

Biomarkers of food intake

- Finding and validating biomarkers of foods by metabolomics

Lars Ove Dragsted

UNIVERSITY OF COPENHAGEN



DINAMIC



FOODBALL
The Food Biomarker Alliance



Why biomarkers of food intake?

Current state of dietary assessment

Subjective dietary instruments

FFQs

Food diaries

24-hour recalls/interviews

Systematic and Random Errors

Time consuming

Based on memory and food composition tables



Biomarkers and their overall classes

Biomarkers in general are defined as “objective measures used to characterize the current condition of a biological system”.

Gao et al. *Genes & Nutrition* (2017) 12:34
DOI 10.1186/s12263-017-0587-x

Genes & Nutrition

REVIEW

Open Access



A scheme for a flexible classification of dietary and health biomarkers

Qian Gao¹, Giulia Praticò^{1,2}, Augustin Scalbert³, Guy Vergères⁴, Marjukka Kolehmainen⁵, Claudine Manach⁶, Lorraine Brennan⁷, Lydia A. Afman⁸, David S. Wishart⁹, Cristina Andres-Lacueva^{10,11}, Mar Garcia-Aloy^{10,11}, Hans Verhagen^{12,13}, Edith J. M. Feskens⁸ and Lars O. Dragsted^{1*}

Abstract

Biomarkers are an efficient means to examine intakes or exposures and their biological effects and to assess system susceptibility. Aided by novel profiling technologies, the biomarker research field is undergoing rapid development and new putative biomarkers are continuously emerging in the scientific literature. However, the existing concepts for classification of biomarkers in the dietary and health area may be ambiguous, leading to uncertainty about their application. In order to better understand the potential of biomarkers and to communicate their use and application, it is imperative to have a solid scheme for biomarker classification that will provide a well-defined ontology for the field. In this manuscript, we provide an improved scheme for biomarker classification based on their intended use rather than the technology or outcomes (six subclasses are suggested: food compound intake biomarkers (FCIBs), food or food component intake biomarkers (FIBs), dietary pattern biomarkers (DPBs), food compound status biomarkers (FCSBs), effect biomarkers, physiological or health state biomarkers). The application of this scheme is described in detail for the dietary and health area and is compared with previous biomarker classification for this field of research.

Keywords: Biomarker, Classification, Nutrition, Ontology, Exposure, Effect, Susceptibility, Metabolomics, Review





Biomarkers and their overall classes

Biomarkers in general are objective measures used to characterize the current condition of a biological system.

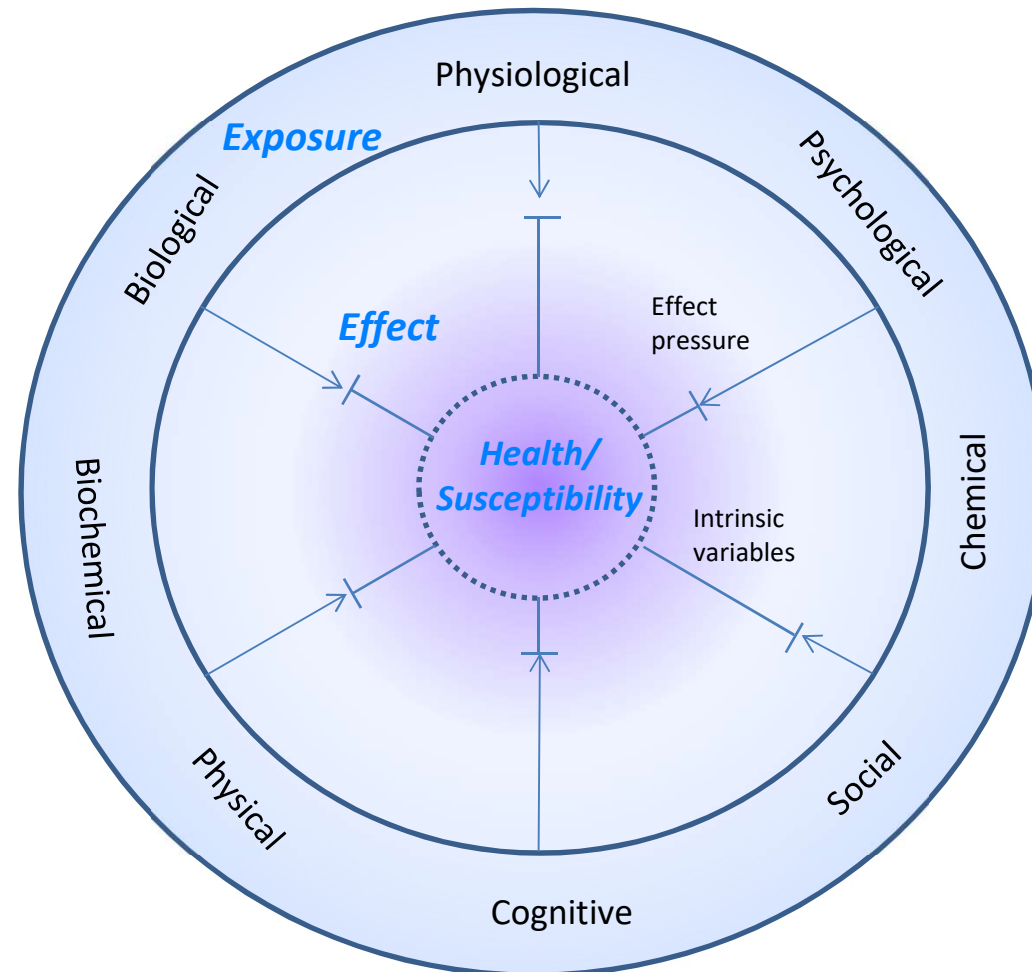
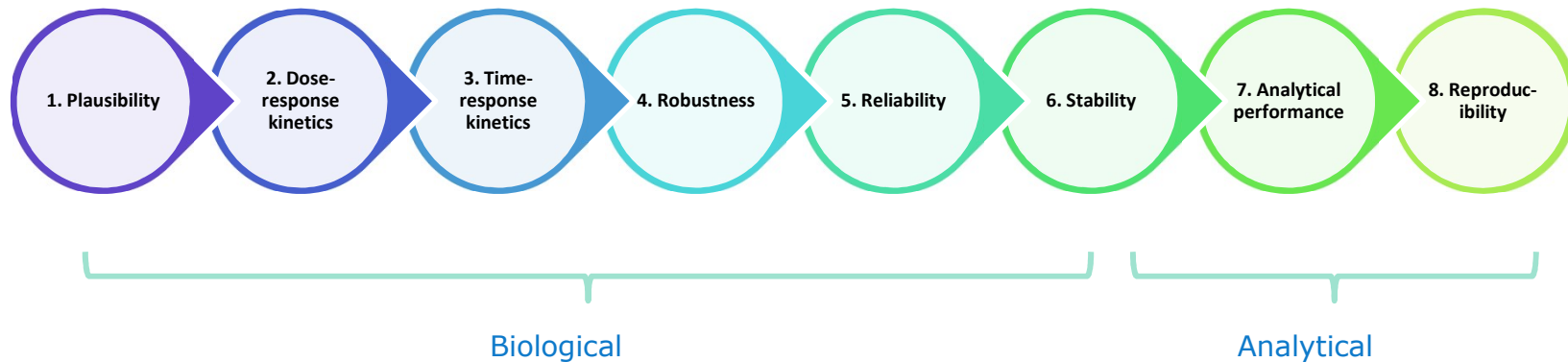


Figure. Diversity of interaction between the biological system with intrinsic system variables and the surrounding environmental variables.



Validation of food intake biomarkers



Dragsted *et al.* *Genes & Nutrition* (2018) 13:14
<https://doi.org/10.1186/s12263-018-0603-9>

Genes & Nutrition

REVIEW

Open Access

Validation of biomarkers of food intake—critical assessment of candidate biomarkers



L. O. Dragsted^{1*}, Q. Gao¹, A. Scalbert³, G. Vergères⁴, M. Kolehmainen⁵, C. Manach⁶, L. Brennan⁷, L. A. Afman⁸, D. S. Wishart⁹, C. Andres Lacueva^{10,11}, M. Garcia-Aloy^{10,11}, H. Verhagen^{12,13}, E. J. M. Feskens⁸ and G. Praticò^{1,2}

Abstract

Biomarkers of food intake (BFIs) are a promising tool for limiting misclassification in nutrition research where more subjective dietary assessment instruments are used. They may also be used to assess compliance to dietary guidelines or to a dietary intervention. Biomarkers therefore hold promise for direct and objective measurement of

