

AGENDA

- Introduction to microstructure work at RISE Agrifood and Bioscience
- Microstructure of bread and dough
 - Different structural components
 - Different techniques
 - Different types of bread
- How to use microstructure in product development
 - Example from frozen stored bread and dough



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Microstructure work at RISE Agrifood and Bioscience

- A large variety of different **preparation techniques** of food and other “soft” material structures
- Microscopy analysis at **different length scales**
- **Dynamic processes** (e.g. visualising phenomena over time like foam formation, aggregation, phase separation, disintegration, migration, etc.)
- **Knowledge of materials** and their properties

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Instrumentation

Instruments

- **Light microscope (LM)**
Nikon Microphot FXA, structural level:
2mm-0.5mm
- **Confocal Laser Scanning
Microscopes (CLSM)** Leica SP5
and SP2, structural level: 2mm-0.2mm
- **Transmission electron
microscope (TEM)** LEO 906E
120 kV, structural level: 20mm-5nm
- At RISE in Stockholm and Borås
**Confocal Raman, SEM, IR, AFM,
ToF-SIMS, XPS)**
- At Chalmers:
 - **ESEM** Quanta FEG ESEM 200 med EDX-
analys, 20mm- 2nm
 - **TEM** Tecnai 200kV, structural level: 20mm-
2nm
 - **FIB-SEM**
 - **RAMAN spectroscope**
- At Lund University:
 - **X-ray Tomograph**

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Preparation equipment

- **Balzer freeze-etching unit,**
BAF 400T
- **Plunge freezing device**
- **Cryo-stage to the TEM**
- **Leica Cryostate, CM 1900**
- **Ultramicrotome, RMC**
PowerTome XL
- **Tissue processor for
plastic embedding,** Leica EM TP

Accessories

- Specially designed **microfluidic
flow cells** for microscopy
observation
- **Micro injection tool**
- **Heating/cooling cell** for CLSM
- **Microtensile test** for CLSM
- **Hybrid Detector** at CLSM SP5
- **Mass transport,** measuring of
local diffusion (FRAP, RICS)
- **Image Analysis** (Using both commercial
and in-house developed software)

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RISE at MAX IV and ESS – Applied research using x-rays and neutrons

- RISE will enable utilization of MAX IV and ESS by industry as a tool in material and product development
- RISE “pilots” guide industry, from their material challenge, through measurements to final material solution
- RISE will integrate x-ray and neutron techniques in their toolbox and use them in combination with our exciting instruments and methods to solve industrial and societal challenges
- RISE is currently expanding it’s capabilities by employing three specialists and an area coordinator
- Project leaders: **Niklas Lorén** and **Claes Holmqvist**

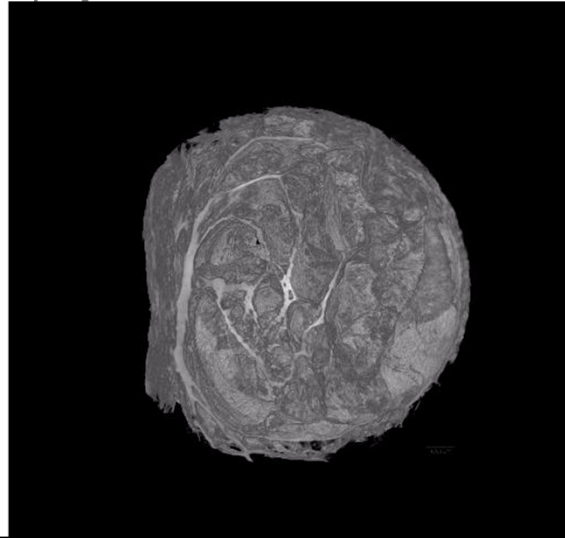
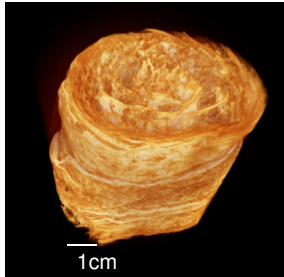
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Microstructure of a croissant

