

# The role of wheat endogenous lipids and their enzymatically released hydrolysis products in bread making

**Sara MELIS and Jan A. DELCOUR**

ICBC 2021 | 30 March 2021 | Online event

**KU LEUVEN**

Laboratory of Food Chemistry and Biochemistry (LFCB)  
Leuven Food Science and Nutrition Research Centre (LFoRCe)

# About me



KU LEUVEN

## Master of Bioscience Engineering: Food Technology

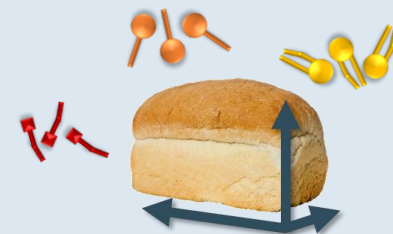
- Sep 2012 – Jun 2014
- The role of different egg yolk fractions during the production of pound cake



KU LEUVEN

## Doctoral researcher (Lab of Food Chemistry & Biochemistry)

- Sep 2014 – Jun 2019
- The role of wheat endogenous lipids and their enzymatically released hydrolysis products in bread making



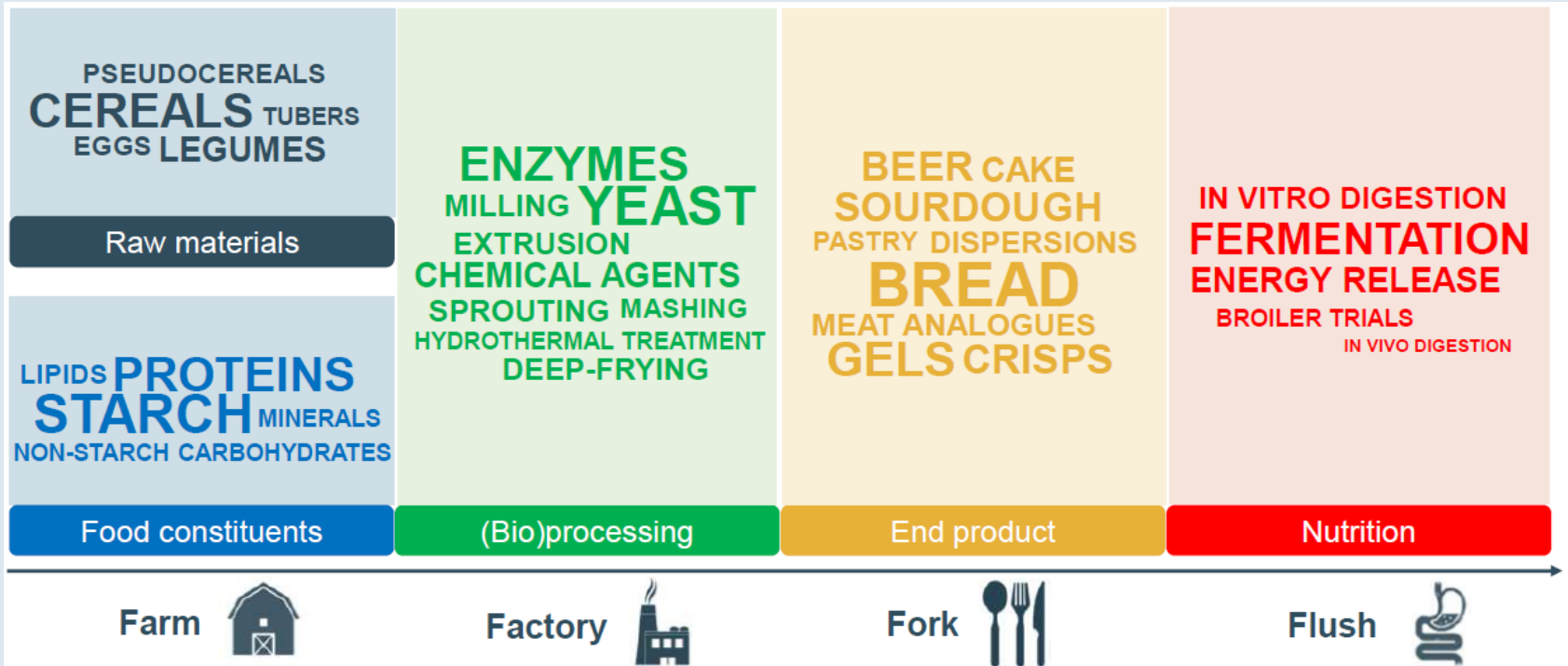
KU LEUVEN

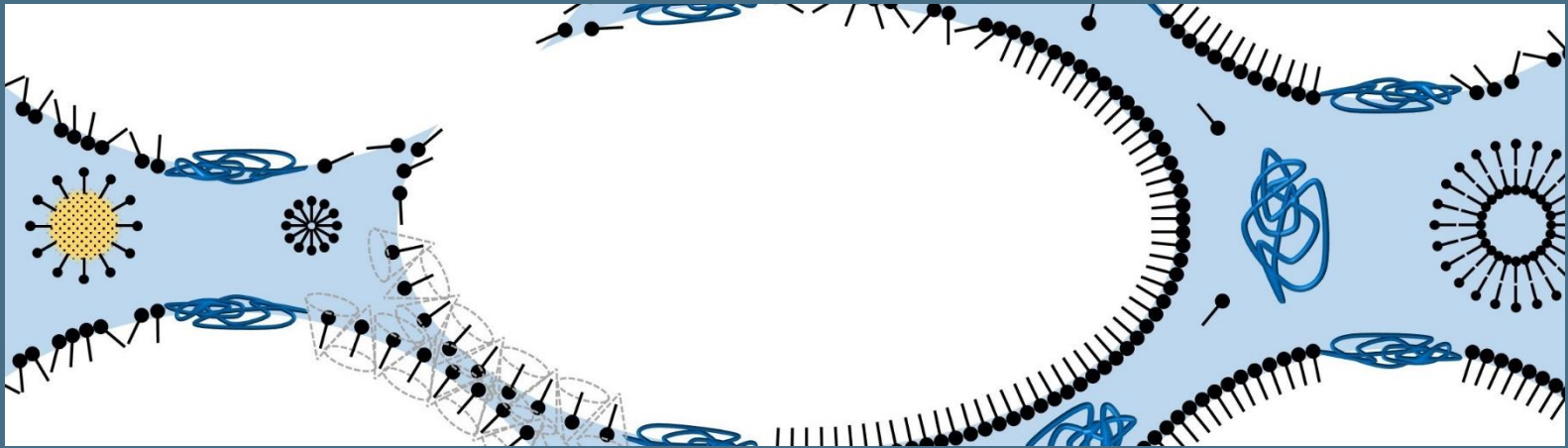
## Postdoctoral researcher (Lab of Food Chemistry & Biochemistry)

- Jun 2019 – Aug 2019: Knowledge driven product development
- Sep 2019 – ...: Coordination FIBRAXFUN project
  - “A knowledge base for exploiting novel wheats rich in arabinoxylan dietary fibre throughout the wheat value chain”