Food groups covered in the FoodBALL project



spices and herbs



alcoholic beverages



legumes



fruit and vegetables



**non-alcoholic
beverages**



fats and oils



confectionary



Meats, eggs and dairy



cereals and wholegrain

Food groups and subgroups

Dairy products

Milk

Butter

Cheese

Yoghurt

Whey

Casein



Eggs



Enriched Eggs



Foods of Animal Origin

Meat



Red Meat

Fish and Seafood

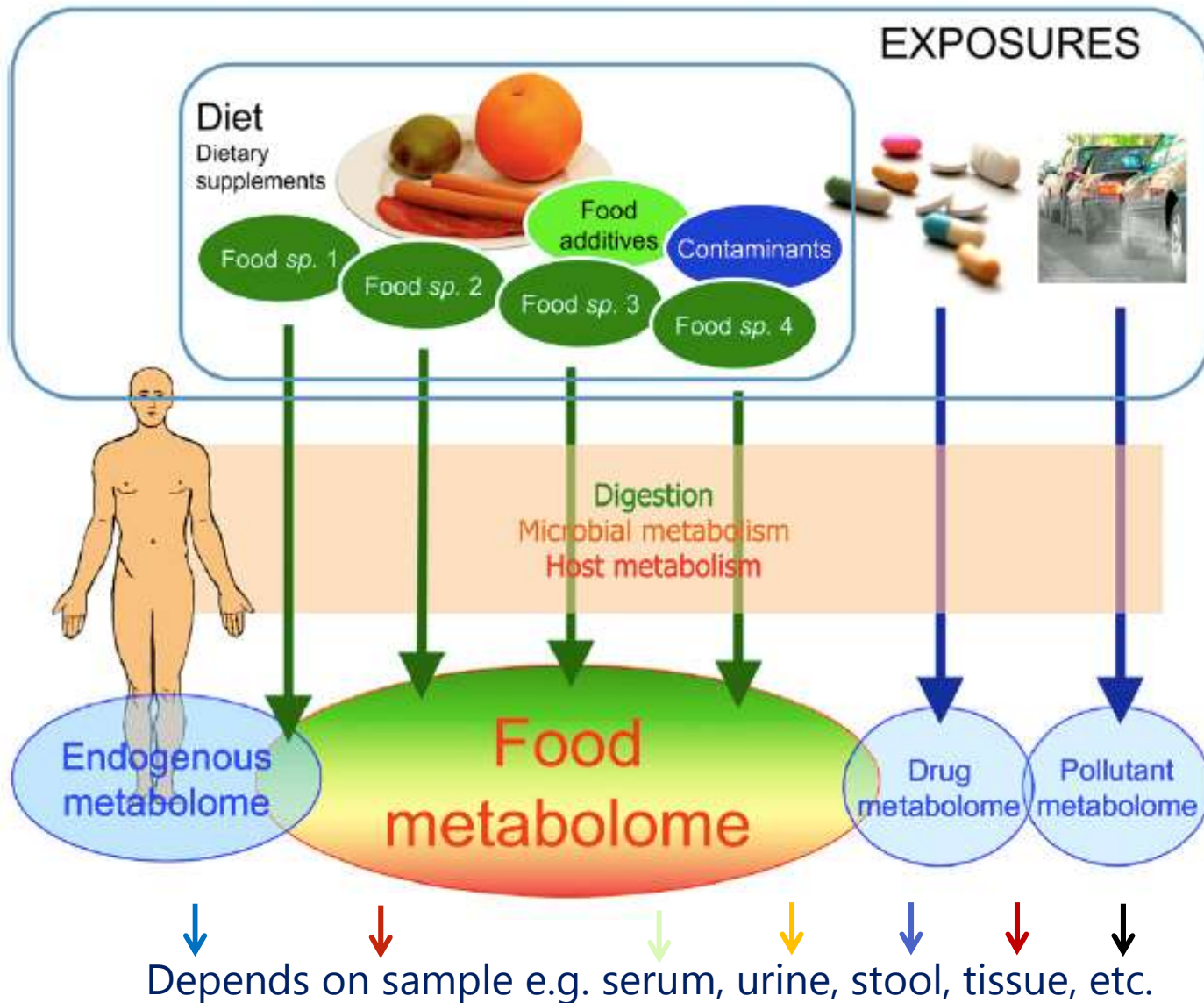
Poultry and other
white meat

Offal meat

Processed Meat
Heated Meat



The food- and other metabolomes



LC-MS metabolomics workflow

Sample extraction (removal of protein and other large molecules) and analysis

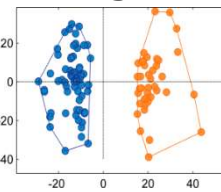
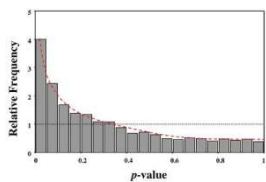


Data analysis

Data is pre-processing

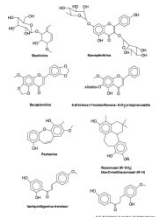
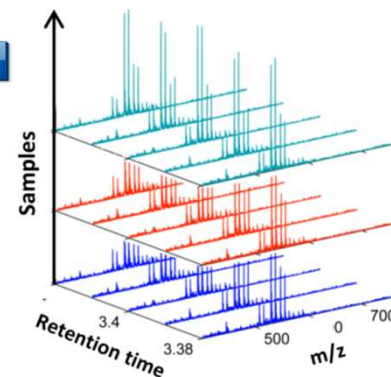
Univariate/FDR

PCA
ASCA
PLS-DA



PREPROCESSED DATA

	Feature 1	Feature 2	Feature n
Ret.T	0.81	0.82	...
m/z	50.57	100.85	...
Sample 1	45534	5445	...
Sample 2	54	425	...
Sample 3	561	538	...

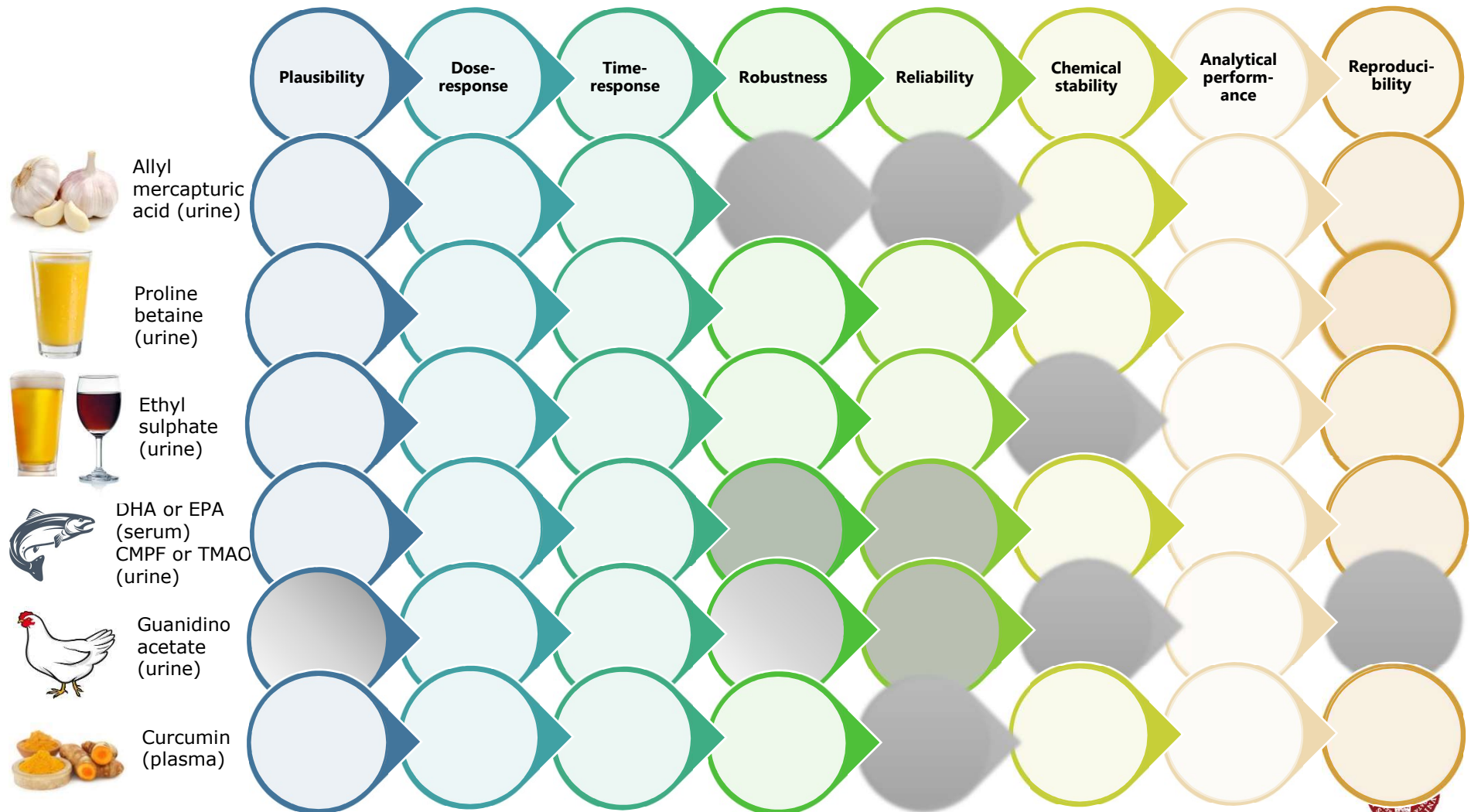


Feature identification and biological interpretation





Some of the better validated food intake biomarkers



Others include markers of WG rye and wheat, some legumes, oils, meat, and dairy

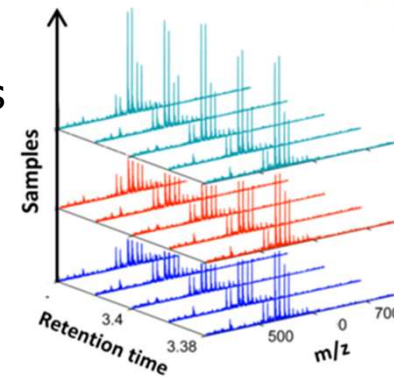
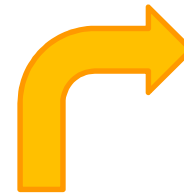
MEAL STUDY DESIGNS

