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SEEDFOOD

Unlocking the biophysical properties of rapeseed storage proteins:

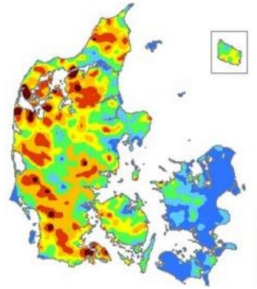
Tailoring innovative strategies for
sustainable protein production

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Department of Food Science, UCPH

KØBENHAVNS UNIVERSITET

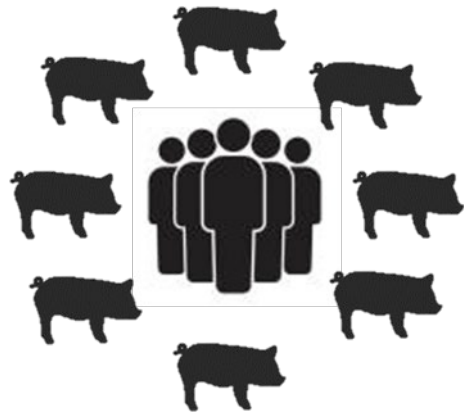


Danish agriculture



Over 60% of total area is cultivated

Over twice as many pigs as people



Wide range of dairy products even available in Australia

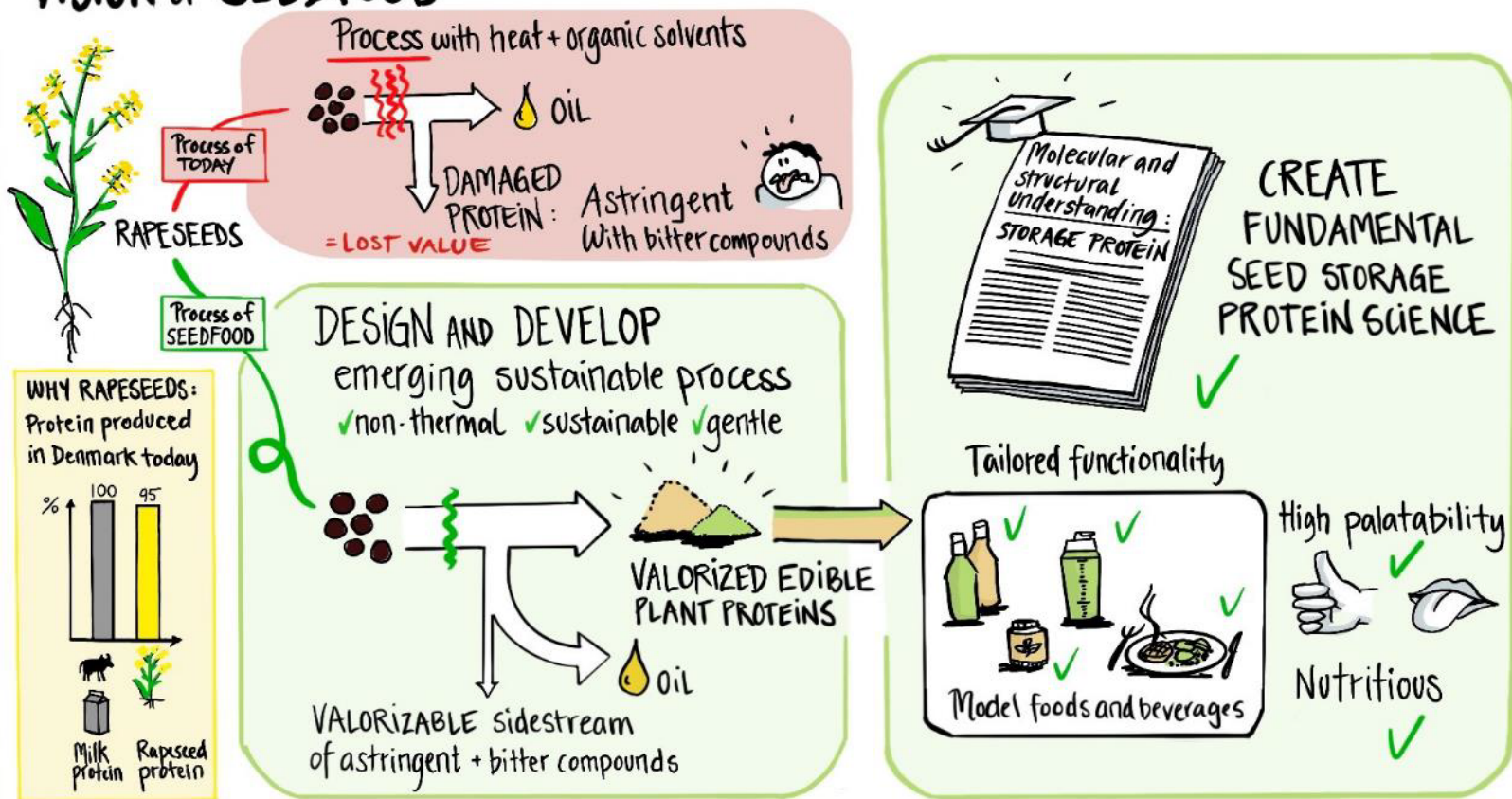


Large focus from political and public side to push the green transition

Why SEEDFOOD?

⇒ High impact – transformation and valorization of existing source of plant protein

VISION OF SEEDFOOD