-Length scales in microscopy



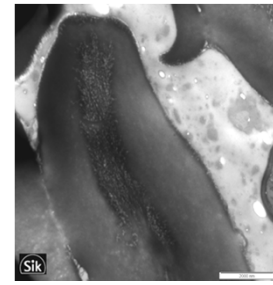
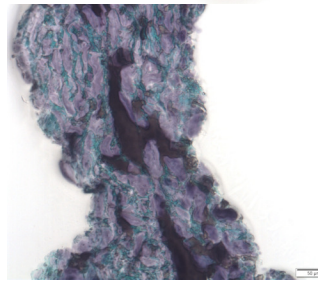
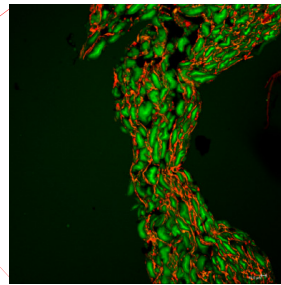
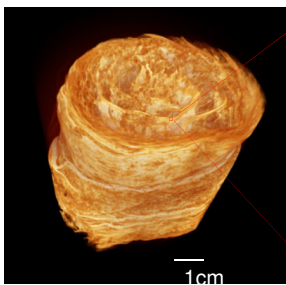
X-ray tomography of croissant



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Length scales in microscopy



3D-visualisation
using **x-ray**
tomography

Gluten protein
is red and starch
granules are
green in **CLSM**

In **LM** starch is shown in
violet (amylose dark blue
and amylopectin
pink/brown, protein in
light green and fat in
brown

A granule
surrounded by
the gluten in
TEM

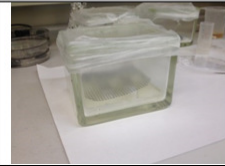
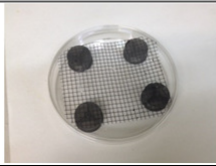
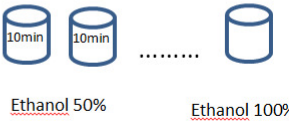

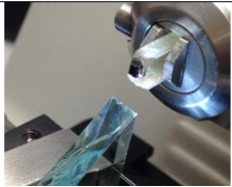
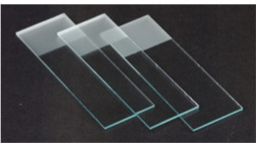
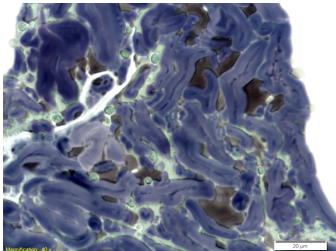
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Microstructure of bread using microscopy at different length scales (LM, TEM)

■ starch
■ amylose
■ amylopectin
■ protein
■ fat

RISE

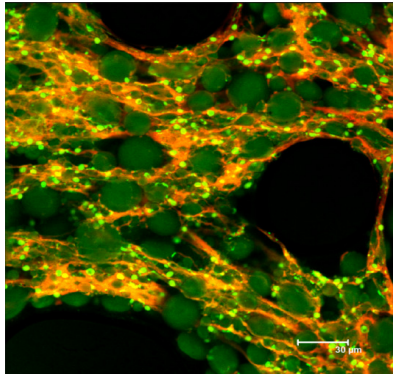
LM and TEM preparation

<p>1. Fixation (days) Formlaldehyde/glutar-aldehyde</p> 	<p>2. Fixation 2 (hours) Oso4</p> 	<p>3. Dehydration Graded ethanolseries</p> 	<p><i>For brittle materials, e.g. bread, snacks, that often containing a lot of cereals, careful fixation followed by plastic embedding and analyse in LM is the solution.</i></p> <p><i>LM is the outstanding method for distinguish between amylose and amylopectin</i></p>
<p>4. Embedding in plastic</p> 	<p>5. thin-sectioning</p> 	<p>6. Staining</p> 	 <p>RISE</p>