– finding biomarkers with metabolomics

Test Day

No test food consumption 2 days prior

Metabolomics



330 ml Test Food/Drink

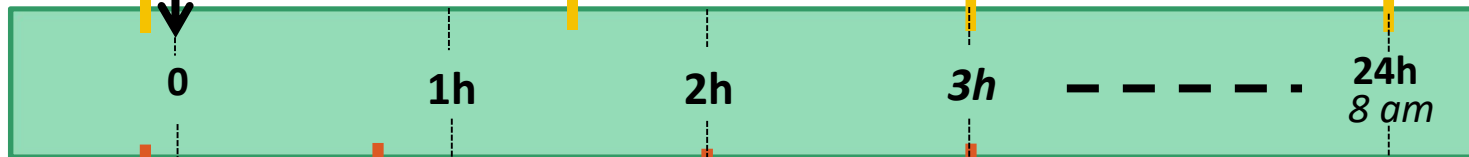


+ a suitable control food



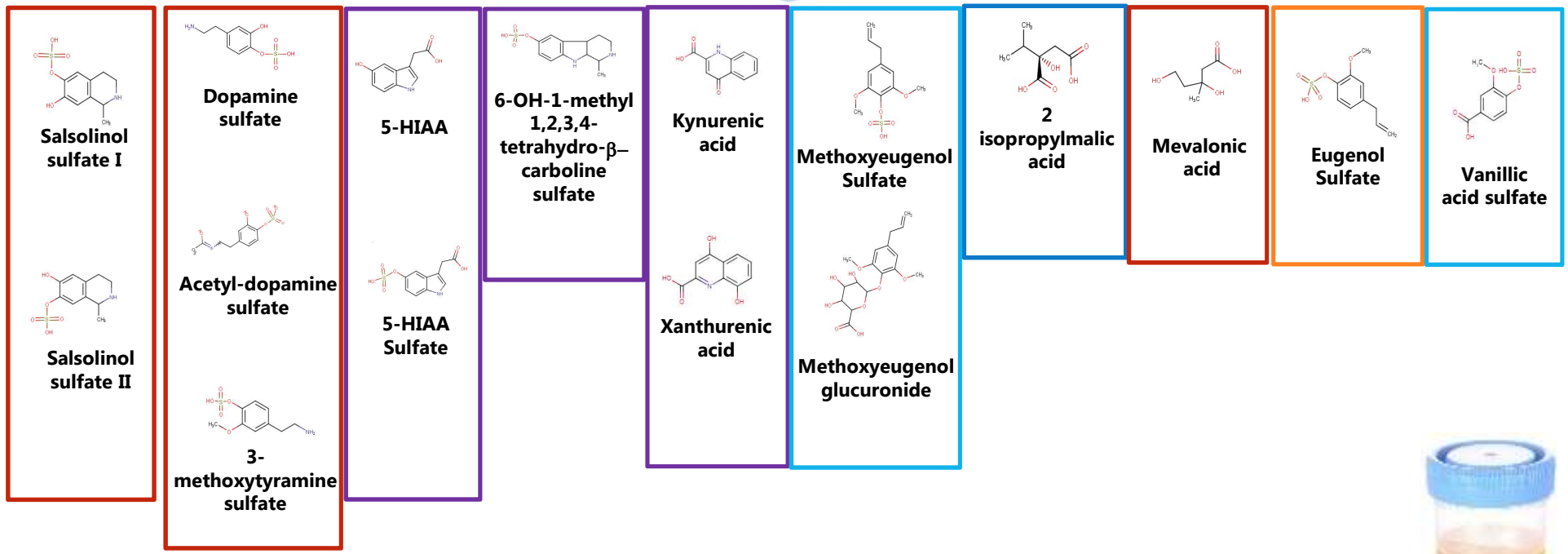
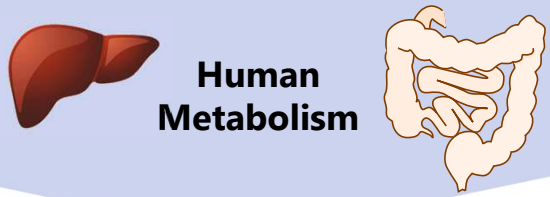
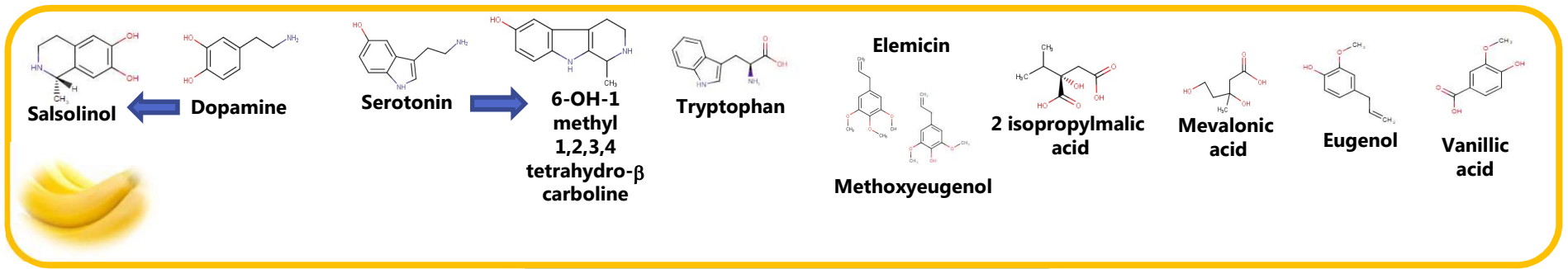
Urine Collection