- ✓ Produced in high amounts
- ✓ Good nutritional quality
- ✓ Good digestibility score* compared to other plant proteins
- ✓ New gentle processes for isolation of rapeseed proteins
- ✓ Structure/techno-functional relationship in food science context

60 mio tons/year worldwide
17 mio tons/year in the EU

*PDCAAS: Protein Digestibility Corrected Amino Acid Score
Bos et al (2007) *J. Nutr.* 137, 594-600
Gilani & Lee (2003) *Encyclopedia of Food Science and Nutrition*, pp 4873-4879. Academic Press

SEEDFOOD purpose and main hypothesis



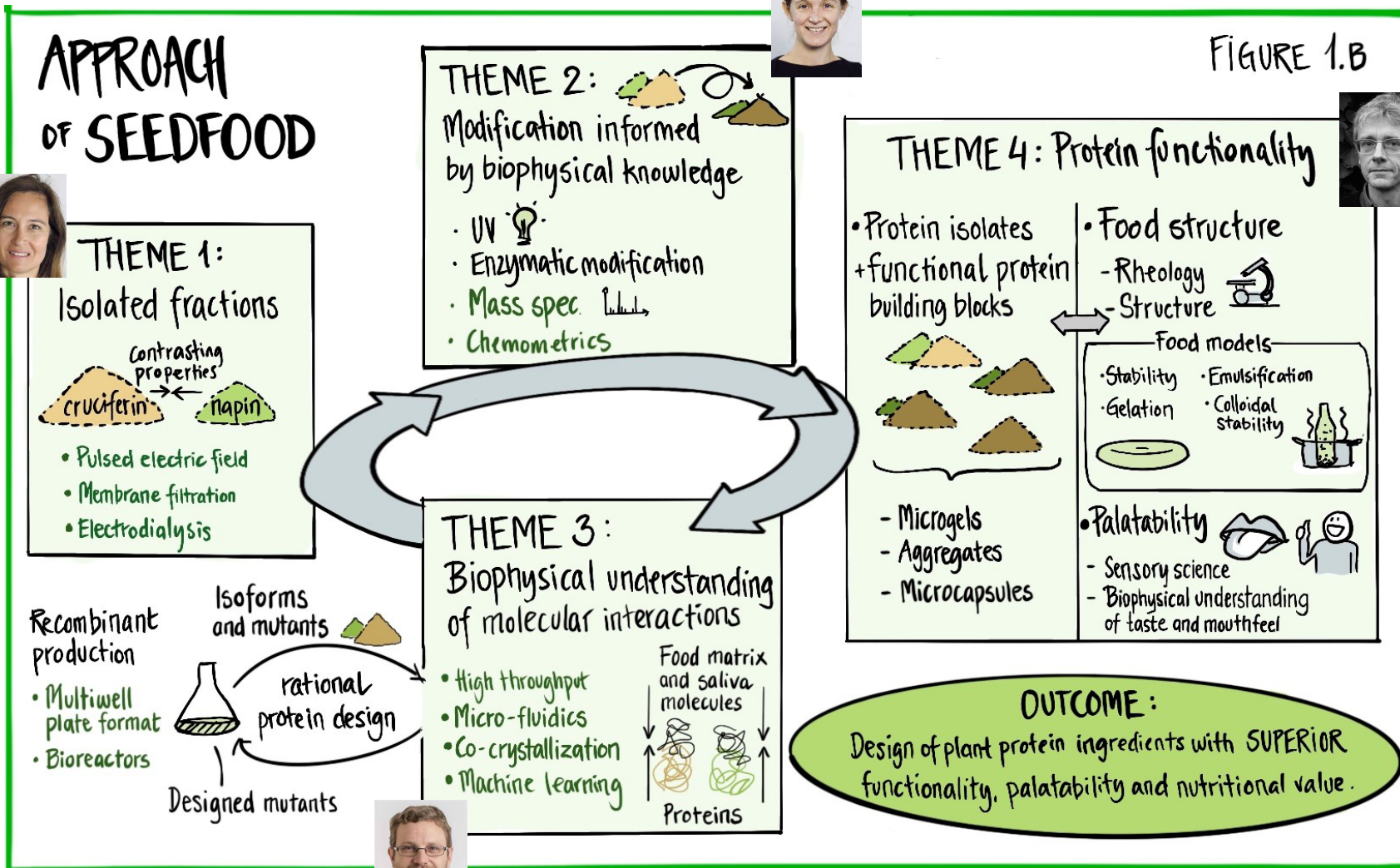
The purpose of SEEDFOOD is to advance insights in fundamental protein science of seed storage proteins to get highly **functional and palatable properties** through protein design and understanding of molecular interactions