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Microscopy on dough

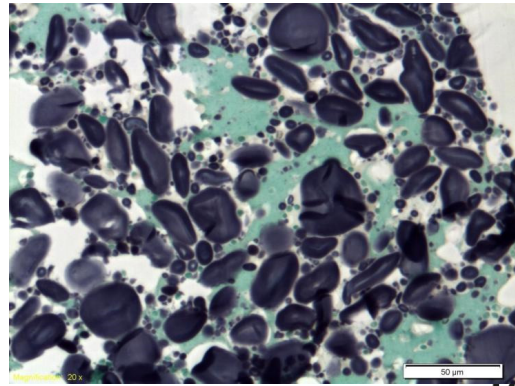
CLSM



gluten
starch granules
yeast cells

Yeast cells
starch granules
gluten

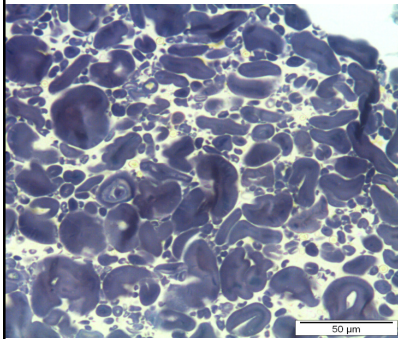
LM



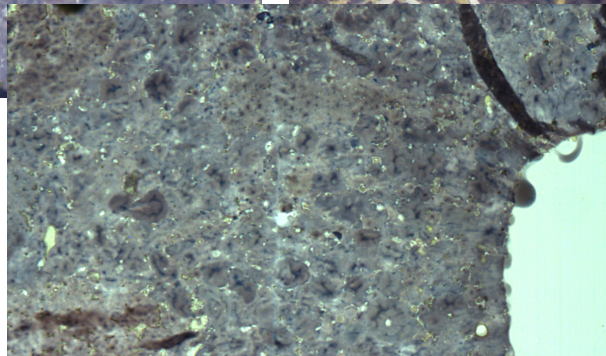
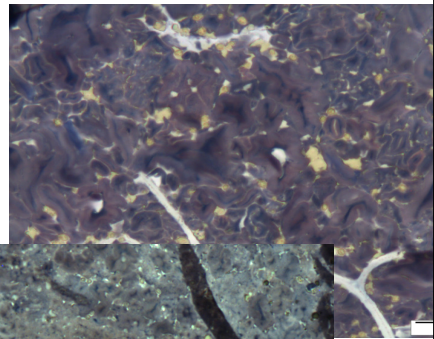
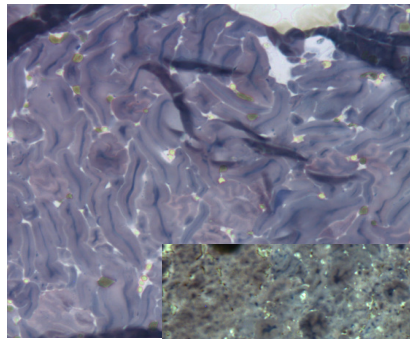
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Different types of bread