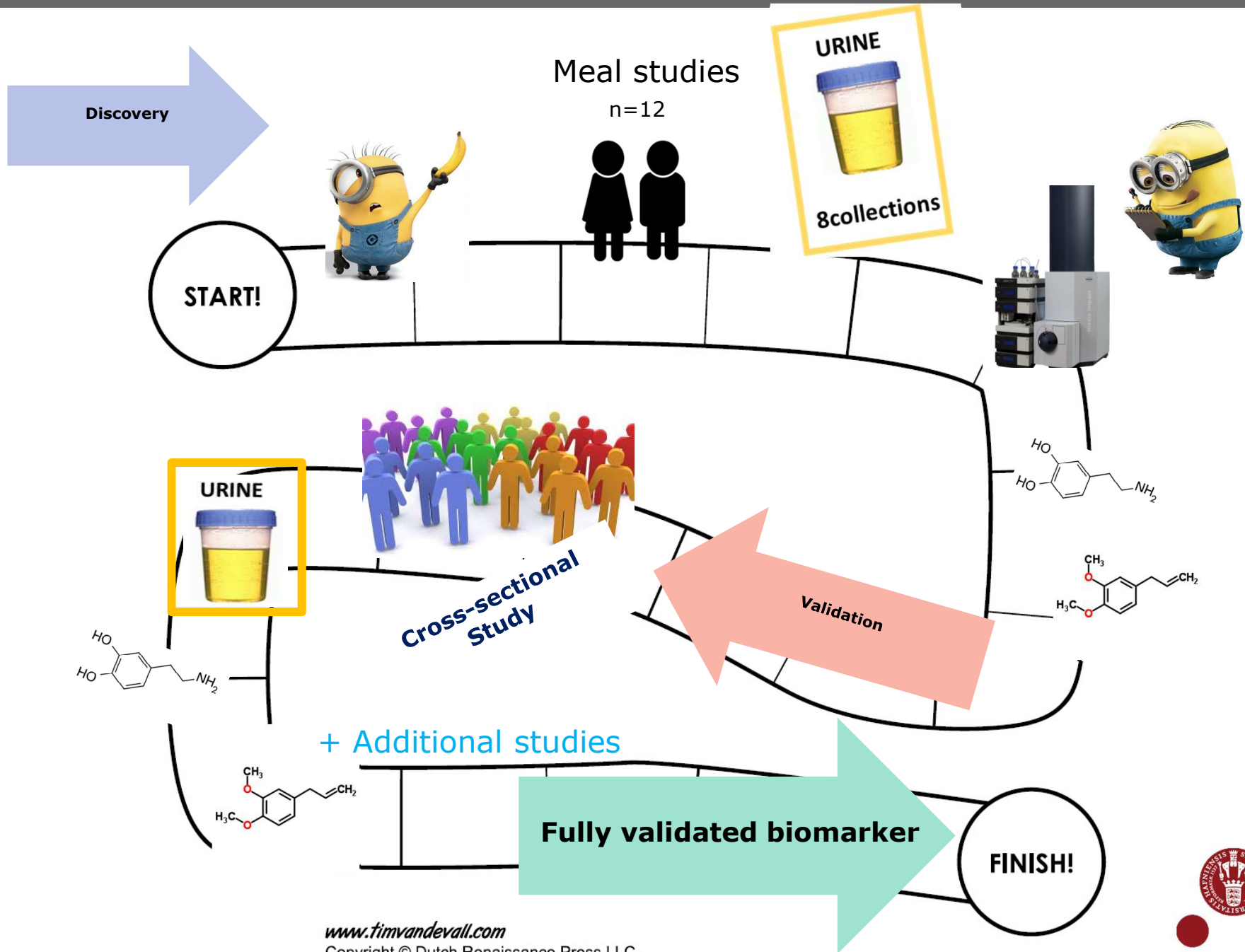
24 h pooled



Blood Collection





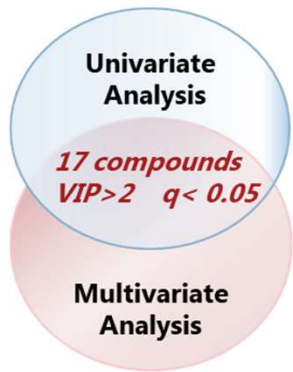


MultiBiomarker for banana intake

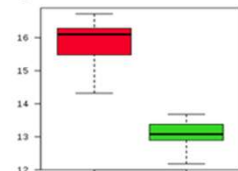


Discovery

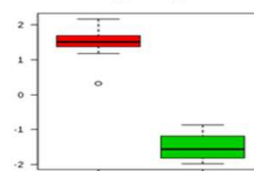
Validation



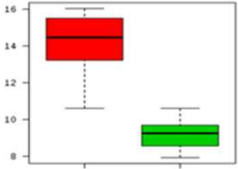
Dopamine sulfate



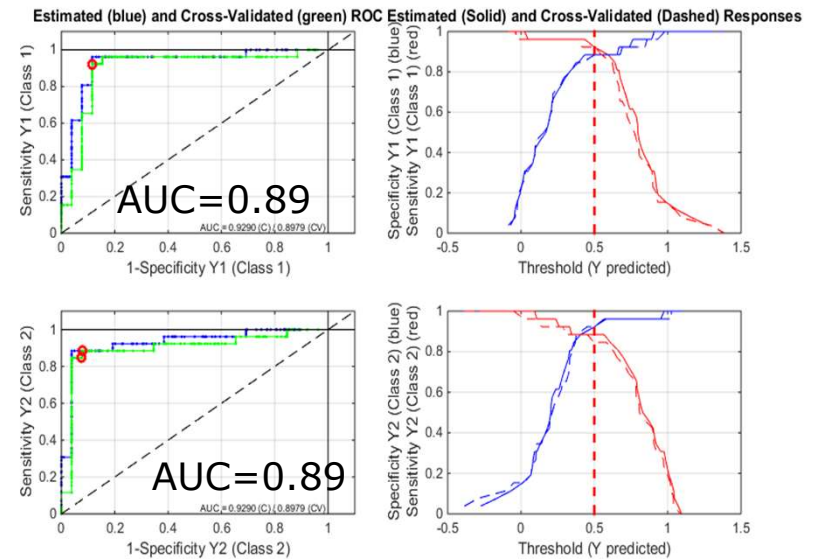
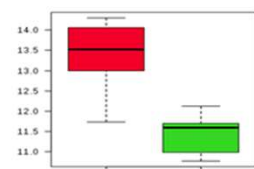
Metoxyeugenol



Salsolinol sulfate



Mevalonic Acid



The KarMen Study

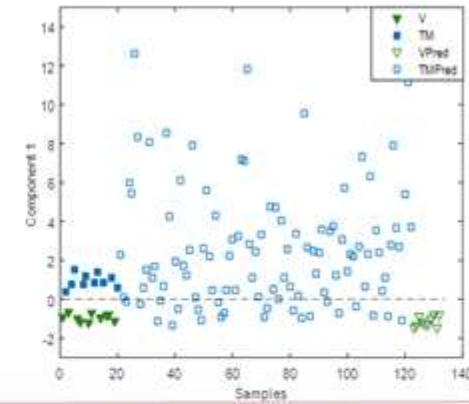
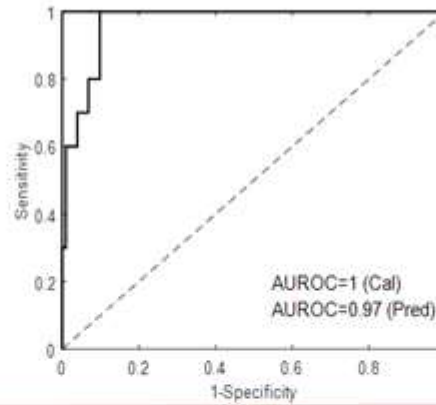


Meat multimarkers – stepwise assessment

Step 1

Omnivorous Vs. Vegetarians

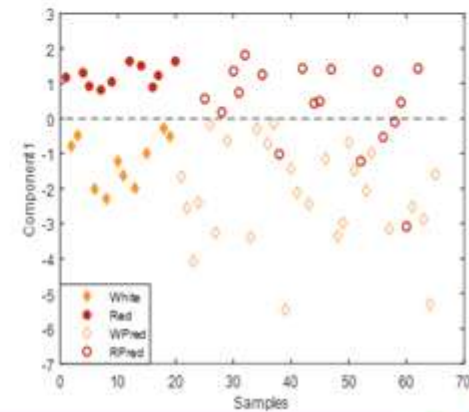
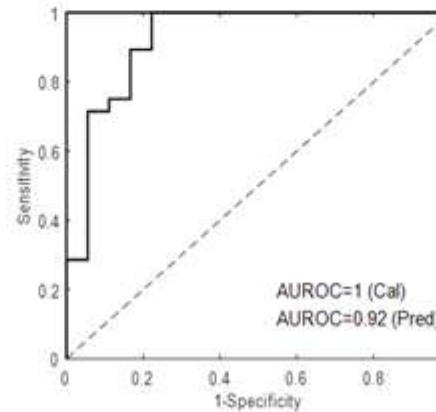
Anserine		
	TM	V
TM	86	17
V	0	10



Step 2

Red vs. white meats

Ans : Car, Ans, Car, Cre		
	R	W
R	12	6
W	0	28

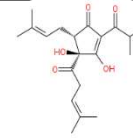


PREVIEW

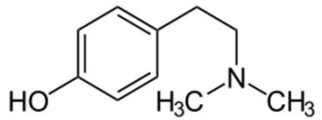


Hypothesis:

Beer biomarkers = dried barley germ markers
 + boiled hops markers
 + fermentation
 (+ biotransformation and/or degradation)