# Lab of Food Chemistry & Biochemistry





# The role of wheat endogenous lipids and their enzymatically released hydrolysis products in bread making

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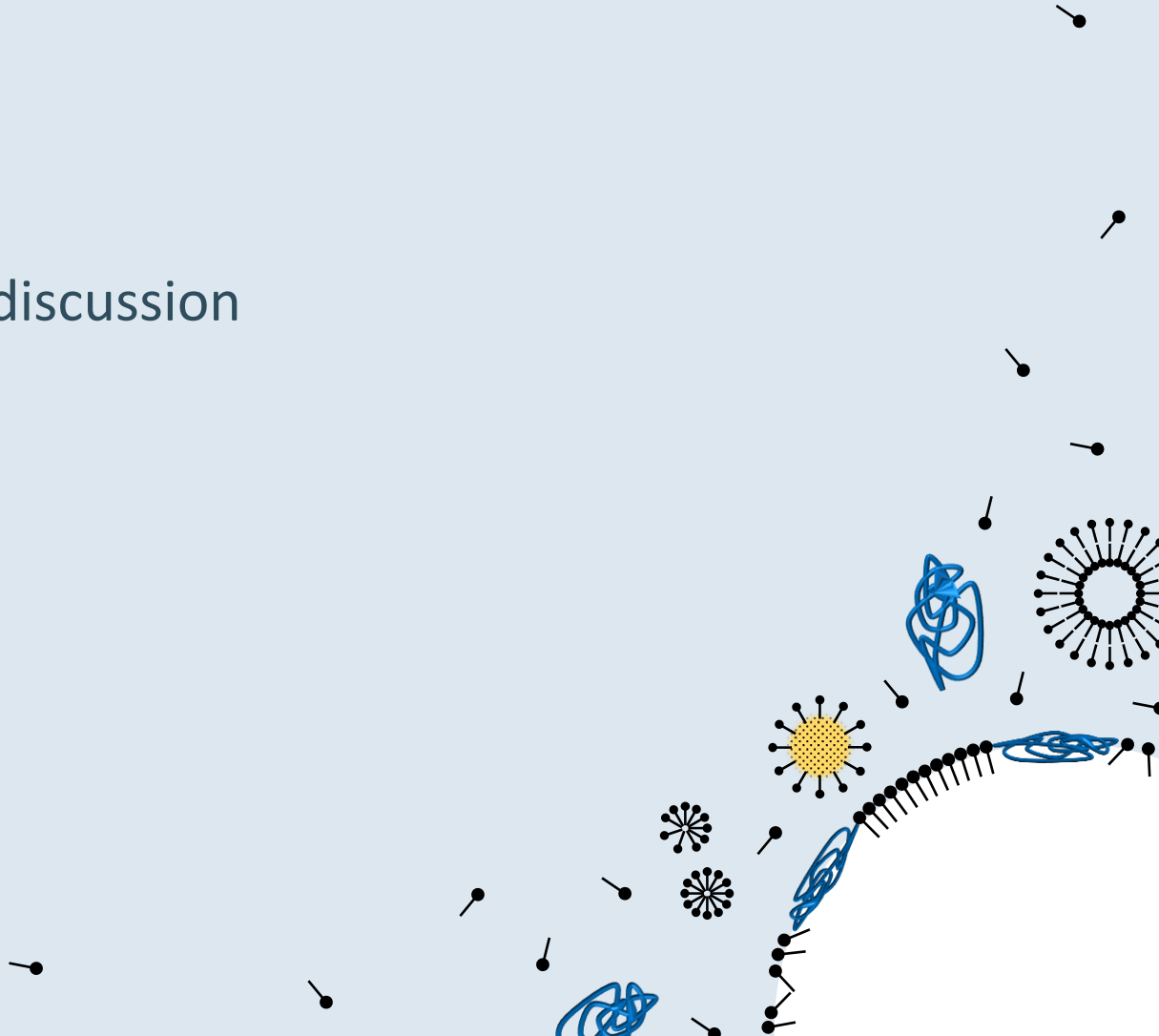
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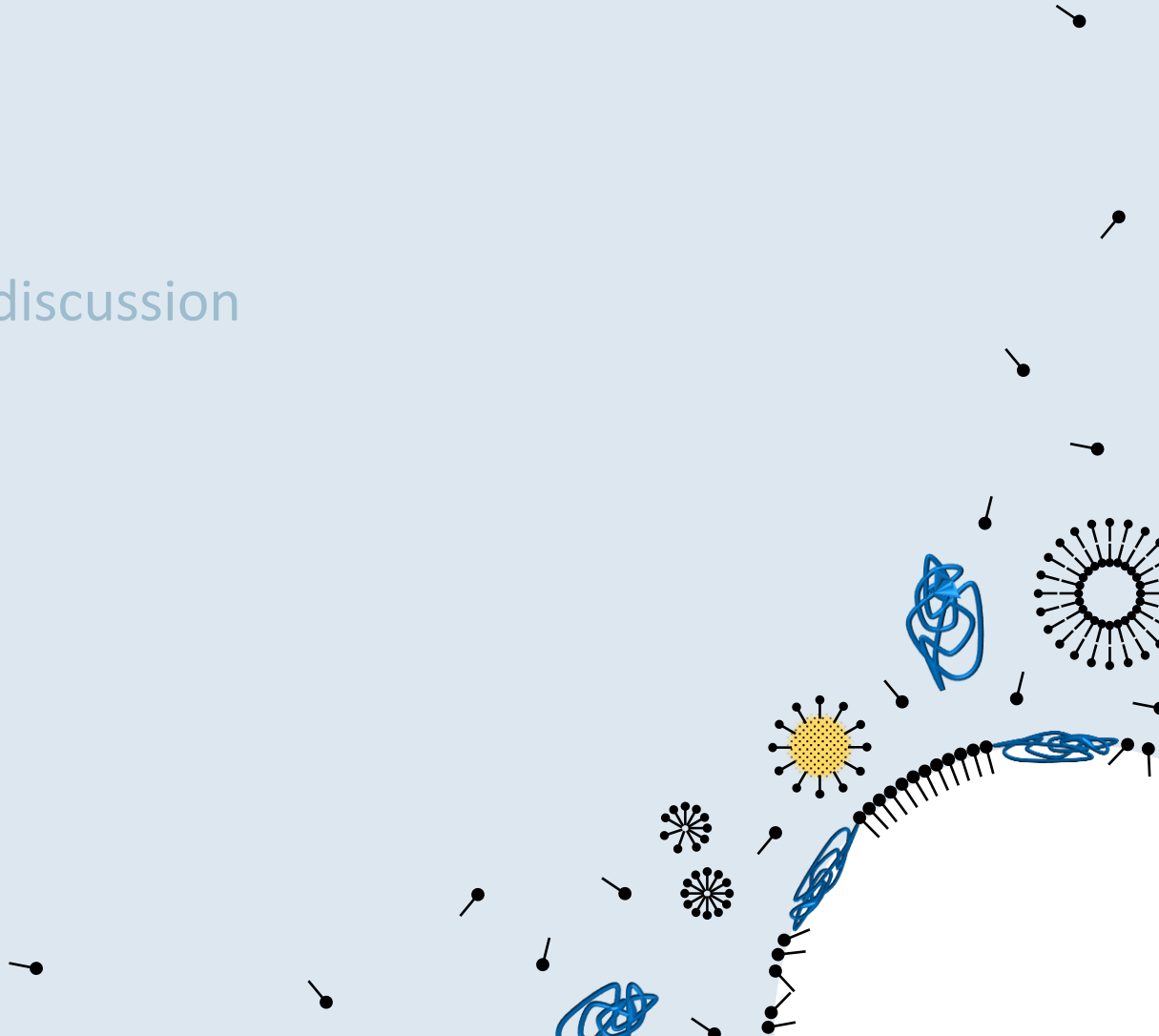
# Overview

- Introduction
- Objective
- Results and discussion
- Conclusions



# Overview

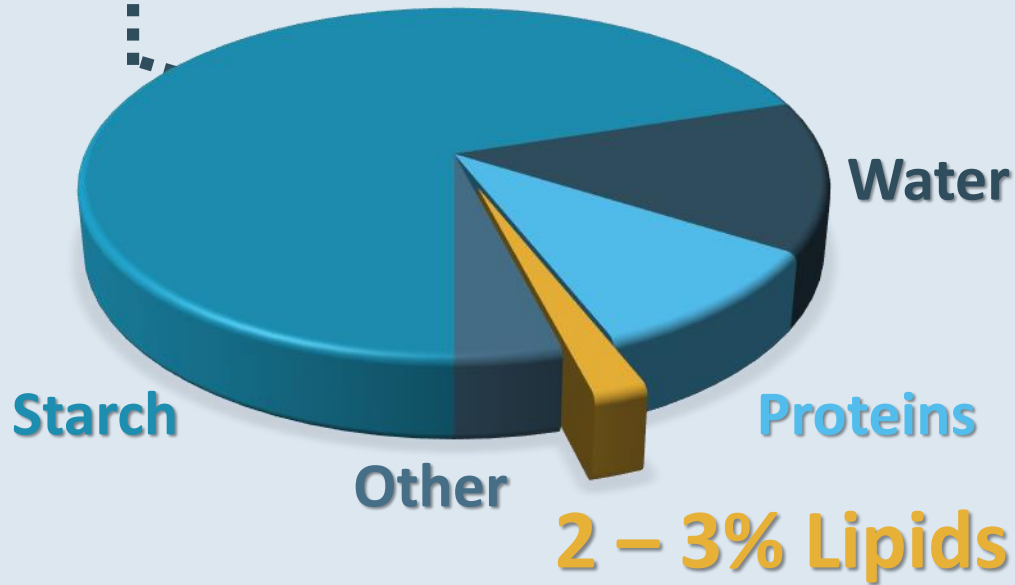
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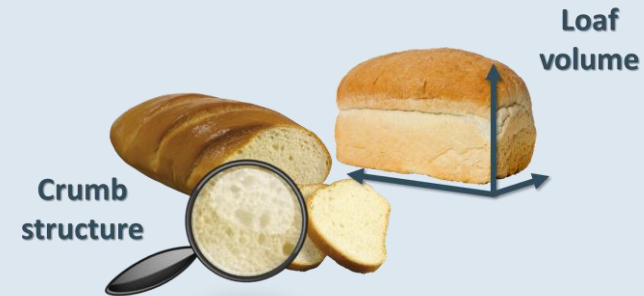


