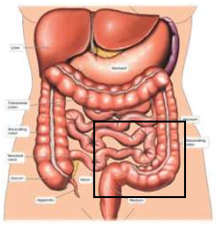
Combinations of SCFA and metabolic profile

To investigate acute effects of distal colonic infusions of SCFA combinations on substrate and energy metabolism



Canfora et al, Scientific reports, 2017

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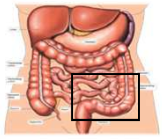
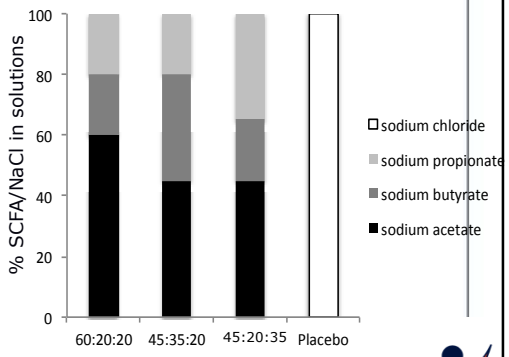
Intervention protocol (1)



Double blind, placebo controlled, randomized crossover study with 4 distal infusions:

- Placebo:
  40mmol NaCl
- High sodium acetate (60:20:20):
  24mmol NaAc, 8mmol NaBu, 8mmol NaPr
- High sodium butyrate (45:35:20):
  18mmol NaAc , 14mmol NaBu, 8mmol NaPr
- High sodium propionate (45:20:35):
  18mmol Na Ac, 8mmol NaBu, 14mmol NaPr
- All diluted in 200mL water



Department of Human Biology *Canfora et al, Scientific reports, 2017*

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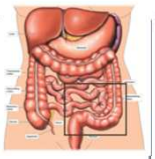
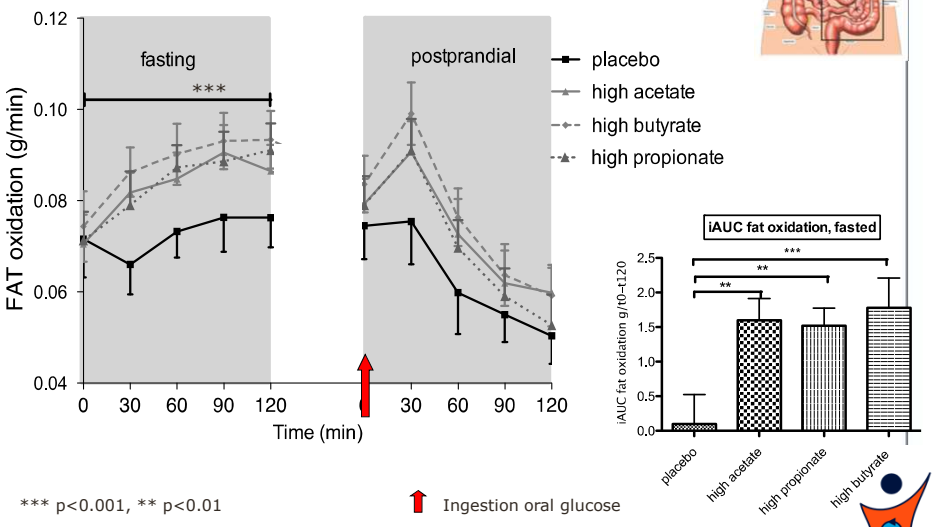
SCFA increase Fat oxidation