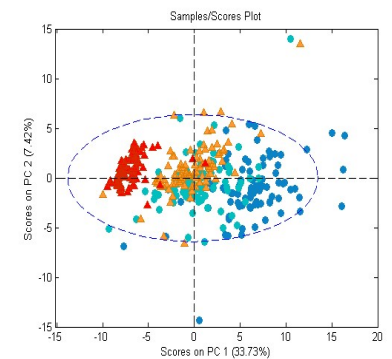
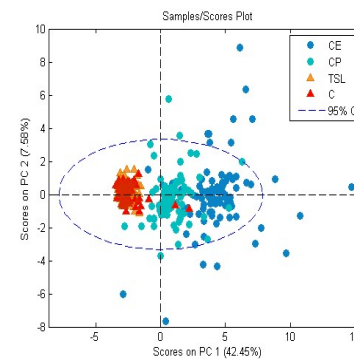
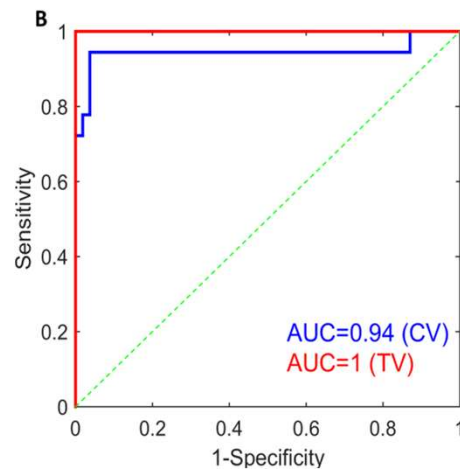
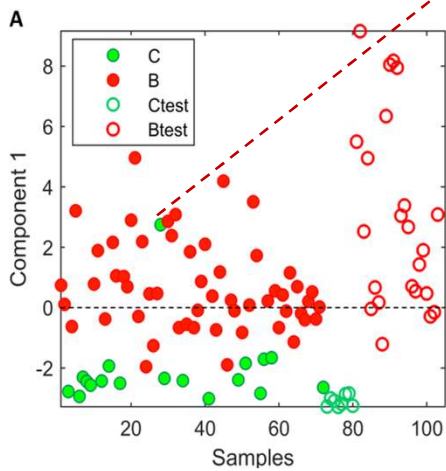
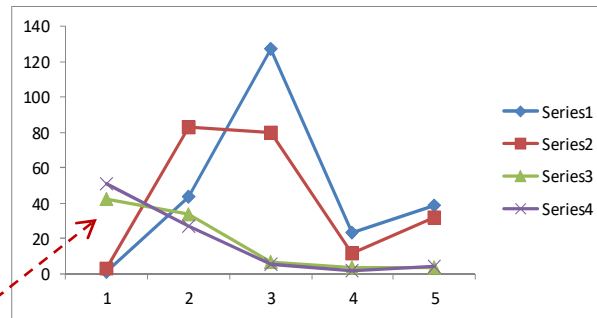
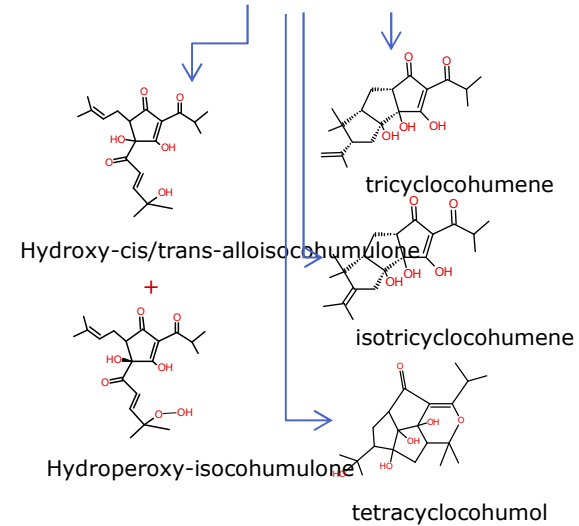
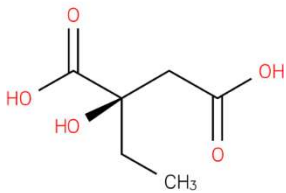


e.g. Iso- α -acids

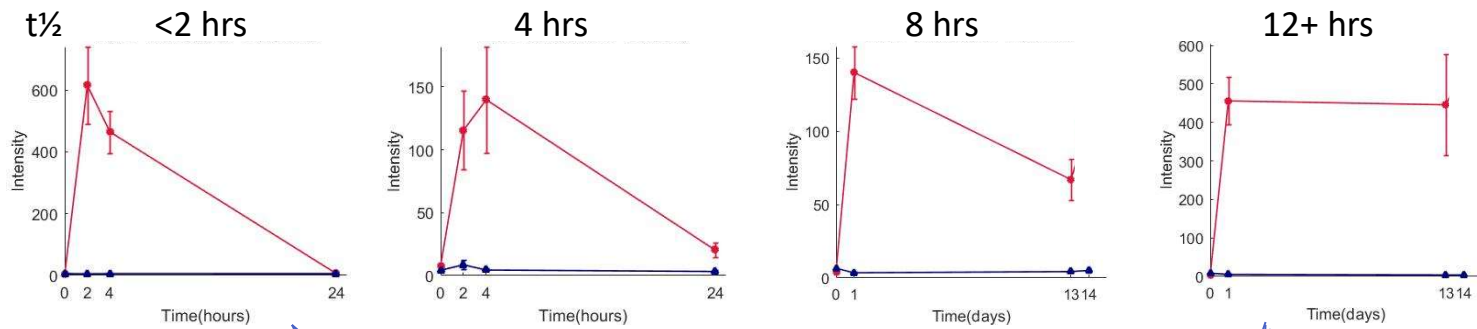


e.g. hordenine

e.g. ethyl malate (not EtOH !)



The interplay of kinetics, consumption frequency, and sampling frequency



Food Consumption intervals

Sampling frequencies

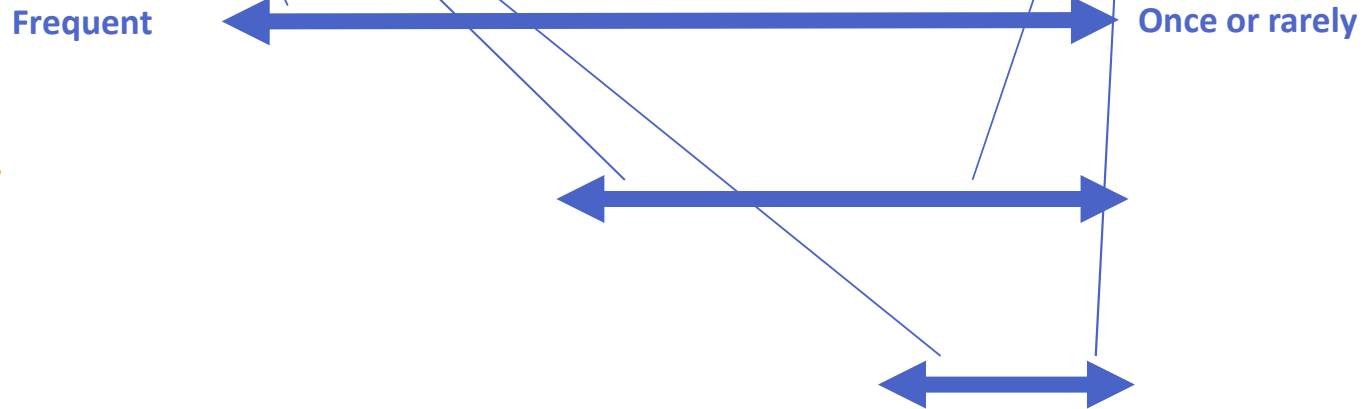
Rarely
(~monthly)

Frequent

Once or rarely

Intermittently
(~weekly)

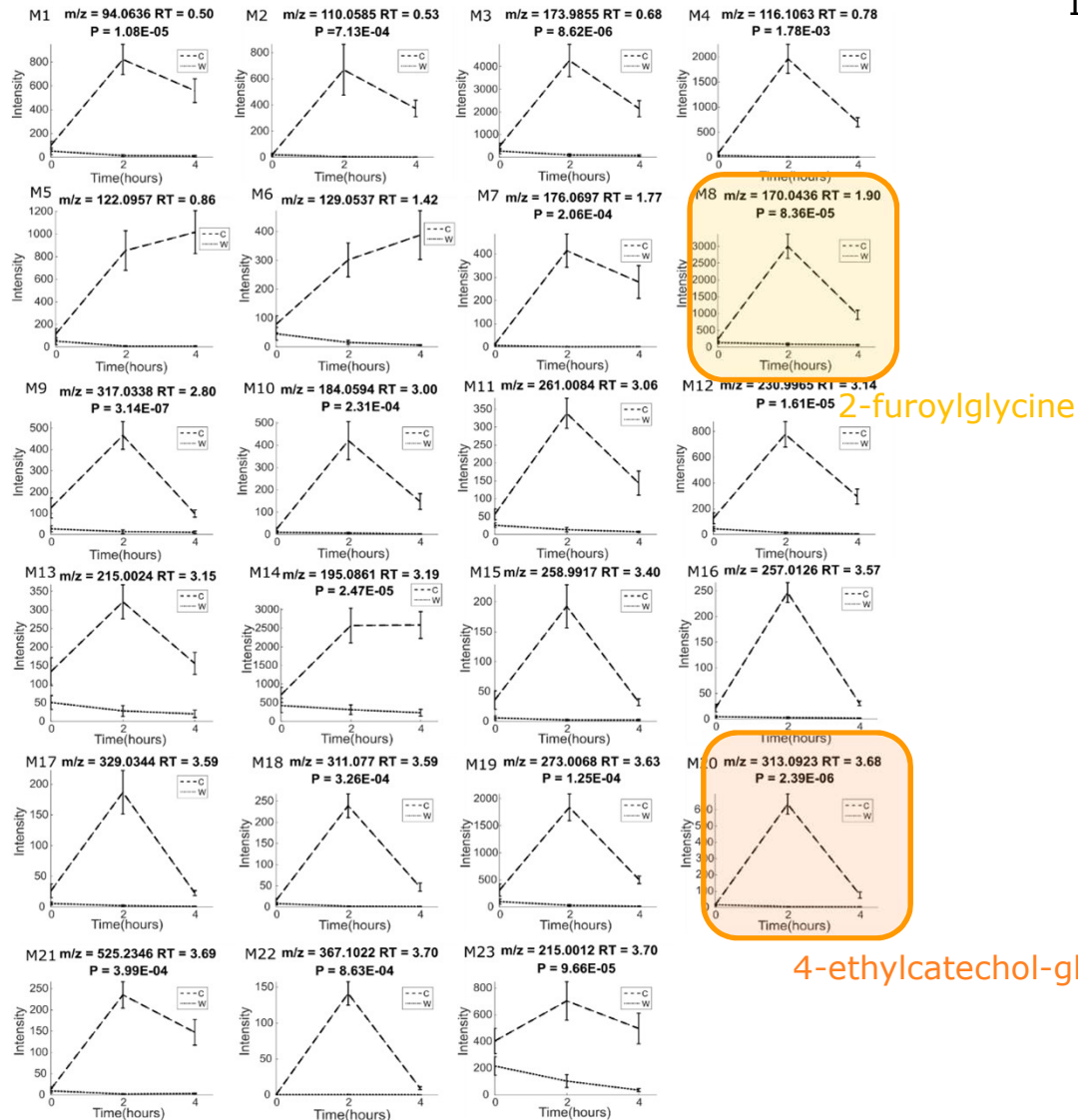
Frequently
(~daily)