# Introduction

## BREAD



**Important influence  
on bread quality!**



# Introduction

## LIPIDS

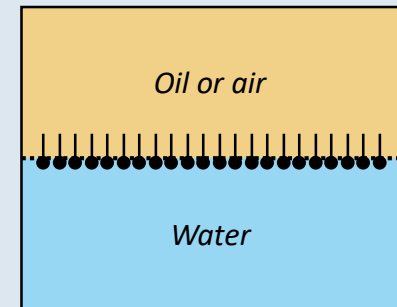
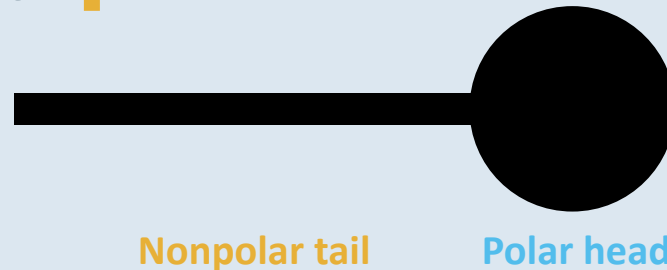
*Lipids are fatty acids and their derivatives, and substances related biosynthetically or functionally to these compounds*

■ Acylglycerols + **Free fatty acids (FFAs)** — **Nonpolar lipids**

■ Galactolipids

— **Polar lipids**

■ Phospholipids





# Introduction

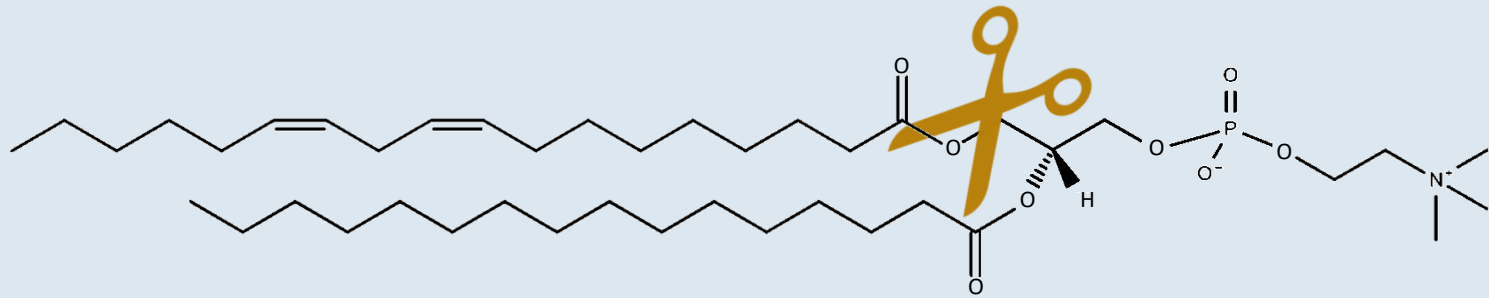
## *LIPIDS IN BREAD MAKING: IMPACT ON BREAD QUALITY*

- Investigated in different ways
  - Flours varying in bread making quality
  - Fractionation-reconstitution experiments
- Lipase technology

# Introduction

## LIPASES

*Lipases are lipid degrading enzymes catalyzing the hydrolysis of (phosphodi)ester bonds of glycerol(phospho)lipids*

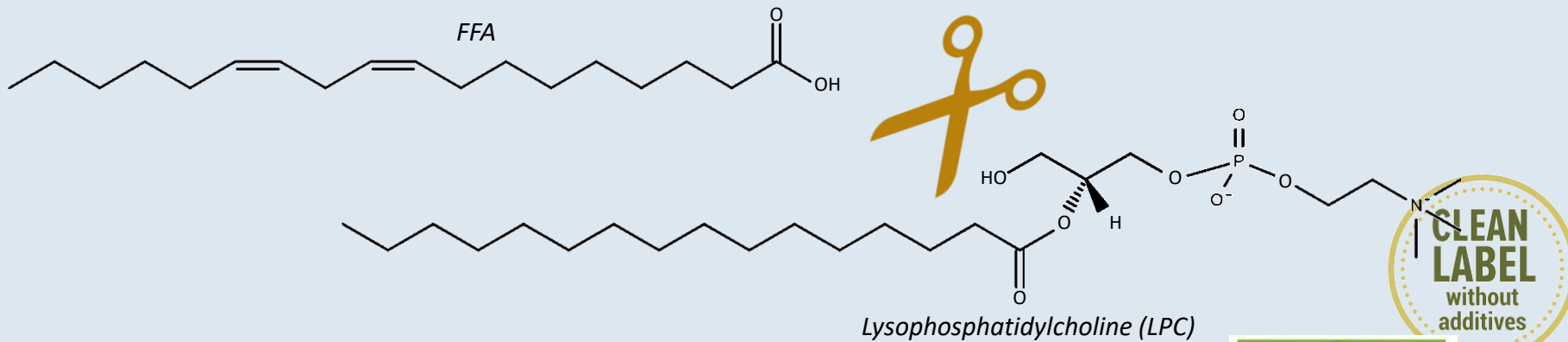


*Phosphatidylcholine (PC)*

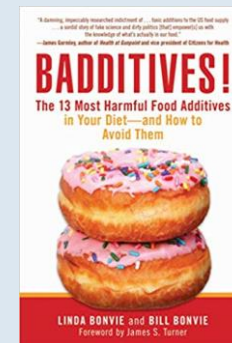
# Introduction

## LIPASES

*Lipases are lipid degrading enzymes catalyzing the hydrolysis of (phosphodi)ester bonds of glycero(phospho)lipids*



- Excellent research tools
- Potential (clean label) alternatives for certain additives in bread making



# Introduction

## *LIPIDS IN BREAD MAKING: IMPACT ON BREAD QUALITY*

- Investigated in different ways
  - Flours varying in bread making quality
  - Fractionation-reconstitution experiments
- Lipase technology
  - Delivered new insights
  - Limited/no information
    - Role of enzymatically released hydrolysis products
    - Interplay between wheat endogenous lipids and their enzymatically released hydrolysis products



# Introduction

## *MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY*

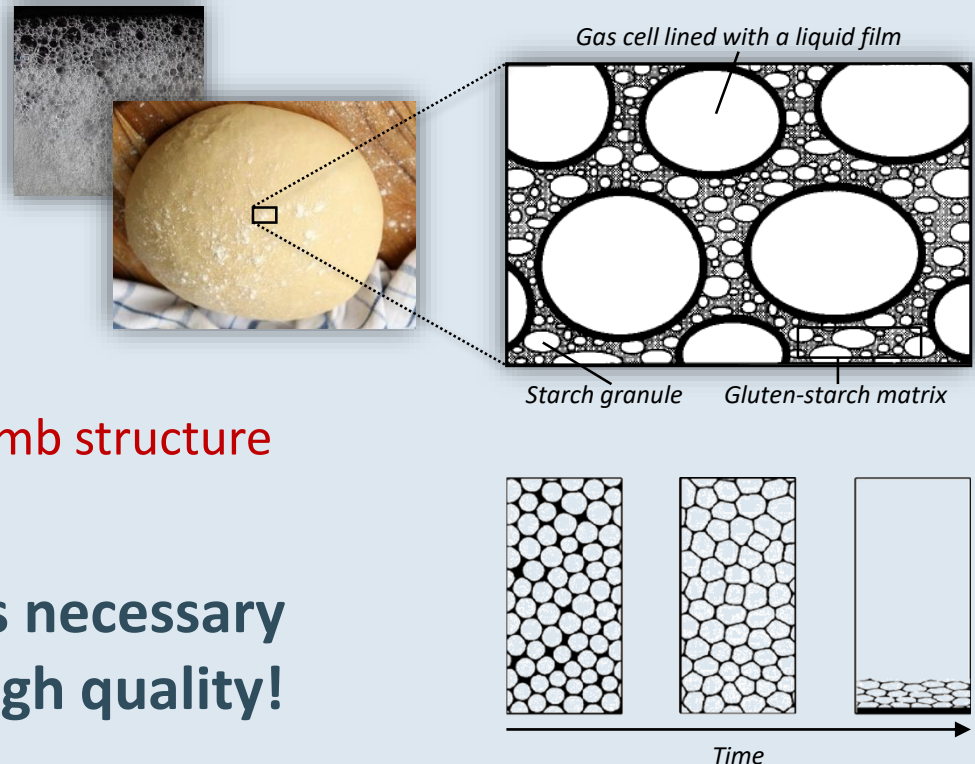
- Impact of lipids on loaf volume and crumb structure is related to the stability of gas cells in bread dough