Main hypothesis:

Rapeseed proteins can be transformed to functional and palatable proteins by gentle processing and protein modification

Cross-disciplinary research organized in four themes

Co-PI
Prof. Lilia Arhné



PI
Prof. Marianne N. Lund

FIGURE 1.B

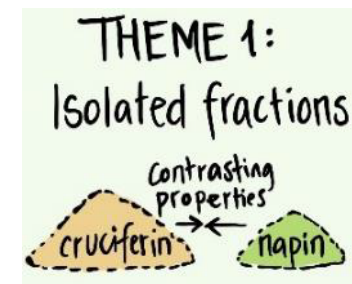


Co-PI
Prof. Taco Nicolai



Co-PI
Prof. Alexander Büll

Major rapeseed proteins: Cruciferin and napin



Cruciferin

~300-420 kDa
15 subunits of 12-60 kDa
pI ~7; T_d = 91 °C
~60% of rapeseed proteins



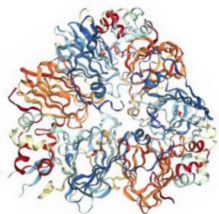
Napin

~12-16 kDa
Two subunits of ~ 4 and 9 kDa
pI 11 ; T_d = 110 °C
~20% of rapeseed proteins

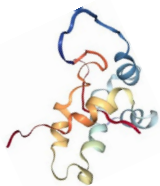
Challenges:

- ⇒ **Poor functionality: Large differences in pI, denaturation and gelation temperatures**
- ⇒ **Glucosinolates, phenolics, phytate and myrosinase must be removed**
- ⇒ **'Close-to commercial' rapeseed protein isolates have problems with taste and color**

Identified knowledge gaps



~60% Cruciferin
~300 - 420 kDa
pI ~7; T_d = 91 °C



~20% Napin
~12 - 16 kDa
pI 11 ; T_d = 110 °C

- ⇒ Functional properties of purified, native RPs are largely unexplored
- ⇒ Mechanism and kinetics behind aggregation is unknown for RPs
- ⇒ How can sustainable technologies be utilized for gentle RP isolation?
- ⇒ How can we modify RPs to improve functionality and palatability?
- ⇒ The mechanism of protein astringency is unknown
- ⇒ Molecular insight of interactions among plant proteins and other molecules is very limited

Working on the edge between disciplines makes it possible to create unique basic knowledge and radical solutions

Biophysical understanding



Surface properties, protein structures and their interactions on a molecular level