*** p<0.001, ** p<0.01

Ingestion oral glucose

Department of Human Biology *Canfora et al, Scientific reports, 2017*

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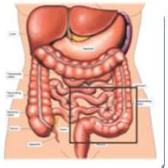
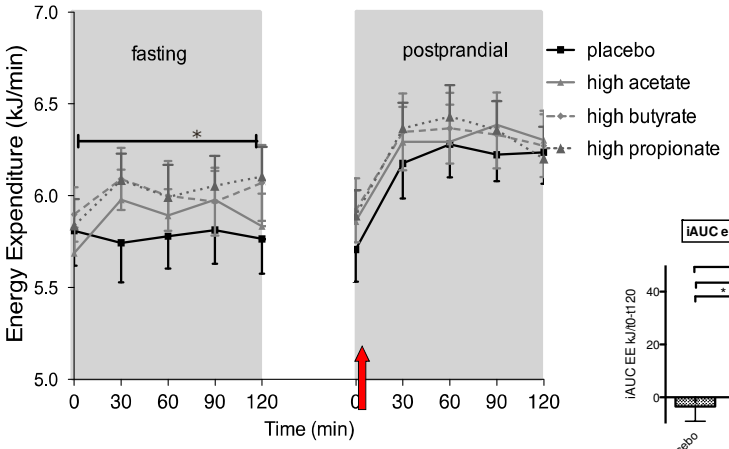
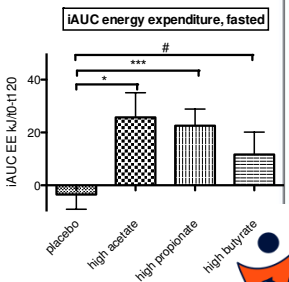
SCFA increase Energy Expenditure





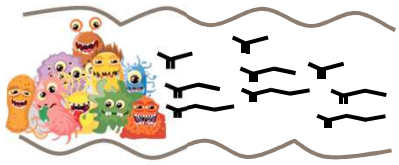


*** p<0.001, * p<0.05, # p<0.1

Ingestion oral glucose solution

Canfora et al, Scientific reports, 2017

Acetate maybe a target to prevent and treat obesity-related co-morbidities



Fat oxidation ↑
Energy Expenditure ↑
PYY ↑
IL-1β ↓
lipolysis ↓

Canfora et al, Scientific reports, 2017

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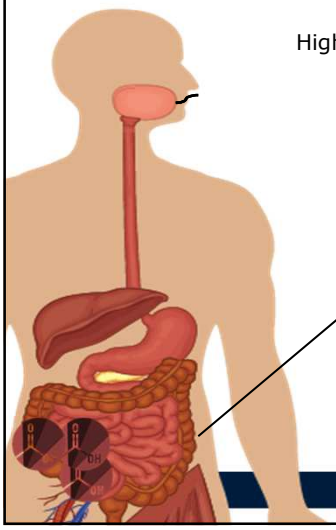
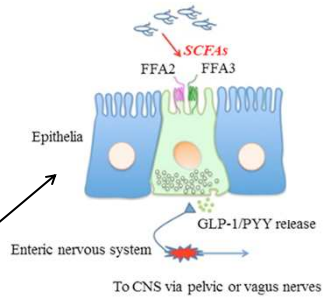
Why do we hypothesize that the distal colon should be targeted?



Higher density of PYY-producing L-cells in the distal colon



Kuwahara (2014), *Frontiers in Endocrinology*

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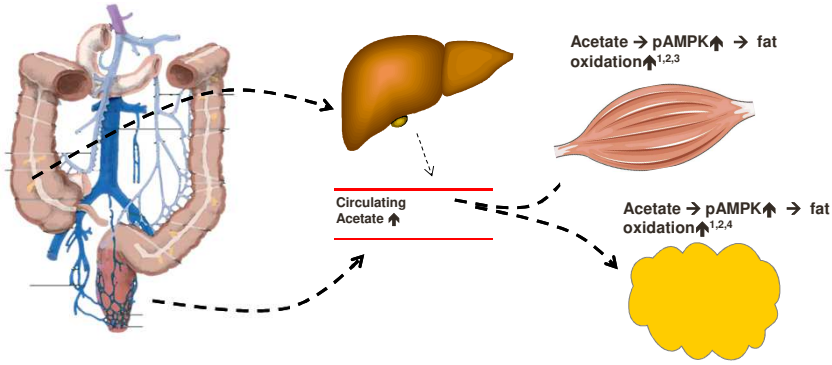
Increased fat oxidation and EE only after distal administration:

Acetate bypass the liver → circulating acetate↑ → uptake in oxidative tissues → pAMPK↑ → fat oxidation↑



[1]Sakakibara *et al., 2006;* [2]Yamashita *et al., 2009;* [3]Kimura *et al., 2013;* [4] den Besten *et al., 2015*

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Summary on acetate

- Abundant animal and limited human in-vivo data: a beneficial role of acetate in the control of body weight, glucose homeostasis, and insulin sensitivity.
- In contrast, some animal data suggest that acetate promotes the development of obesity and insulin resistance.
- Human studies are warranted to evaluate the 'acetate discrepancies' with respect to effects on metabolic health.