"normal" white wheat bread

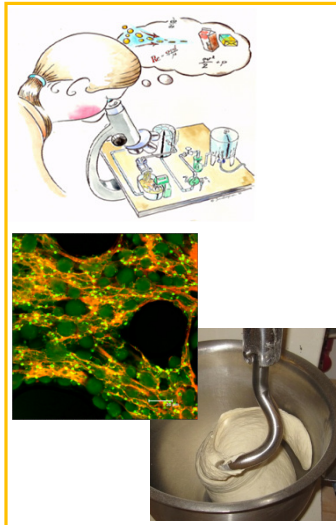


glutenfree



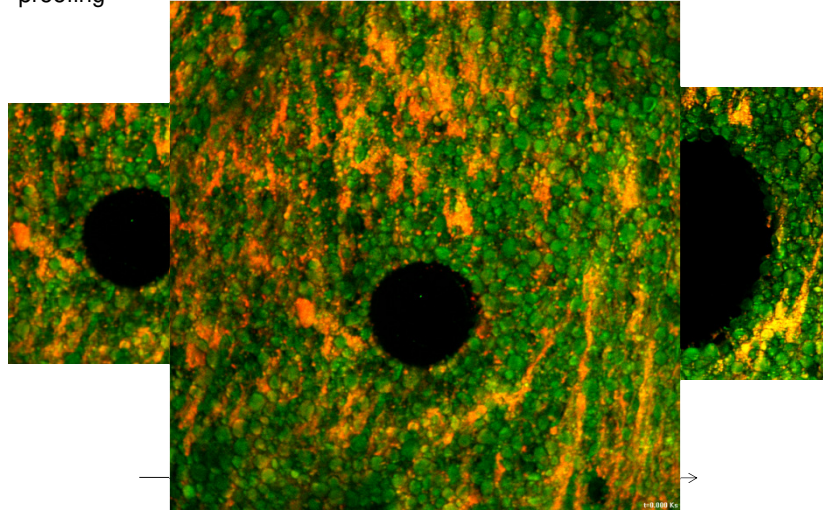
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Example of dynamic study -Dough -proofing



■ Yeast
■ Starch
■ protein

Developing of a bubble and deformation of material components during proofing



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FROZEN BREAD AND DOUGH

April 2011-Mars 2014

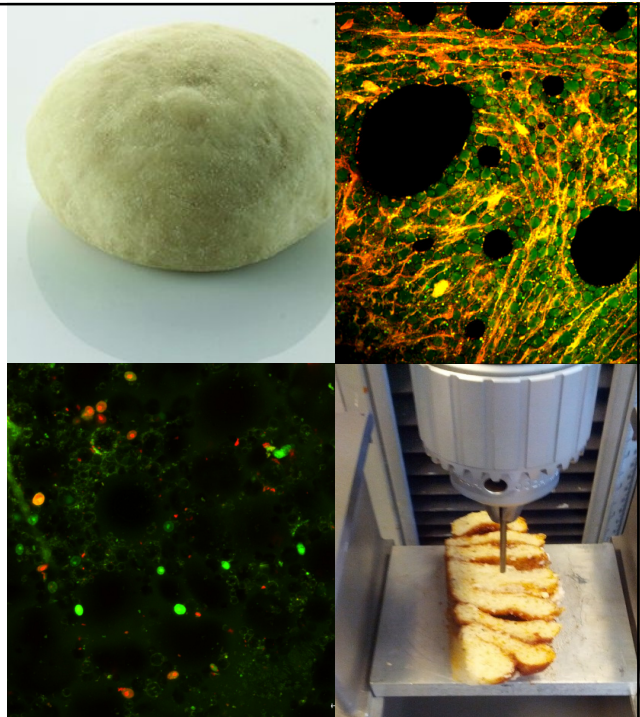
Camilla Öhgren, RISE

Maud Langton, SLU

Research Institutes of Sweden

Bioscience and Materials

AGRI-FOOD AND BIOSCIENCE

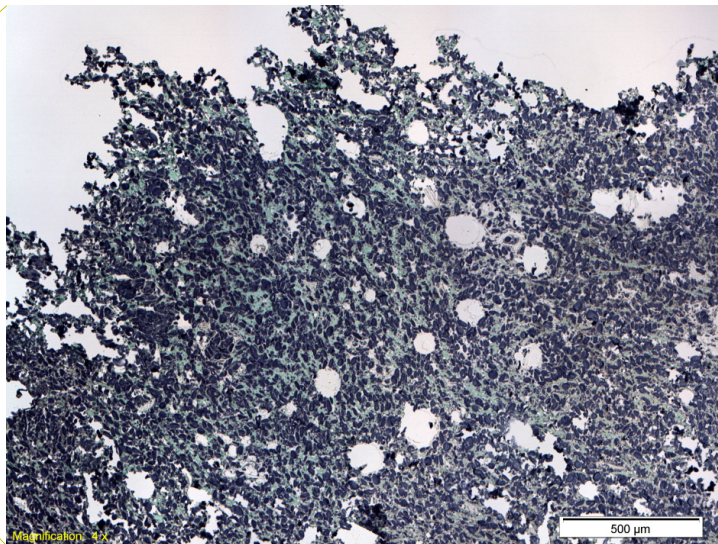
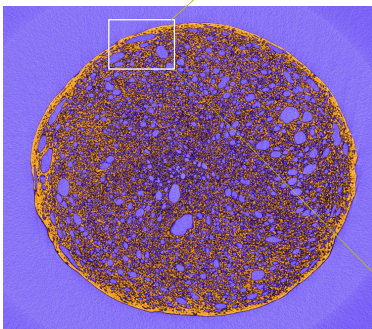
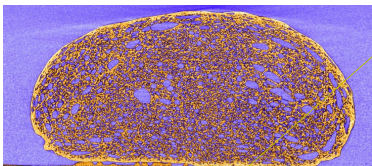