- Bread dough = foam
  - Unstable
  - Gas is lost over time

👉 **Low loaf volume**

👉 **Coarse, irregular crumb structure**

- **Stabilization of gas is necessary to obtain bread of high quality!**



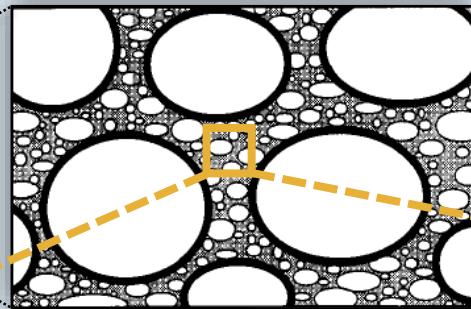
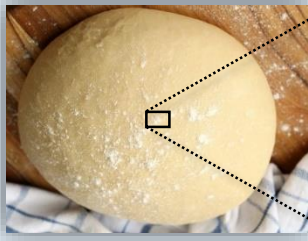
# Introduction

## MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY

- Impact of lipids on gas cell stability in bread dough

- Indirect** → Strength of gluten-starch matrix

- Direct**

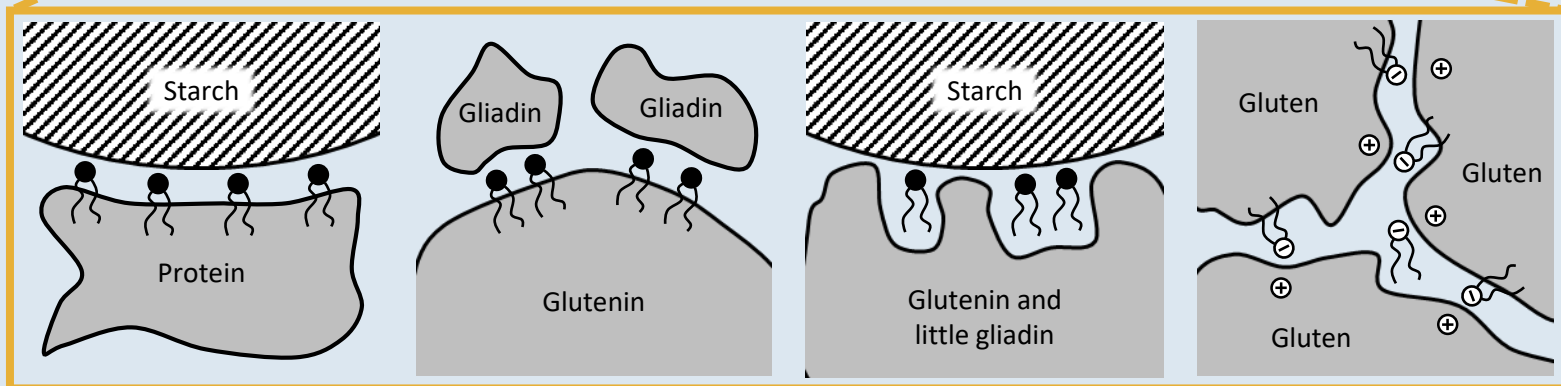


Early stages of fermentation

○ Starch granules

○ Gas cells lined with a liquid film

■ Gluten-starch matrix

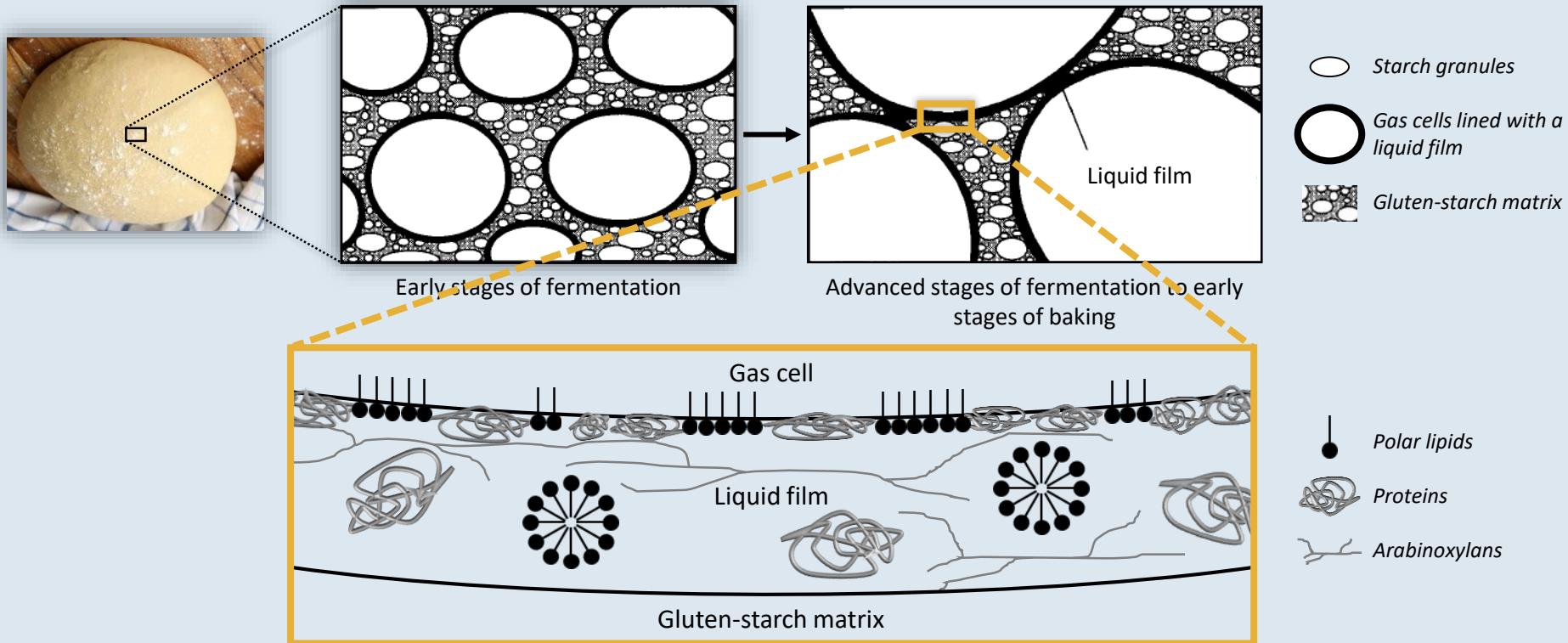




# Introduction

## MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY

- Impact of lipids on gas cell stability in bread dough
  - Indirect → Strength of gluten-starch matrix
  - **Direct** → Presence in liquid films