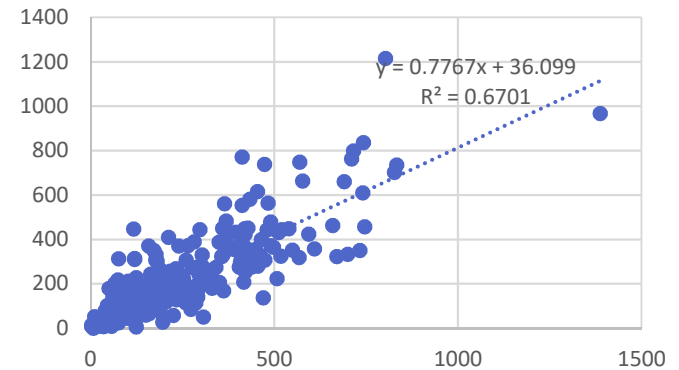
Using several markers may provide better coverage due to variation in kinetics



Markers of recent coffee intake

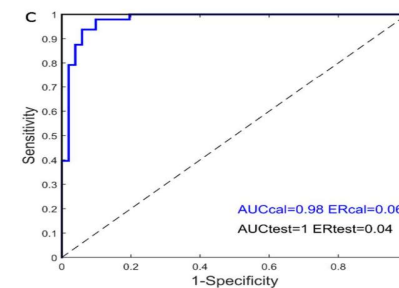


Independent analyses in NU-AGE samples

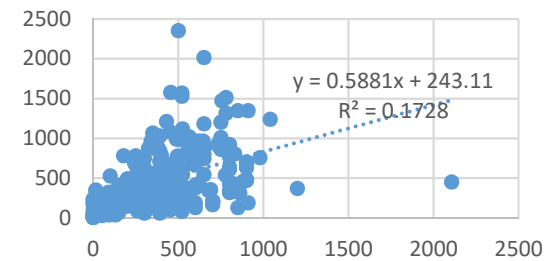


Concordance with Nu-AGE 24hr recall

qualitative

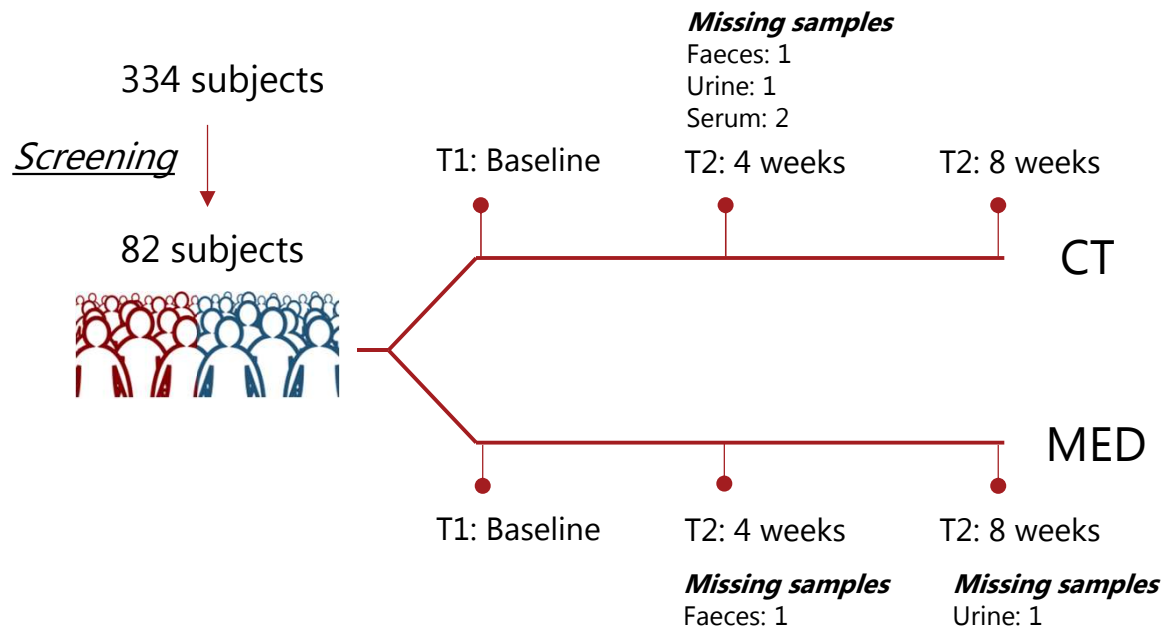


quantitative



UNINA intervention

<https://clinicaltrials.gov/ct2/show/study/NCT03071718>
 ClinicalTrials.gov Identifier: NCT03071718



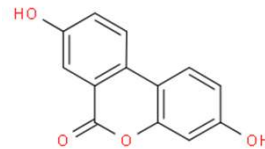
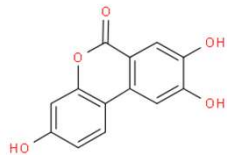
- Data**
- Anthropometrics
 - Food diaries
 - Metabolic markers
 - Metagenomics (16S+WGS)
 - **Metabolomics**

Biomarkers of nut consumption

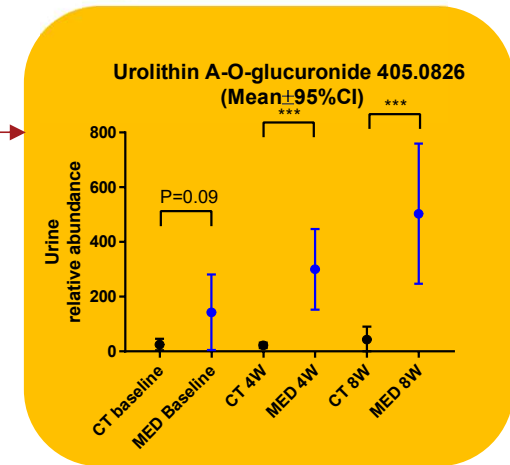
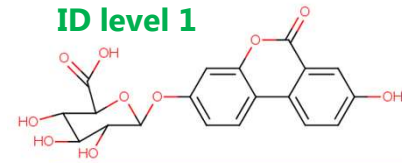
Found in e.g. pomegranate, walnuts, berries and grapes

→ Ellagitannins

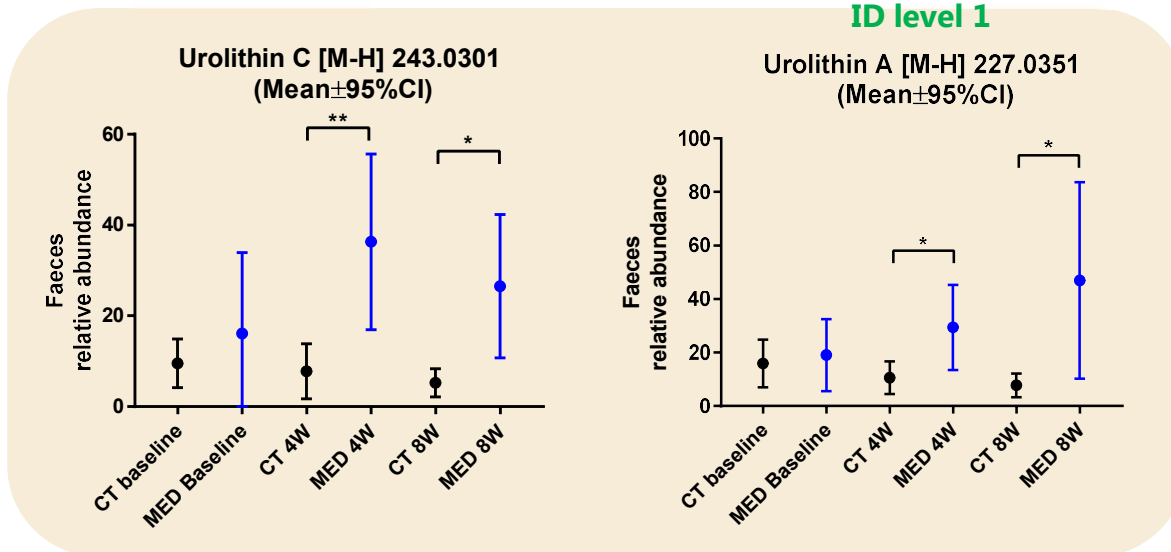
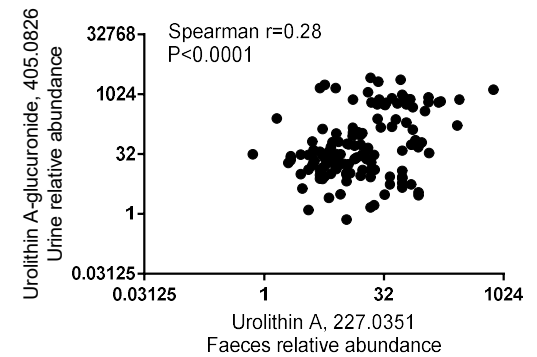
Gut microbiota