PARTICIPANTS:

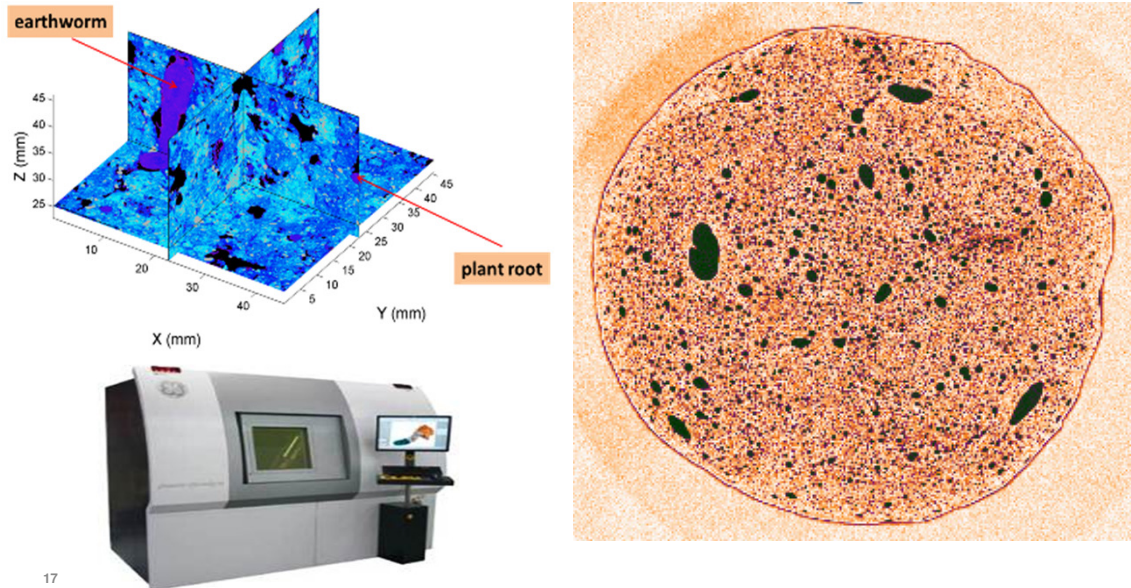
Dafgårds	(SIK) RISE	Swedish Board of Agriculture
Fazer	Chalmers	
Lantmännen	SLU	
Ewalco		
Norlander-Zeelandia		
Jästbolaget		
JBT		
Sveba-Dahlen		



Partly proofed dough –x-ray tomography at SLU, Uppsala



Partly proofed dough –x-ray tomography at SLU, Uppsala,



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BACKGROUND

Today, more and more bread is frozen at the bakery...