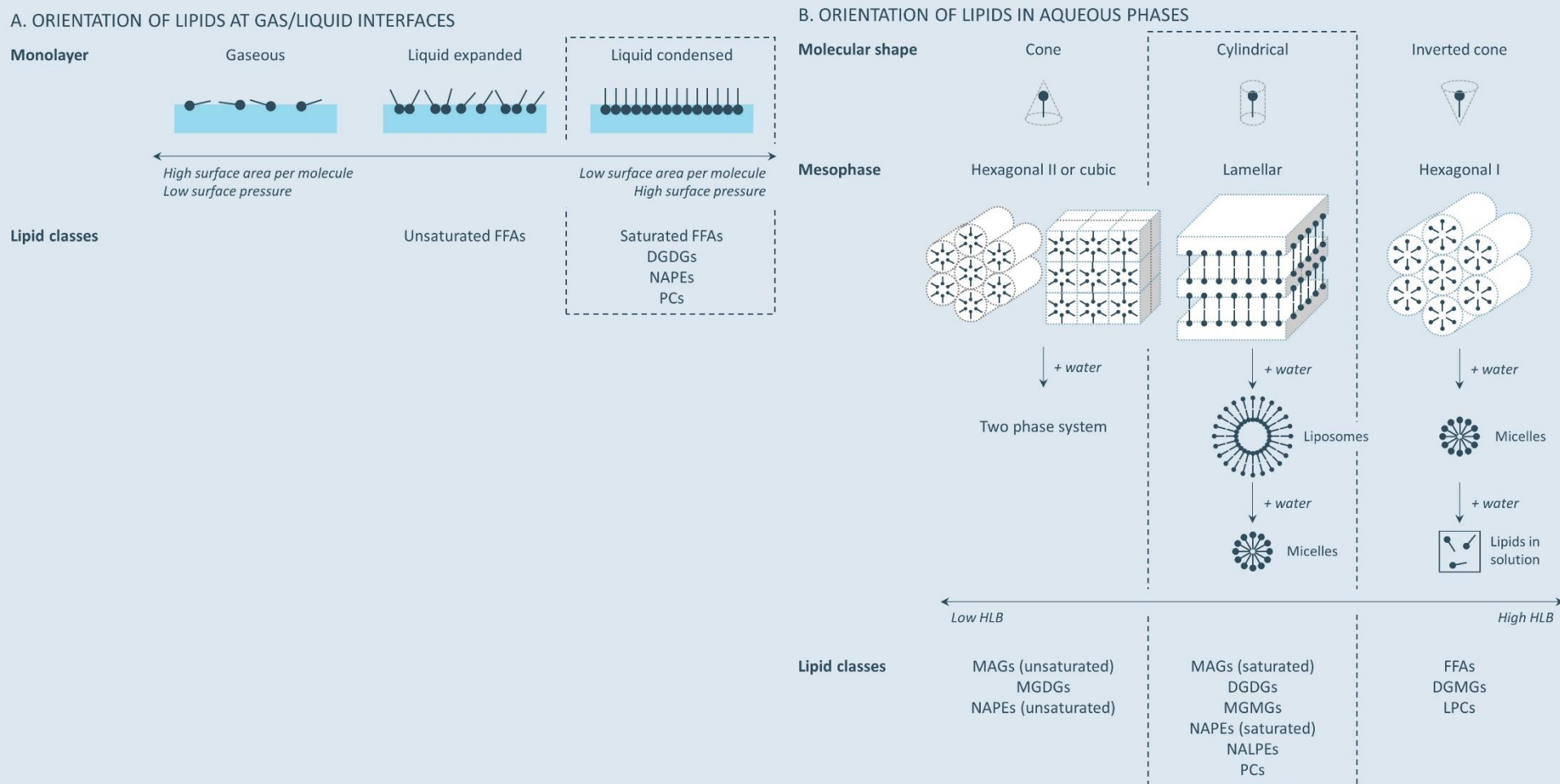


(Gan et al., 1990; Gan et al., 1995; Mills et al., 2003)

# Introduction

## MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY

- Ability of lipids to directly stabilize gas cells depends on



(Eliasson and Larsson, 1993; Gunstone et al., 2007; Kaganer et al., 1999; Krog, 1981; MacRitchie, 1976b)

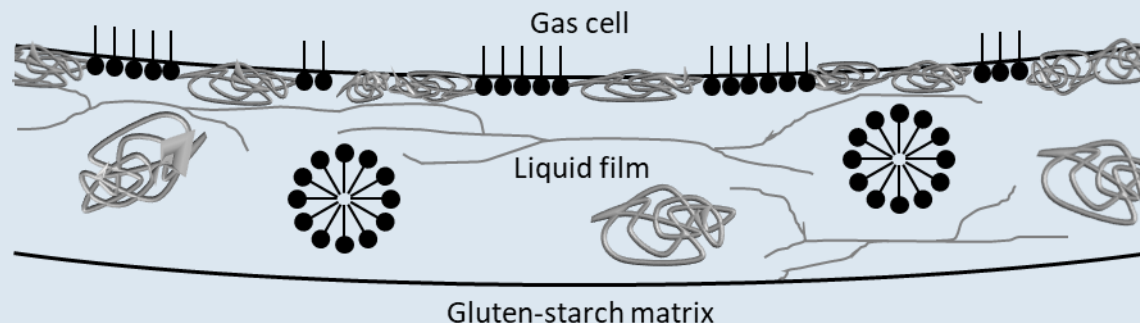
# Introduction

## *MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY*

Leading hypothesis:

Impact of wheat endogenous lipids and their enzymatically released hydrolysis products is exclusively related to their **direct** action as surface-active components

- Flour lipids and lipases → no effect on dough bulk rheology  
(Gerits et al., 2015b; MacRitchie and Gras, 1973; Sloan et al., 2009; Sloan and MacRitchie, 2009)
- Effects of lipids on loaf volume and crumb structure  
~ theoretical ability of lipids to (de)stabilize gas/liquid interfaces  
(MacRitchie, 1976a; MacRitchie, 1977; MacRitchie, 1981; MacRitchie and Gras, 1973)



# Introduction

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- Effects of lipids on loaf volume and crumb structure  
~ theoretical ability of lipids to (de)stabilize gas/liquid interfaces  
(MacRitchie, 1976a; MacRitchie, 1977; MacRitchie, 1981; MacRitchie and Gras, 1973)
- Presence and action of lipids at gas cell interfaces in dough never experimentally proven!



# Introduction

## CONCLUSIONS

- Chung and coworkers (1978)
  - Reviewed the role of wheat flour lipids in bread making
  - Much information available
    - Fragmentary
    - Sometimes contradictory
  - Factors making sustained and consistent progress difficult
  - Little likelihood that a unified theory on the role of wheat lipids in the production of baked goods could be developed