Systematic recombinant protein production

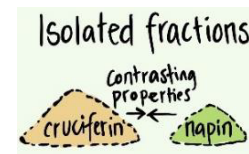


Screening tools to guide protein design

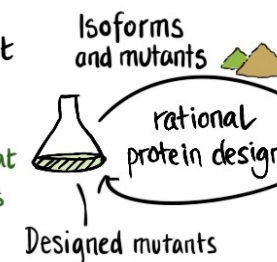


Broad range of different functionalities &

Understanding protein palatability

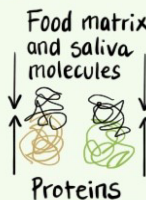


Recombinant production
• Multiwell plate format
• Bioreactors

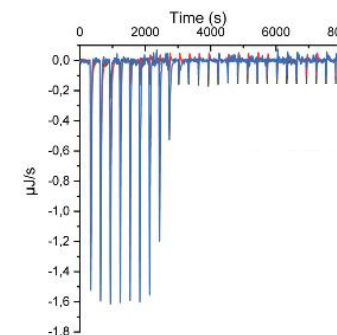


THEME 3:
Biophysical understanding of molecular interactions

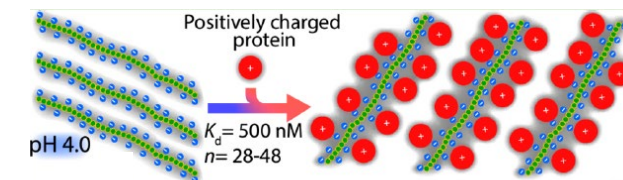
- High throughput
- Micro-fluidics
- Co-crystallization
- Machine learning



Protein-protein interactions

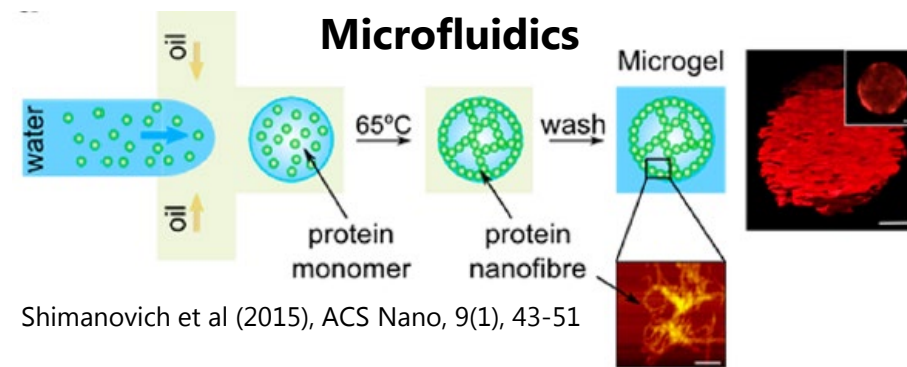


Protein-carbohydrate interactions



Madsen et al (2021), Biomacromolecules, 22(2), 649-660

Microfluidics

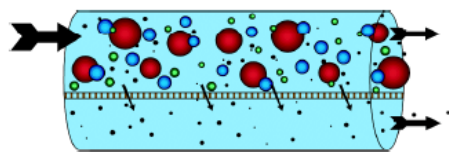
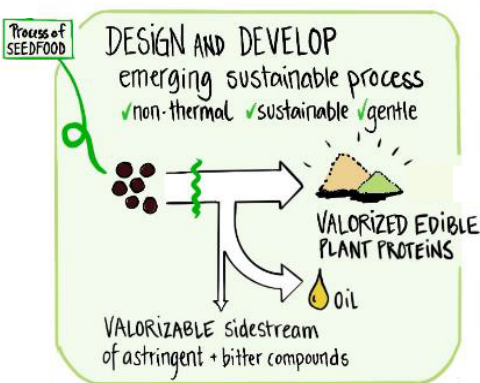


Shimanovich et al (2015), ACS Nano, 9(1), 43-51

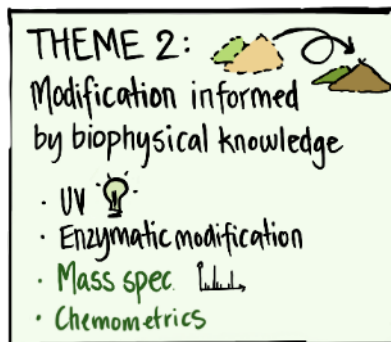
The purpose of SEEDFOOD is to advance insights in fundamental protein science of seed storage proteins to get highly functional and palatable properties through protein design and understanding of molecular interactions

How?