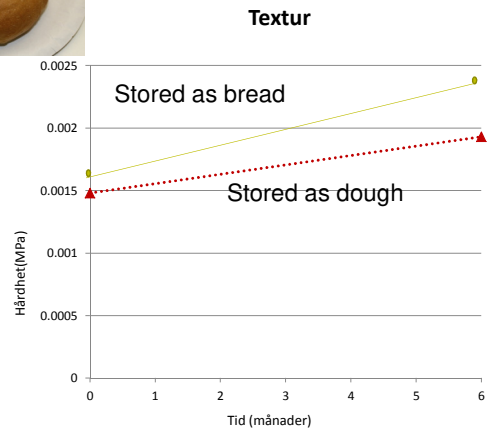
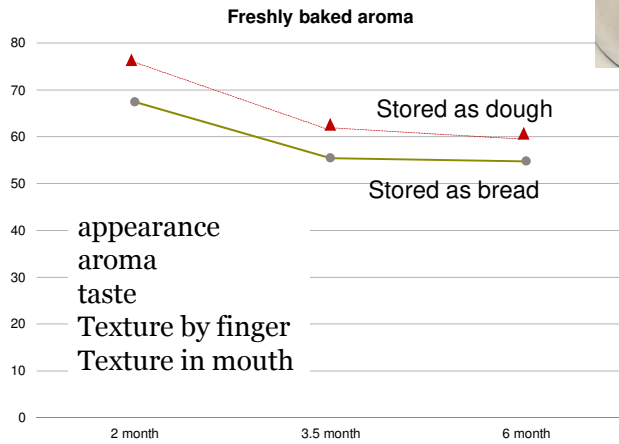
- What are the quality differences between freshly baked bread and the frozen alternatives, and what causes such differences?
- Do the consumers notice the differences?
- How can quality loss be avoided or minimized by changing the baking methods (freezing rate, kneading time) and the recipe (fiber, emulsifiers, enzymes etc)?



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Quality changes during storage -e.g. freshly baked aroma and texture

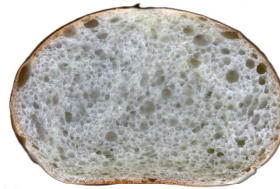


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What is the problem?..... volume decrease!!

Rye/wheat



Stored as dough in freezer



Stored as bread in freezer

Sweat wheat

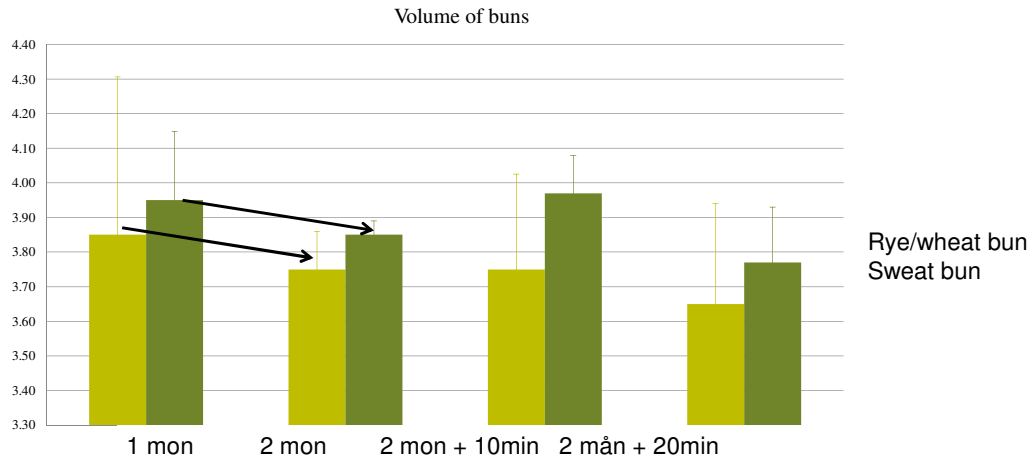


Stored as dough in freezer

Peaked buns!



Extra proofing...?

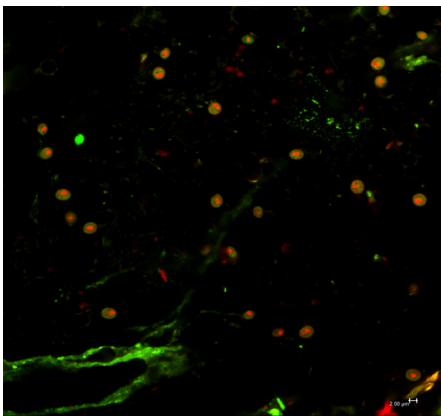


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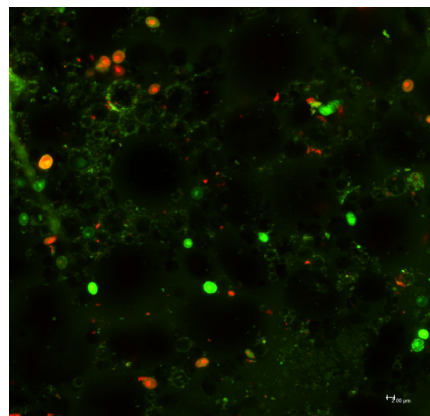
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How are the yeast?