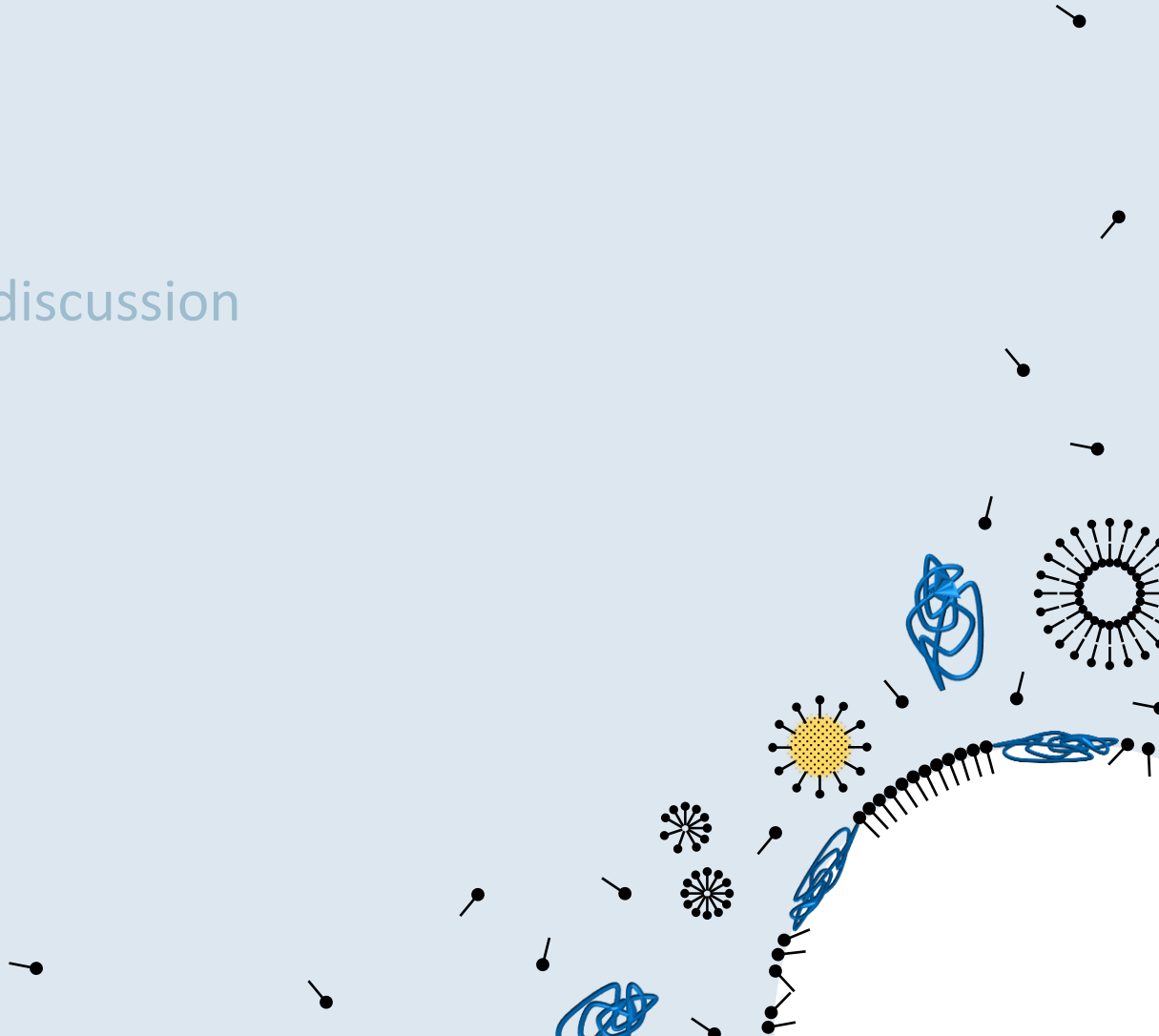
~

*“Consequently, we will learn increasingly more about the role of lipids, but we will be frustrated by the complexity of the effects.”*

~

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# Objective

To unravel the role of wheat endogenous lipids and their enzymatically released hydrolysis products in bread making



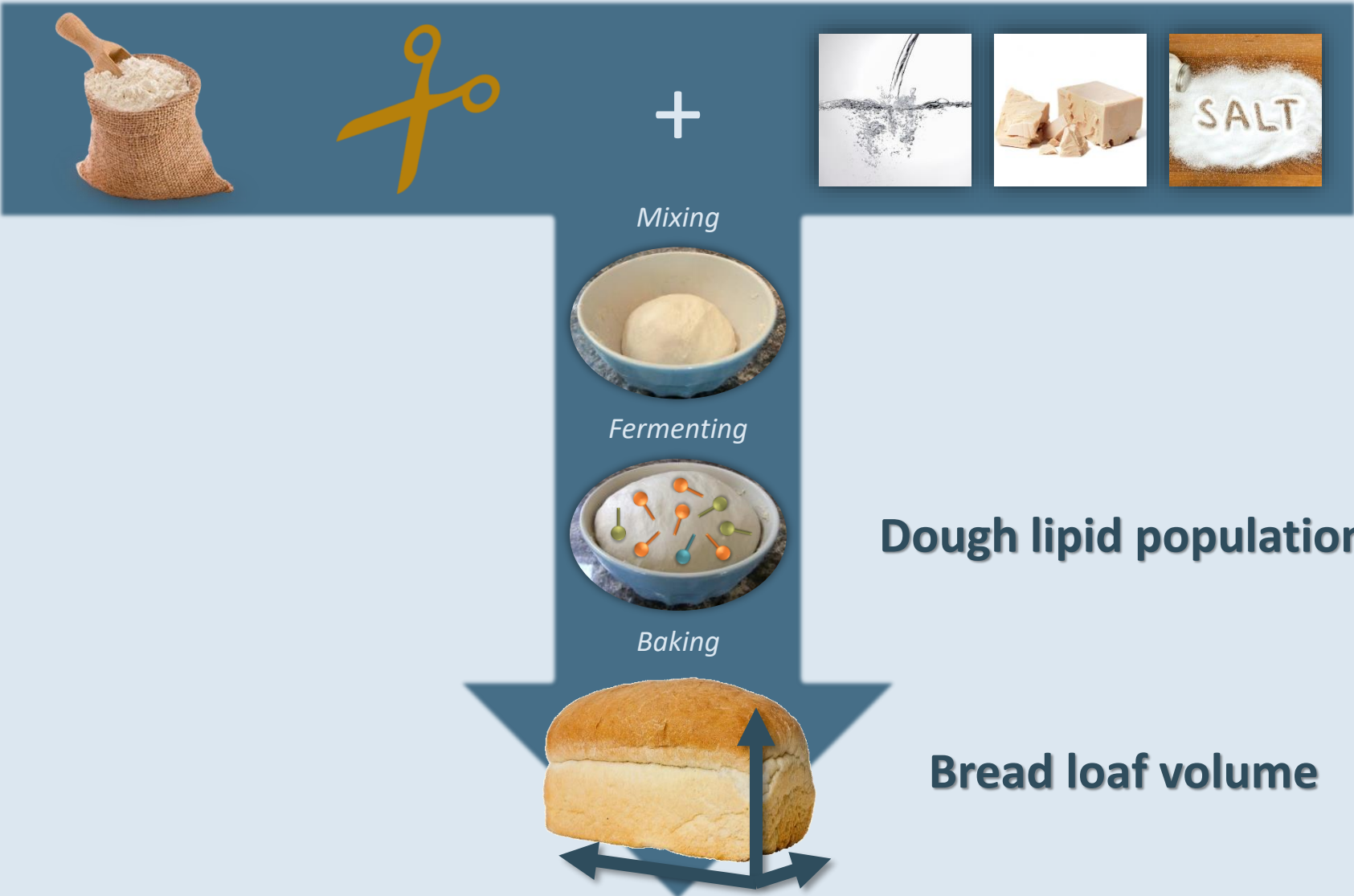
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  - Impact of lipids on bread quality
  - Mechanisms whereby lipids impact bread quality
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# Results and discussion