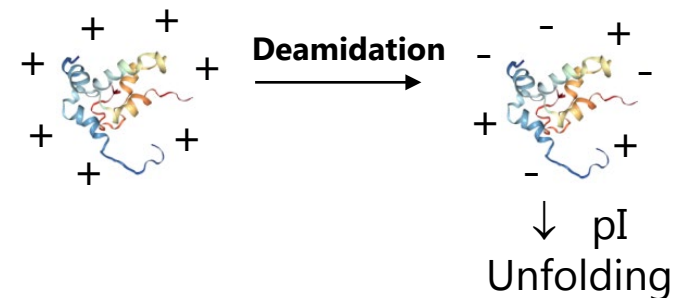
Rapeseed proteins can be transformed to functional and palatable proteins by gentle processing and protein modification



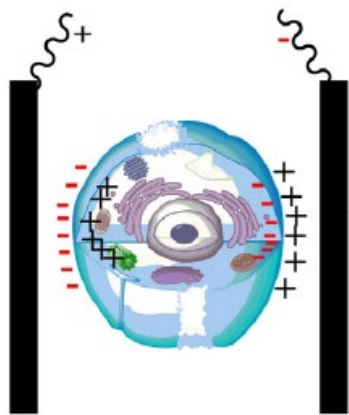
Membrane filtration



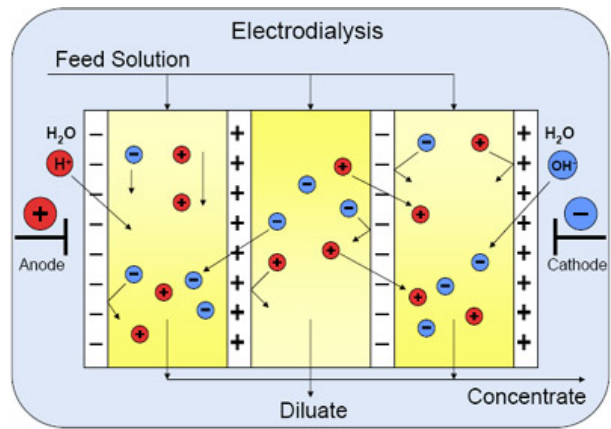
Change surface properties of napin and cruciferin



Pulsed electric field

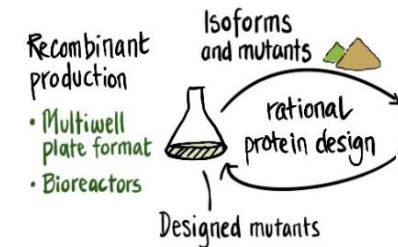
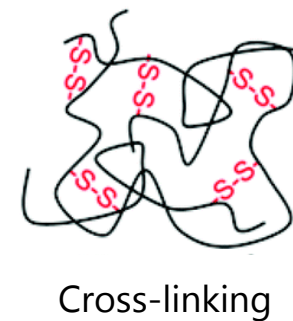


Inactivation of enzymes



Electrodialysis

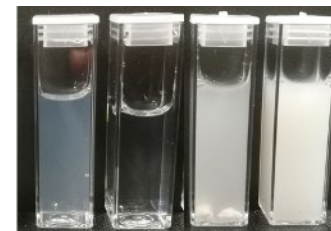
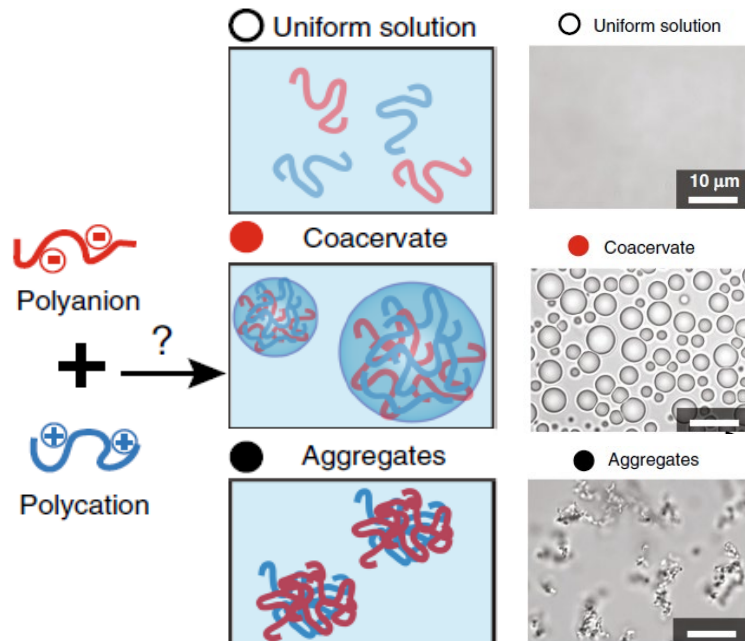
Stabilize protein structures by covalent bonds