Wheat/rye dough
92% dead



Sweat dough
46% dead



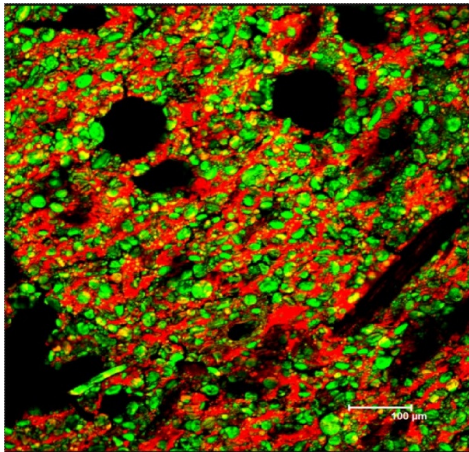
Stored unproofed for 10 month in the freezer

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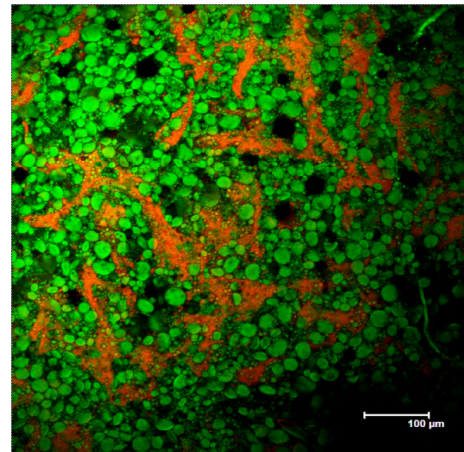
More reasons?

Wheat/rye dough



Gluten
starch

Sweet dough

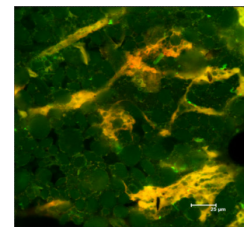
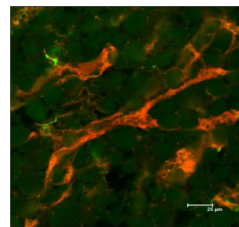
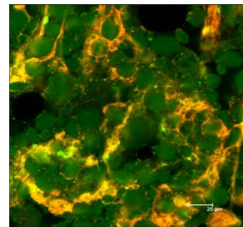
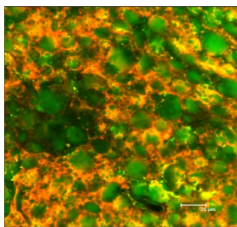


Sugar content?

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Effect of sugar content

Gluten
Starch granules

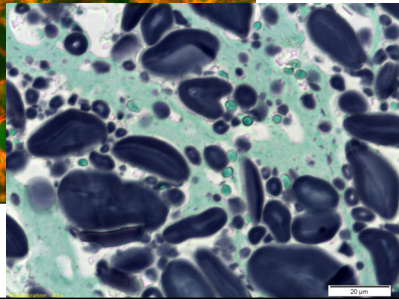
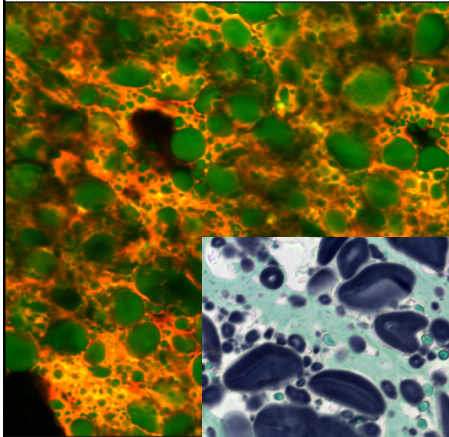