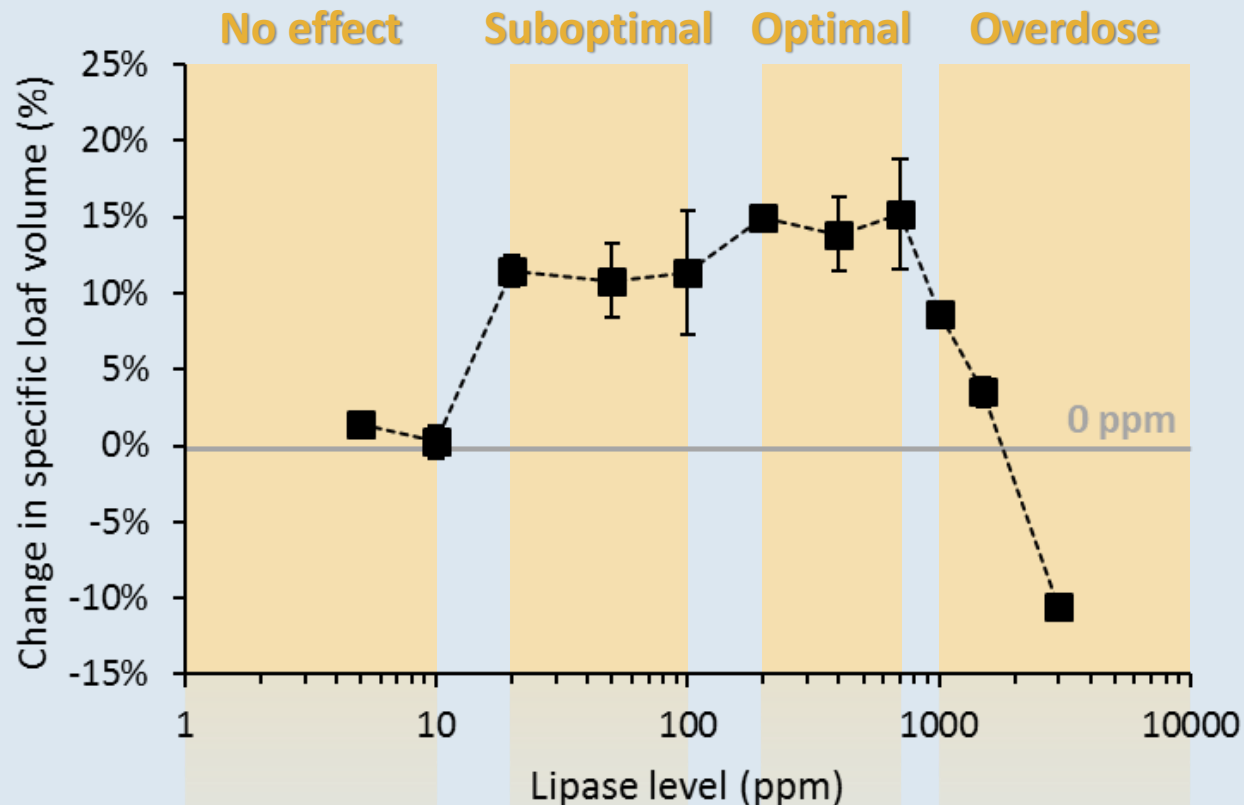
## IMPACT OF LIPIDS ON BREAD QUALITY



# Results and discussion

## IMPACT OF LIPIDS ON BREAD QUALITY

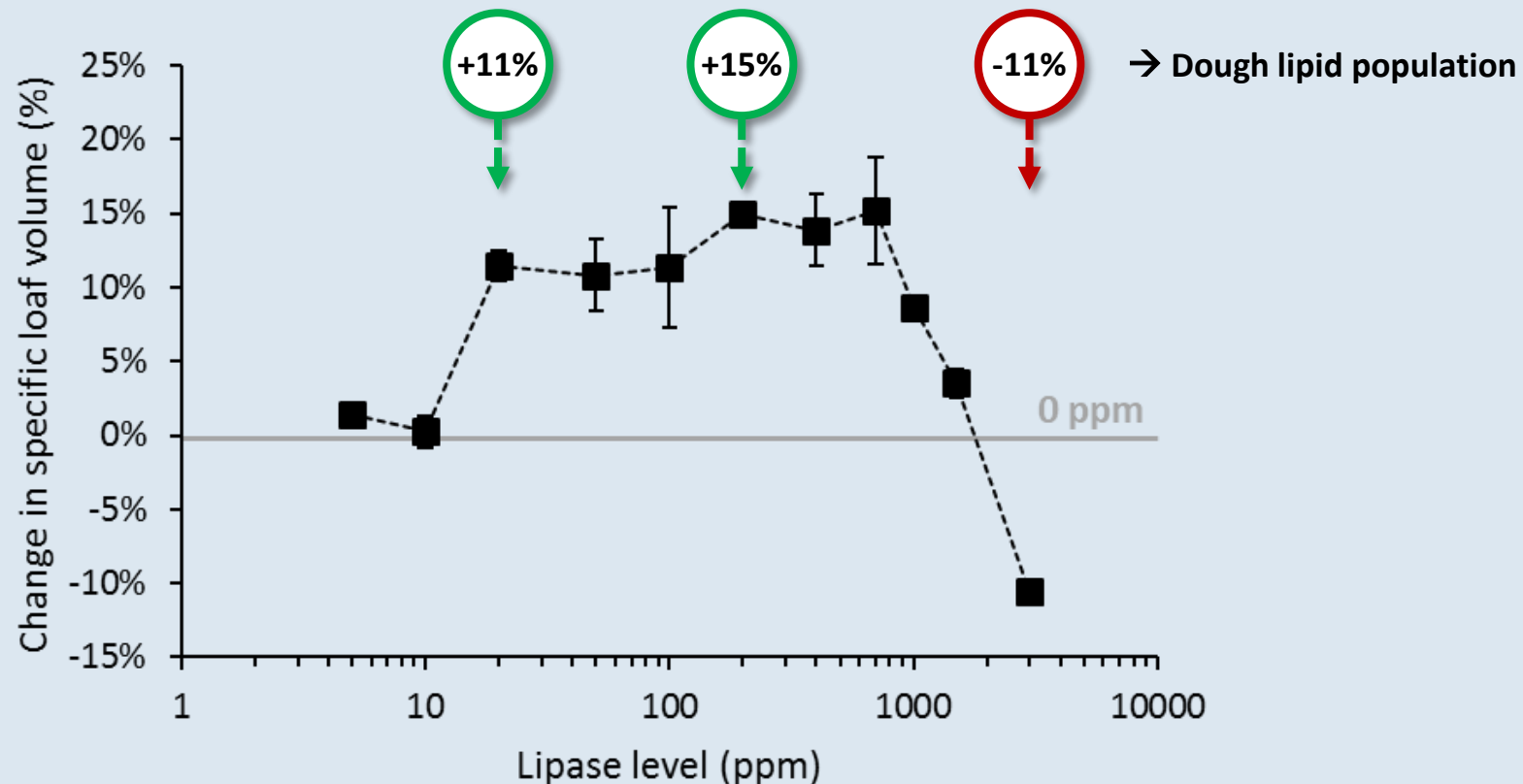
- Change in specific loaf volume as a function of lipase level



# Results and discussion

## IMPACT OF LIPIDS ON BREAD QUALITY

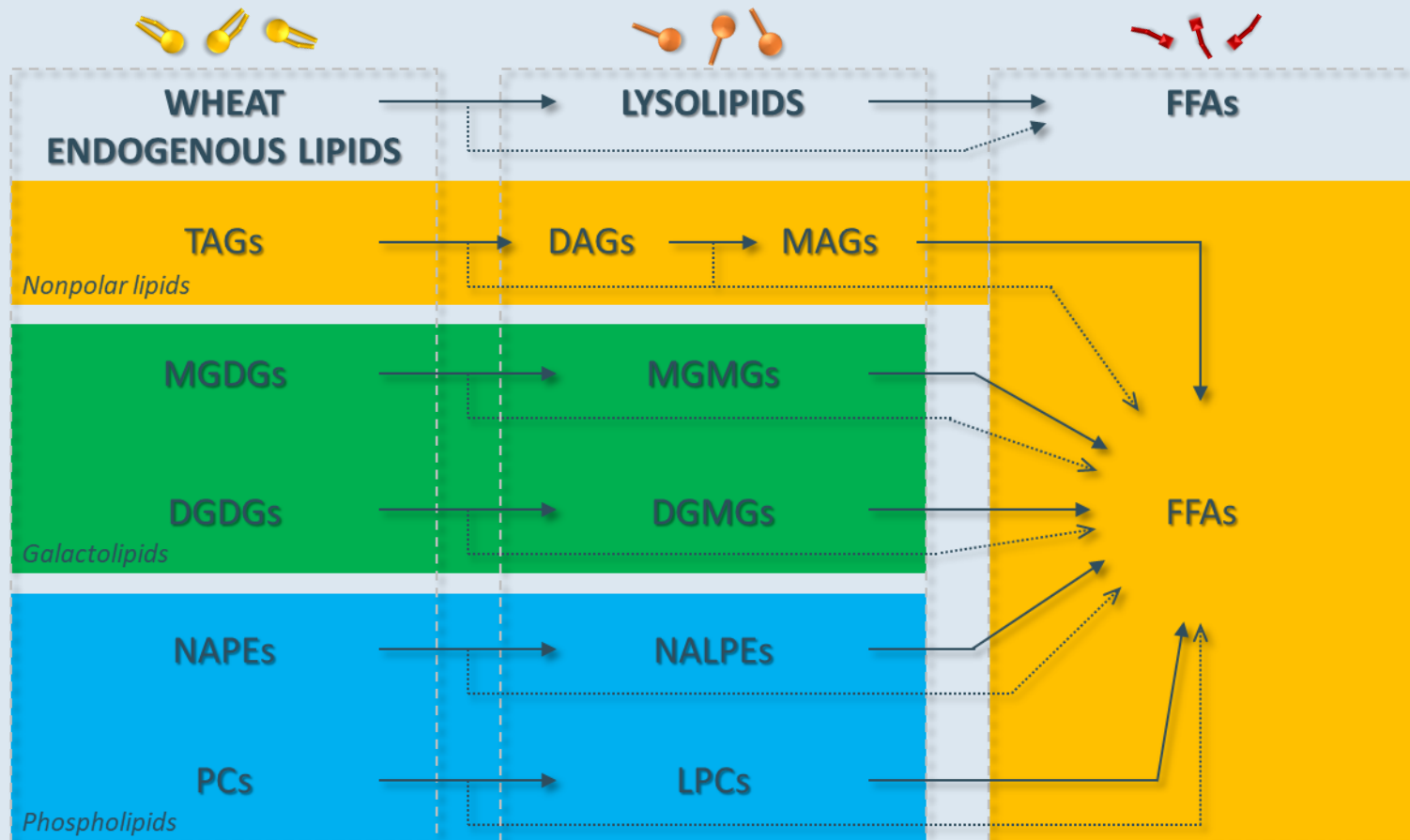
- Change in specific loaf volume as a function of lipase level



# Results and discussion

## IMPACT OF LIPIDS ON BREAD QUALITY

- Dough lipid population analysis (HPLC-ELSD)