ID level 1



Comparable results in urine metabolome



Diet level classification **SYSDIET** healthy Nordic diet vs. average



Study design and subjects

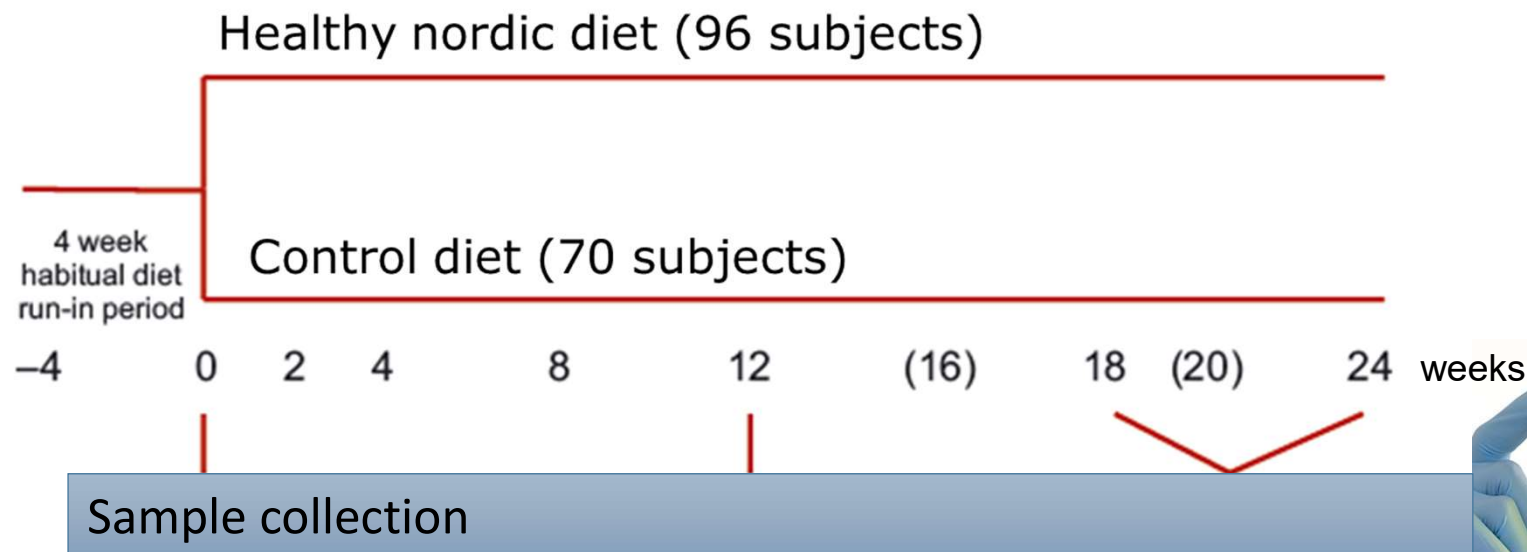
A randomized controlled multi-centre intervention

- Finland (*Kuopio* and *Oulu*)
- Iceland (*Reykjavik*)
- Sweden (*Lund* and *Uppsala*)
- Denmark (*Aarhus*)

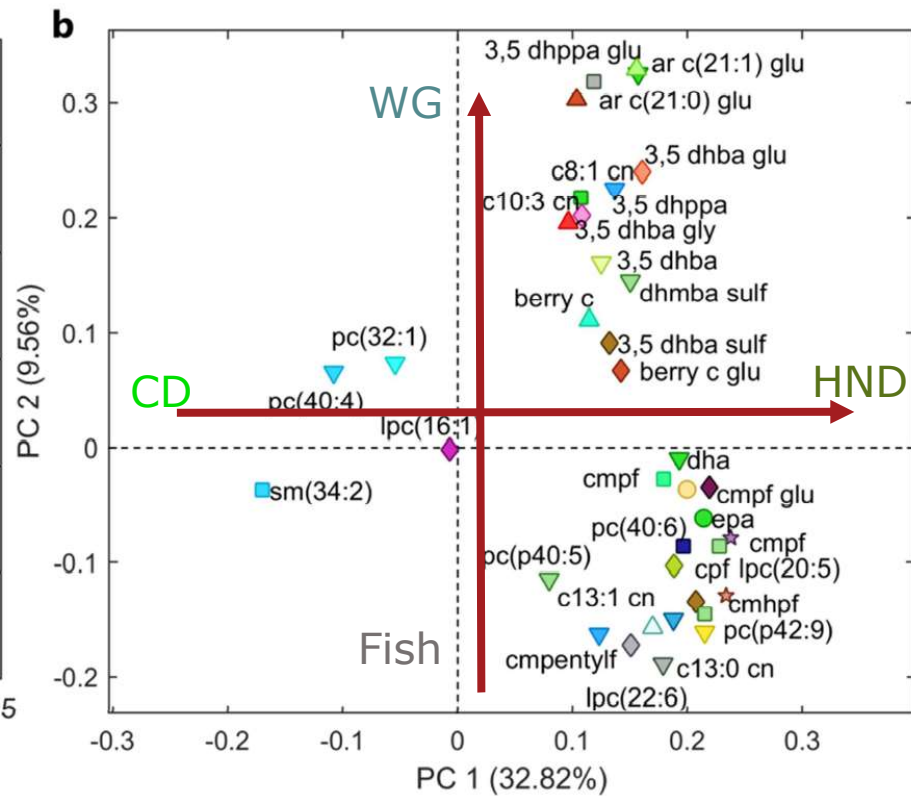
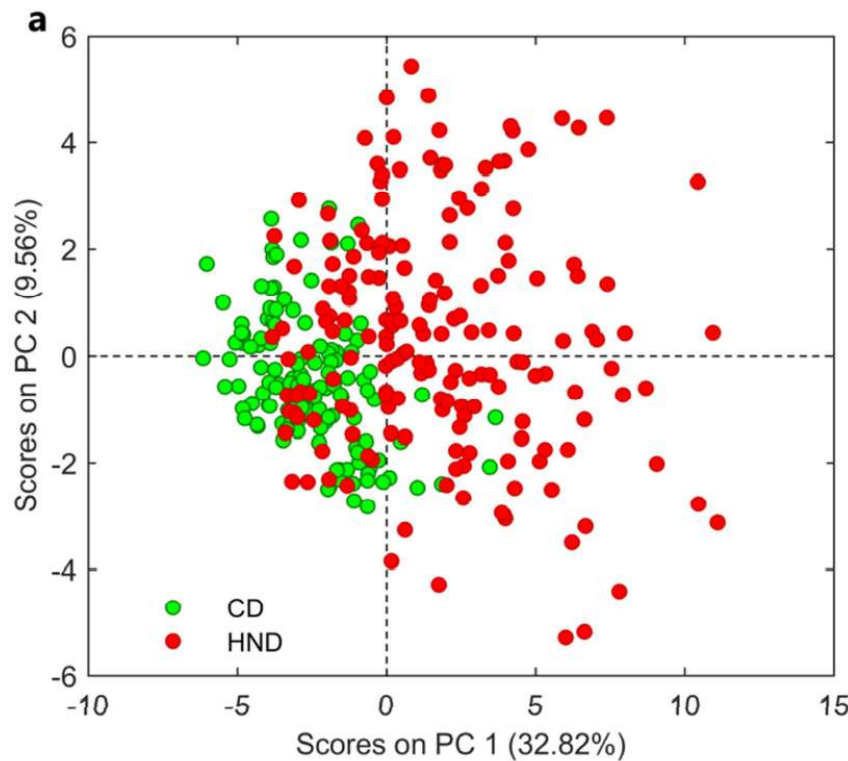