Department of Human Biology *Canfora and Blaak, Curr Opin Clin Nutr Metab Care 2017*

Towards dietary intervention study with galacto-oligosaccharides targeting colonic acetate

Study design

- Double blind, placebo controlled, randomized parallel study
- 46 volunteers:
  - Males and postmenopausal females aged 45-70 years;
  - BMI 28 – 40 kg/m$^{2;}$
    Impaired glucose tolerance (IGT) and/or impaired fasting glucose (IFG);
    Weight stable for at least 3 months (± 2 kg);
    No use of antibiotics, pre- or probiotics for 3 months

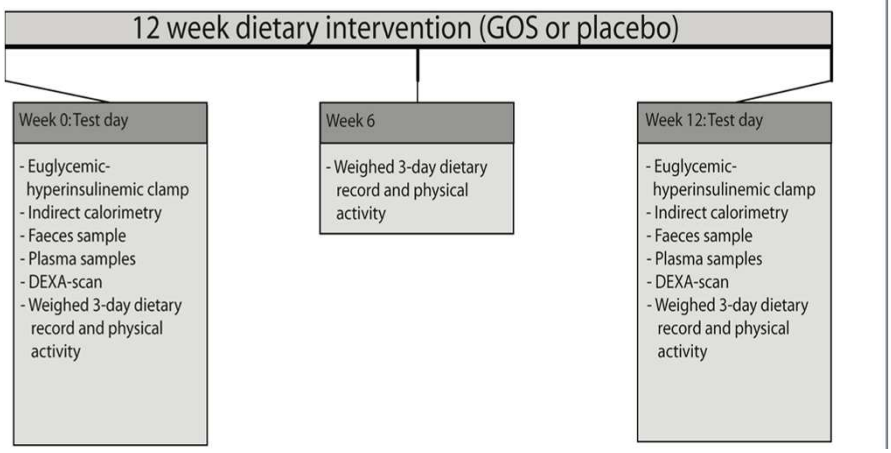
IGT: 2h plasma glucose during 75g OGTT
7.8-11.1 mmol/l
IFG: plasma glucose ≥ 5.6 mmol/l

Intervention groups:

1. Vivinal GOS 3x 5 gram per day for 12 weeks
2. Maltodextrin 3x 4.4 gram per day for 12 weeks (isocaloric placebo)

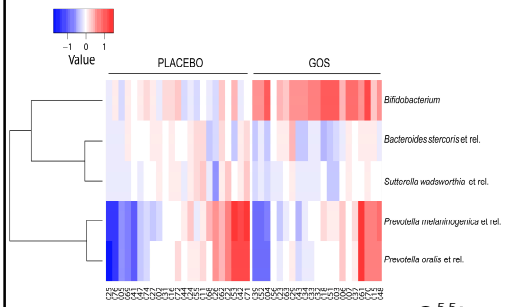
Department of Human Biology *Canfora et al, Gastroenterology, july 2017*

Study design

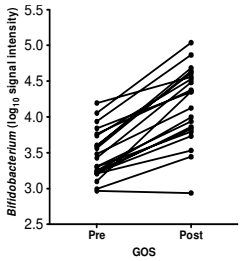


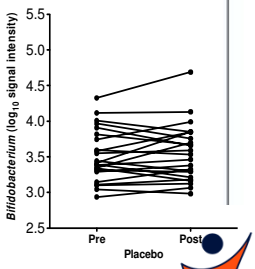
Canfora, *et al.* (2017), *Gastroenterology*