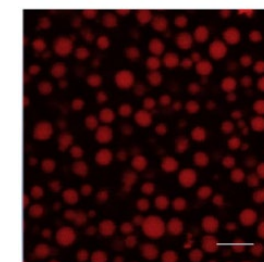
⇒ Broad range of different functionalities & understanding protein palatability

THEME 4: Protein functionality

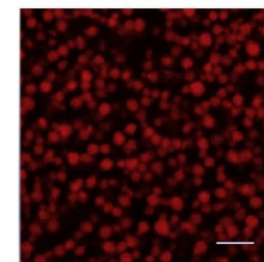
- Food structure
 - Rheology
 - Structure
- Food models
 - Stability
 - Emulsification
 - Gelation
 - Colloidal stability



Colloidal stability



Coacervates



Microgels

heat →

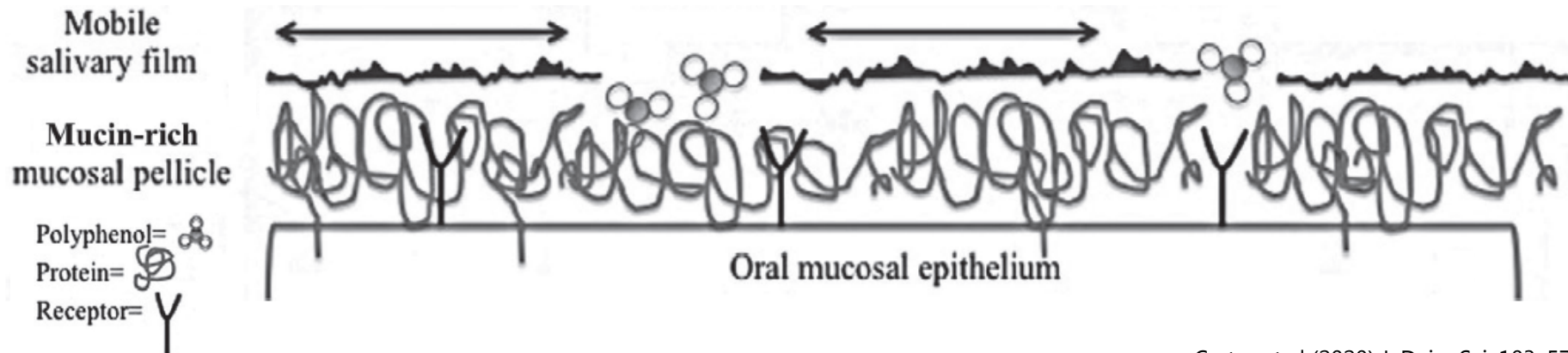
Cakmak et al (2020) Nature Commun. 11, 5949