Inclusion criteria

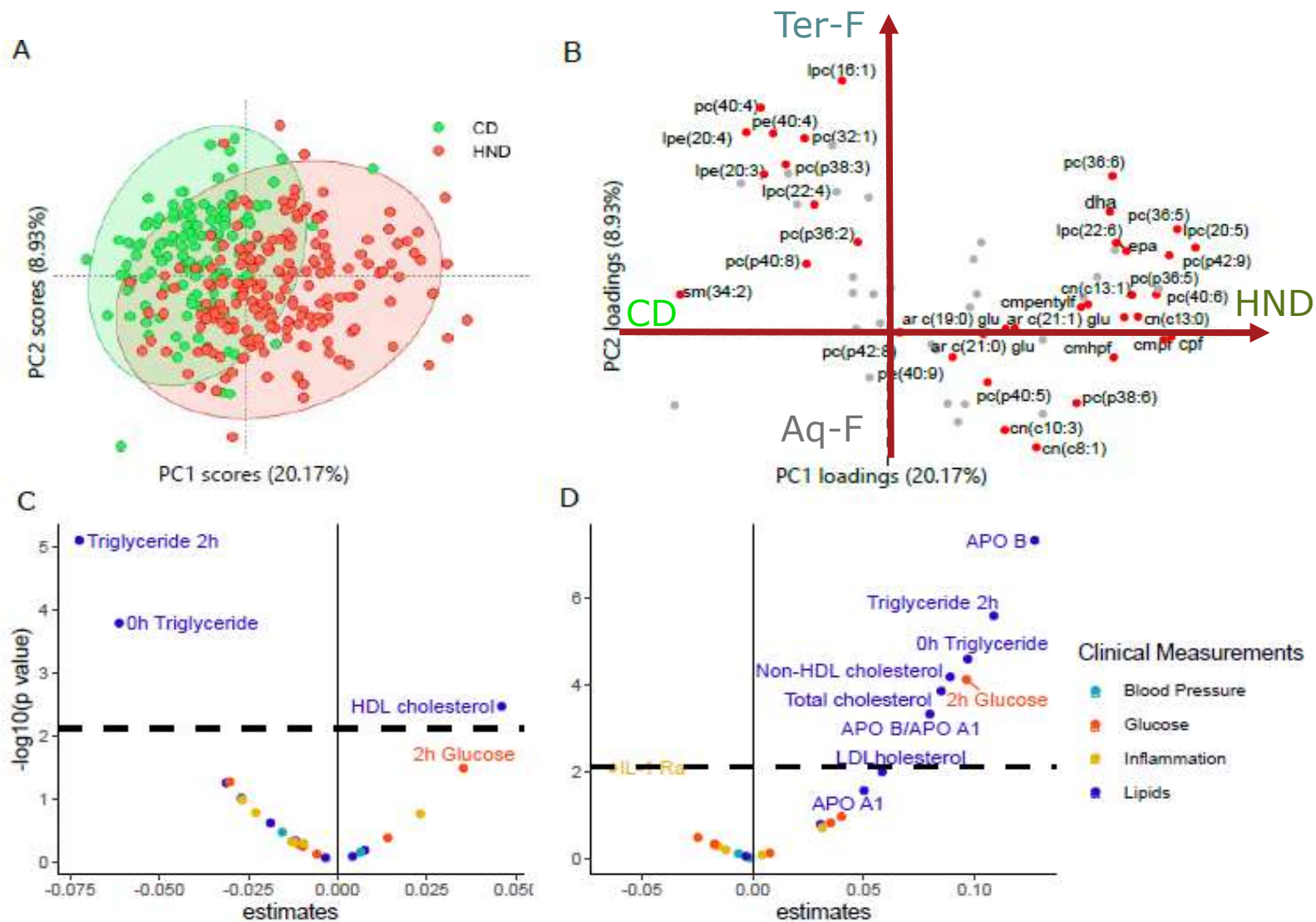
Age 30–65 years, BMI 27–38 kgm⁻²



Classifying urine metabolites – Nordic diets



Classifying serum metabolites – Nordic diets



Current state of Biomarkers of Food Intake

Issues:

- Substitution of dietary records doesn't seem possible at this time
- The one biomarker/one food approach doesn't seem realistic

Reframed purpose

- To increase the trust in dietary assessment
- To get a more refined idea of the human diet (foods vs. food groups)
- To identify misreporters in observational studies and non-compliant participants in intervention studies, and to correct for them
- To gain objectivity, though not necessarily accuracy
- To assess adherence to a dietary pattern



More useful correlates to health
(by correcting for measurement errors)

Conclusions and funding

By accurately classifying, validating and combining biomarkers it is possible to

- 1) Classify the recent intake of several foods and drinks correctly
- 2) Identify misreporting in short-term dietary assessment
- 3) Apply markers in longer-term clinical studies to assess dietary intakes

However, much basic work on biomarkers is still needed to optimize their use and figure out the quantitative aspects!

Support

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- 2) **EU FP7 grant agreement # 312057** supporting the PREVIEW project, (PREvention of diabetes through lifestyle Intervention and population studies in Europe and around the World),
- 3) **Carlsberg Foundation Semper Ardens and postdoc grants** to Lars O. Dragsted and Rastislav Monosic.



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Nu-Age collaborators: Agnes Berendsen, Edith Feskens

DINAMIC collaborators:

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.. and the SYSDIET consortium

Original Article

Journal of INTERNAL MEDICINE

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Effects of an isocaloric healthy Nordic diet on insulin sensitivity, lipid profile and inflammation markers in metabolic syndrome – a randomized study (SYSDIET)

■ M. Uusitupa^{1,2}, K. Hermansen³, M. J. Savolainen⁴, U. Schwab^{1,5}, M. Kolehmainen¹, L. Brader³, L. S. Mortensen³, L. Cloetens⁶, A. Johansson-Persson⁶, G. Onning⁶, M. Landin-Olsson⁷, K.-H. Herzig^{8,9}, J. Hukkanen⁴, F. Rosqvist¹⁰, D. Iggman^{10,11}, J. Paananen¹, K. J. Pulkki¹², M. Siloaho^{1,4,13}, L. Dragsted¹⁴, T. Barri¹⁴, K. Overvad^{15,16}, K. E. Bach Knudsen¹⁷, M. S. Hedemann¹⁷, P. Arner¹⁸, I. Dahlman¹⁸, G. I. A. Borge¹⁹, P. Baardseth¹⁹, S. M. Ulven²⁰, I. Gunnarsdottir²¹, S. Jónsdóttir²¹, I. Thorsdottir²¹, M. Orešič²², K. S. Poutanen^{1,22}, U. Risérus¹⁰ & B. Akesson^{6,23}

From the ¹Institute of Public Health and Clinical Nutrition, University of Eastern Finland; ²Research Unit, Kuopio University Hospital, Kuopio, Finland; ³Department of Medicine and Endocrinology MEA, Aarhus University Hospital, Aarhus, Denmark; ⁴Institute of Clinical Medicine, Department of Internal Medicine, University of Oulu, Oulu, ⁵Institute of Clinical Medicine, Internal Medicine, Kuopio University Hospital, Kuopio, Finland; ⁶Biomedical Nutrition, Pure and Applied Biochemistry, Lund University, ⁷Department of Endocrinology, Skåne University Hospital, Lund, Sweden; ⁸Institute of Biomedicine and Biocenter of Oulu, University of Oulu, Oulu, ⁹Department of Psychiatry, Kuopio University Hospital, Kuopio, Finland; ¹⁰Department of Public Health and Caring Sciences, Clinical Nutrition and Metabolism, Uppsala University, Uppsala, ¹¹Center for Clinical Research Dalarna, Falun, Sweden; ¹²Eastern Finland Laboratory Centre and Department of Clinical Chemistry, University of Eastern Finland; ¹³Institute of Clinical Medicine, University of Eastern Finland, Kuopio, Finland; ¹⁴Department of Nutrition, Exercise and Sport, University of Copenhagen, Copenhagen, ¹⁵Department of Epidemiology, School of Public Health, Aarhus University, ¹⁶Department of Cardiology, Aalborg Hospital, Aarhus University Hospital, ¹⁷Department of Animal Science, Aarhus University, Aarhus, Denmark; ¹⁸Department of Medicine (H7), Karolinska Institute, Stockholm, Sweden; ¹⁹Nofima, Norwegian Institute of Food, Fisheries and Aquaculture Research, Ås; ²⁰Department of Health, Nutrition and Management, Faculty of Health Sciences, Oslo and Akershus University College of Applied Sciences, Oslo, Norway; ²¹Unit for Nutrition Research, University of Iceland and Landspítali – The National University Hospital of Iceland, Reykjavik, Iceland; ²²VTT Technical Research Centre of Finland, Espoo, Finland; and ²³Department of Clinical Nutrition, Skåne University Hospital, Lund, Sweden