GOS increased faecal *Bifidobacterium spp.*



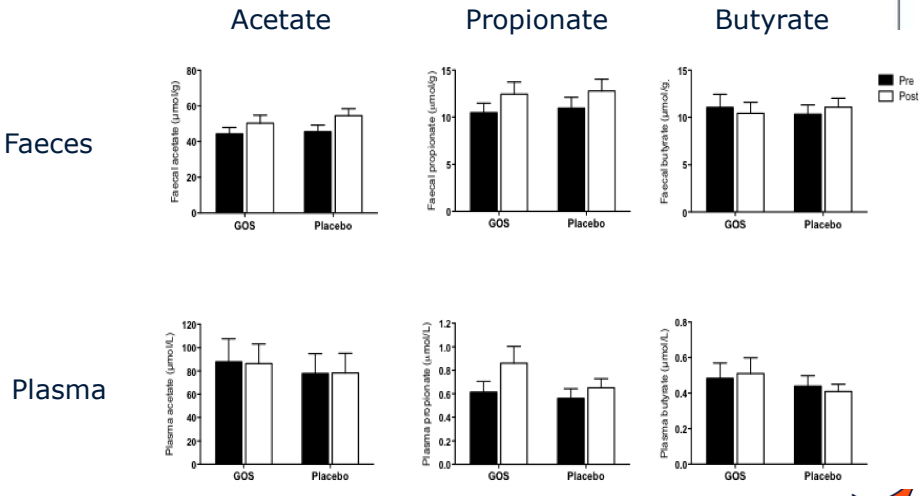
5.0 ± 0.3 fold increased relative abundance (*P*=0.009)





Canfora, *et al.* (2017), *Gastroenterology*

GOS did not affect faecal and plasma SCFA concentrations



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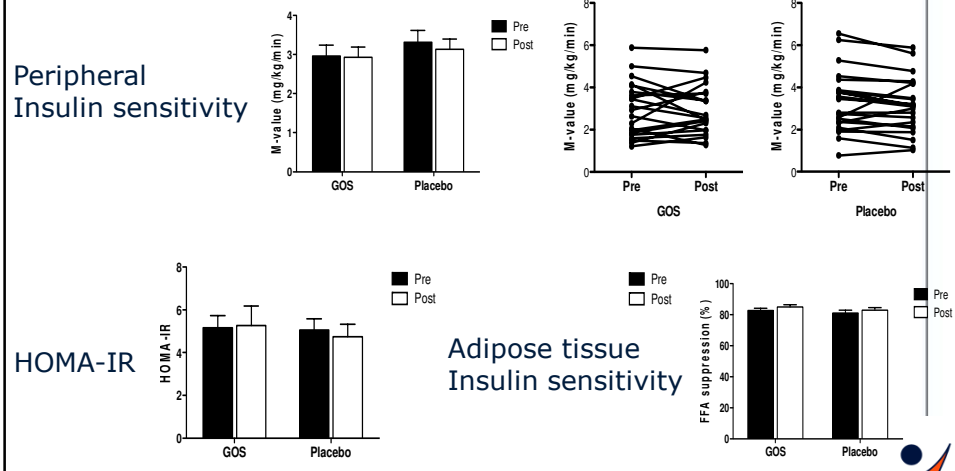
Canfora et al, Gastroenterology, july 2017

GOS did not affect markers of insulin sensitivity



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Canfora et al, Gastroenterology, july 2017

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In Summary

- Modulation of microbiota by means of antibiotics did not affect metabolic health after 7 days and in the longer term
- SCFA may be an important link between gut microbiota and metabolic health
- Distal, but not proximal, acetate infusion may improve fat oxidation and metabolic profile
- Rectal infusion of SCFA combinations all increase fat oxidation, energy expenditure and metabolic profile
- We propose that acetate is the main driver of this effect
- GOS increased bifidobacteria but not insulin sensitivity in prediabetic individuals
- Dietary fiber intervention focussed on targeted production of SCFA may improve intervention outcome with respect to metabolic profile, but this requires further confirmation

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Collaborations/Acknowledgements

TIFOOD NUTRITION

- Gijs Goossens
- Johan Jocken
- Birgitta vsn der Kolk
- Dorien Reijnders
- Emanuel Canfora
- Rudi Stinkens
- Max Vogel
- Jasper Most
- Kenneth Verbo
- Yvonne Essers
- Nicole Hoebers

Dept Human Biolog

And external Collaborations
Top Institute Food and Nutrition
(project GH003, microbiota, energy balance and metabolism)
EU consortia: EU-Lipgene, MIRdiet, EDIPS, Diogenes

Cargill

Diabetes Fonds

EFSD European Foundation for the Study of Diabetes