Chen et al (2020) Food Hydrocoll. 105, 105853

Palatability

- Sensory science
- Biophysical understanding of taste and mouthfeel

Astringency: Interactions of proteins/polyphenols with mucosa



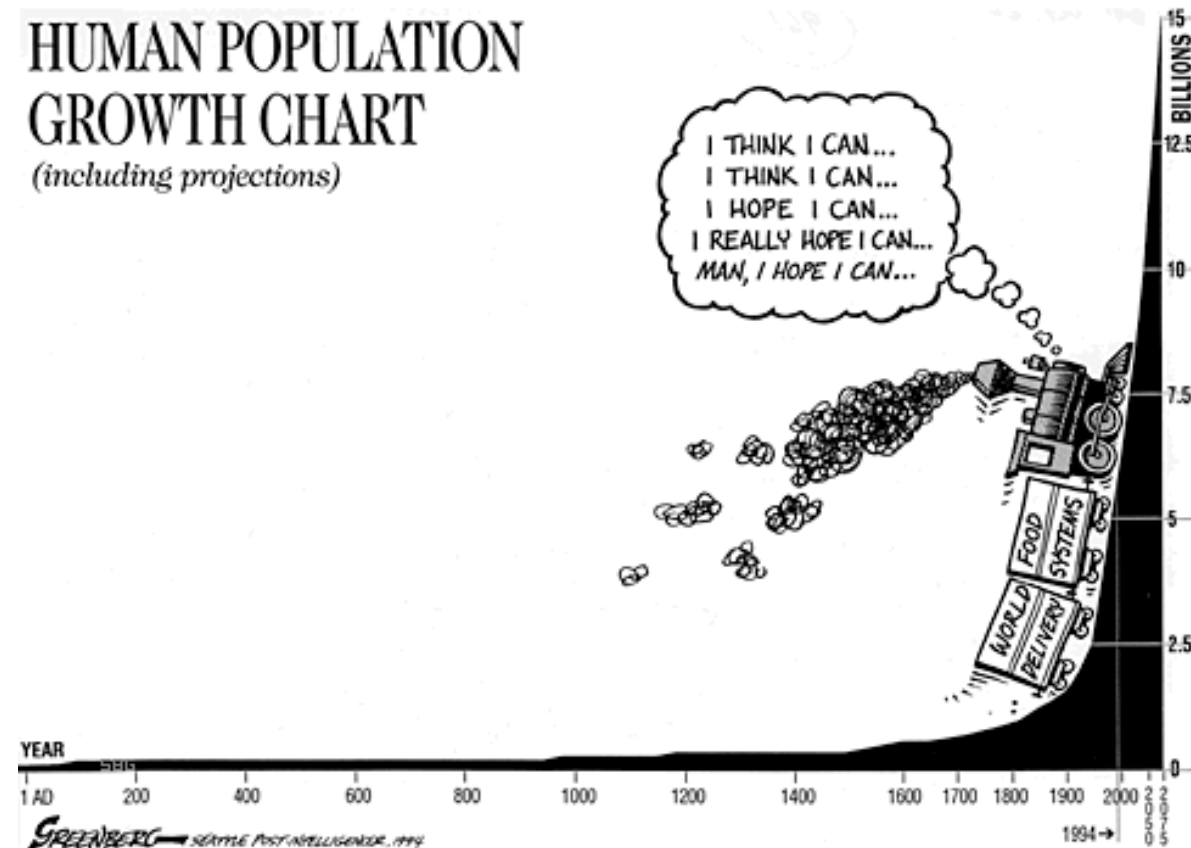
Carter et al (2020) J. Dairy Sci. 103, 5793-5804

Scientific impact and outcome

- ✓ **SEEDFOOD will provide the scientific foundation to transform seed storage proteins for food with superior functionality and high palatability and nutritional value**
- ✓ **Bring new disciplines into food science and create unique knowledge on food proteins**
- ✓ **Interdisciplinary scientific learning environment with education of >20 PhD students/postdocs**



HUMAN POPULATION GROWTH CHART
(including projections)



GREENBERG - SEATTLE POST-INTELLIGENCER, 1994

The SEEDFOOD consortium

Advisory board



Dr. Janitha Wanasundara
Saskatoon R&D Centre
Canada



Prof. Ulrich Kulozik
Technical University of Munich
Germany



Dr. Christophe Schmitt
Nestlé Research Center
Switzerland



Prof. Wender Bredie
Sensory science



Prof. Birte Svensson
Structure/function relationship



Prof. Günther Peters
Machine learning



Assoc. prof. Åsmund Rinnan
Chemometrics



Prof. Taco Nicolai
Protein functionality



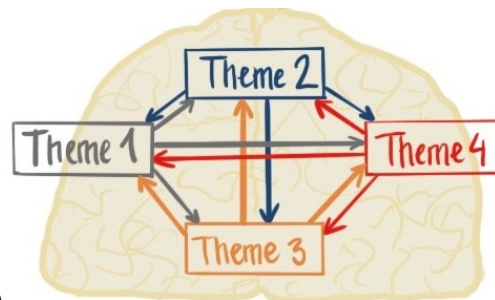
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Assis. prof. Marie S. Møller
Molecular interactions



Prof. Marianne N. Lund
Protein modifications



Prof. Alexander K. Büll
Biophysics



Adj. prof. Jacob H. Nielsen
Protein functionality



Prof. Lilia Ahrné
Processing technology



Prof. Morten Sommer & Dr. Leonie Jahn
Recombinant proteins



Assis. prof. Mahesha M. Poojary
Food analytical chemistry



Prof. Frank Lipinski
Membrane filtration



Prof. Poul Erik Jensen
Plant biochemistry

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