Sugar content

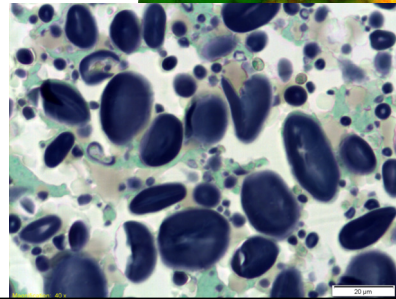
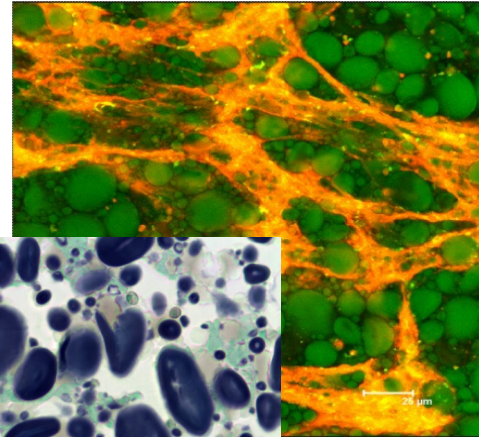
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Effect of freezing

Fresh



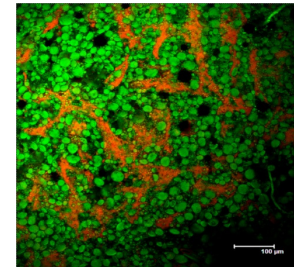
Frozen stored dough



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Why lower volume after frozen storage?

- Dead yeast
- sugar,
- Ice crystals – deterioration of the gluten network
- Ice crystals– dead yeast
- Loss of water
- Or combinations
- ...



Solutions:

- Increase kneading time
- Avoid storage >-15°C
- Avoid too long freeze storage
- Yeast stays alive longer in freezer with small amounts of sugar available, 0.4%

- Eckardt, J., Öhgren, C., Alp, A., Ekman, S., Åström, A., Chen, G., Swenson, J., Johansson, D. and Langton, M. (2013) Long-term frozen storage of wheat bread and dough – Effect of time, temperature and fibre on sensory quality, microstructure and state of water. *Journal of Cereal Science*, 57, 125-133.
- Chen, G., Öhgren, C., Langton, M., F Lustrup, K., Nydén, M. and Swenson, J. (2013) Impact of long-term frozen storage on the dynamics of water and ice in wheat bread. *Journal of Cereal Science*, 57, 120-124.
- Öhgren, C., Fabregat, N., and Langton, M. 2016. Quality of bread baked from frozen dough –effects of rye, and sugar content, kneading time and proofing profile, *LWT-Food science and technology*, 68, 626-633.

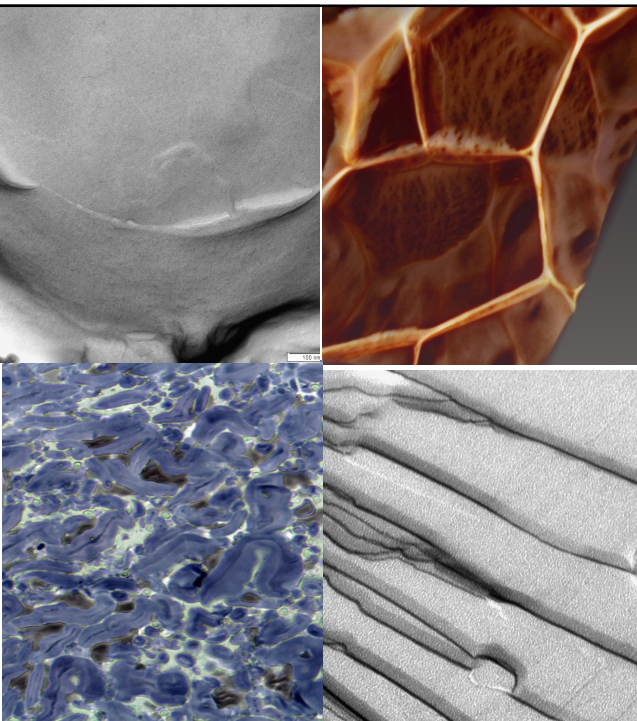
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THANK YOU!

Camilla Öhgren
Maj 2017

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Bioscience and Materials
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FAT DROPLET with emulsifier

BREAD

CARROT

CHOCOLATE