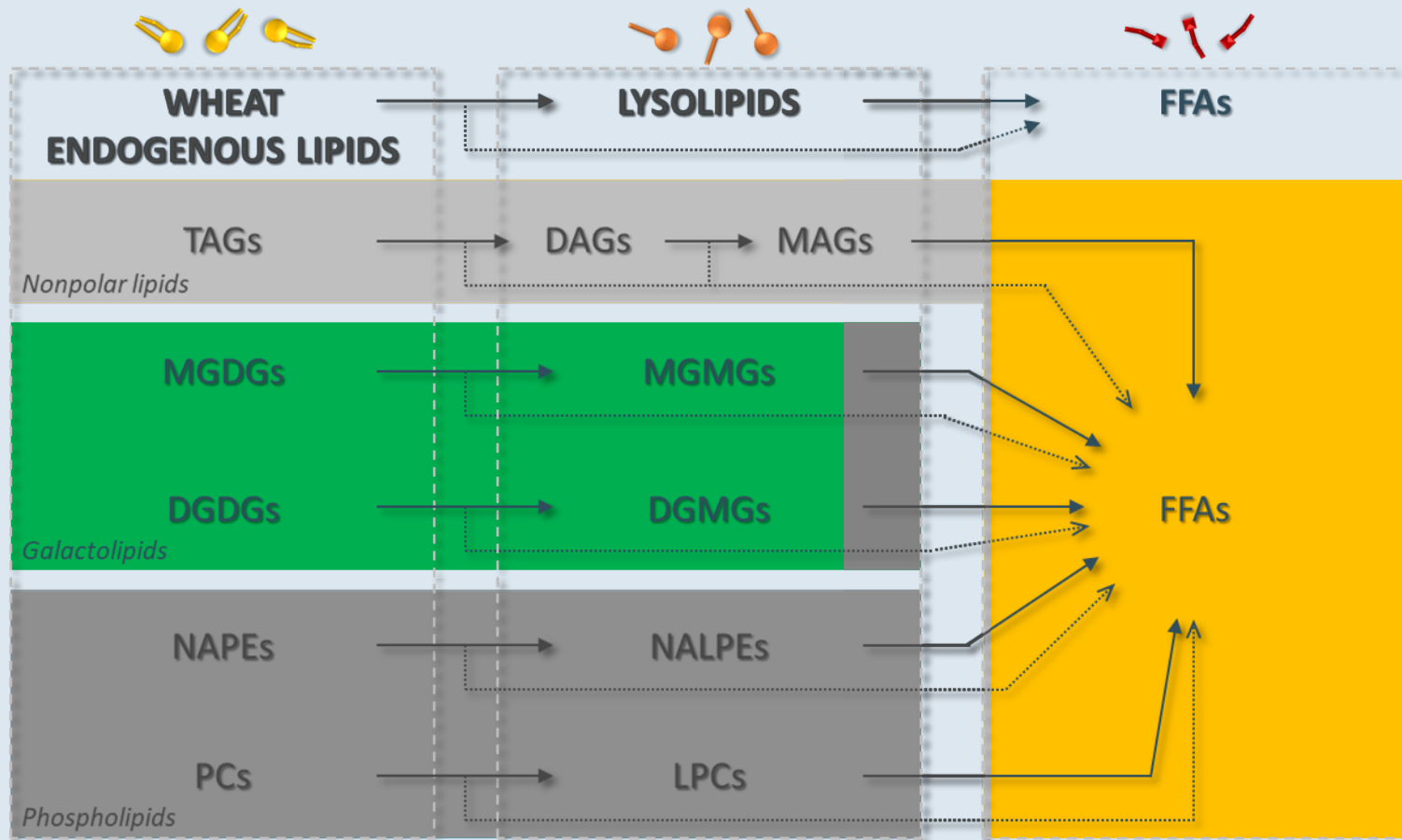




# Results and discussion

## IMPACT OF LIPIDS ON BREAD QUALITY

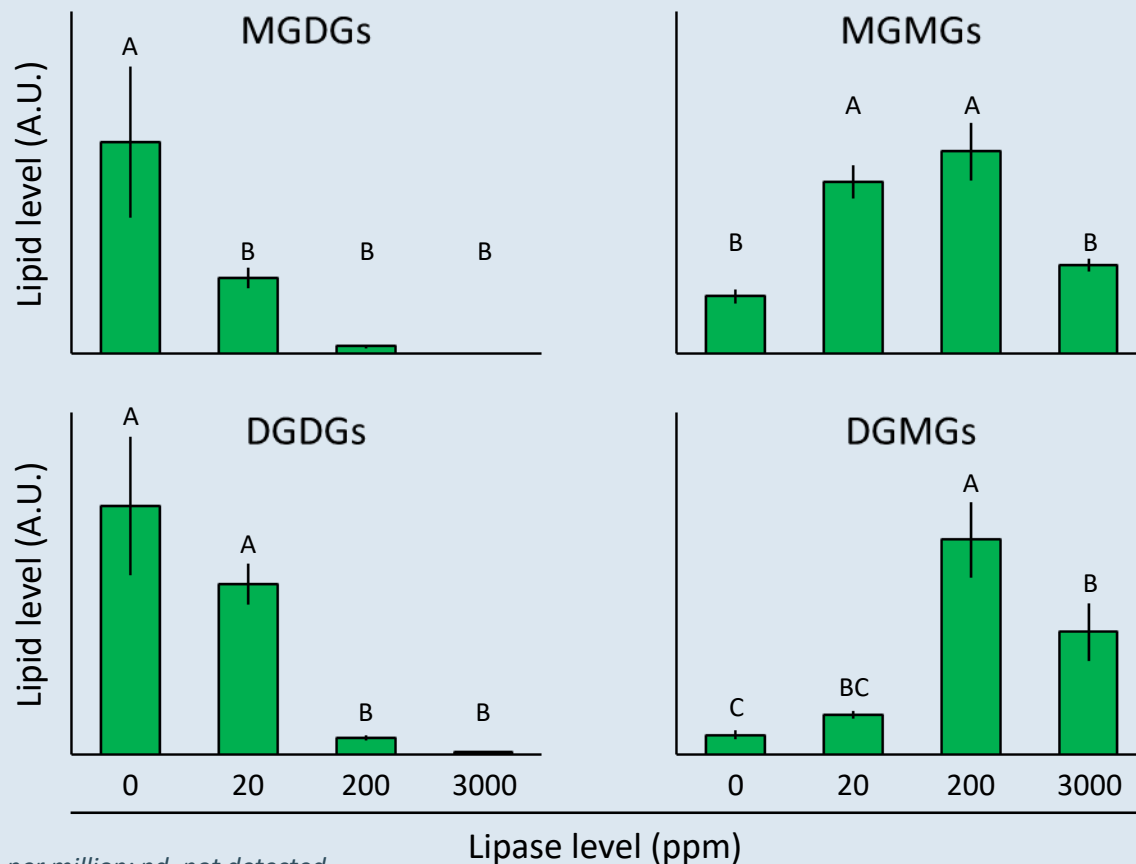
- Dough lipid population analysis (HPLC-ELSD)



# Results and discussion

## IMPACT OF LIPIDS ON BREAD QUALITY

- Lipid levels in dough prepared with and without different levels of lipase



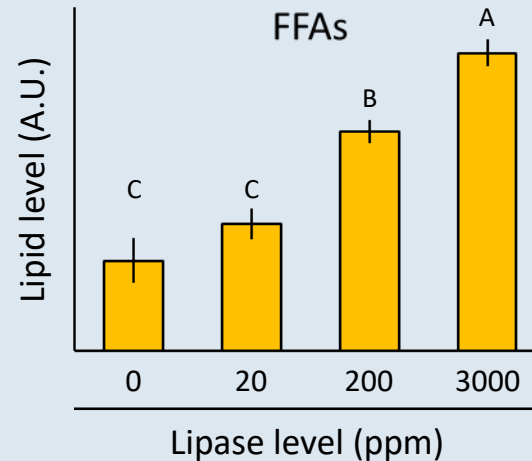
A.U., arbitrary units; ppm, parts per million; nd, not detected.

Bars with different capital letters are significantly different from each other ( $p < 0.05$ ).

# Results and discussion

## IMPACT OF LIPIDS ON BREAD QUALITY

- Lipid levels in dough prepared with and without different levels of lipase

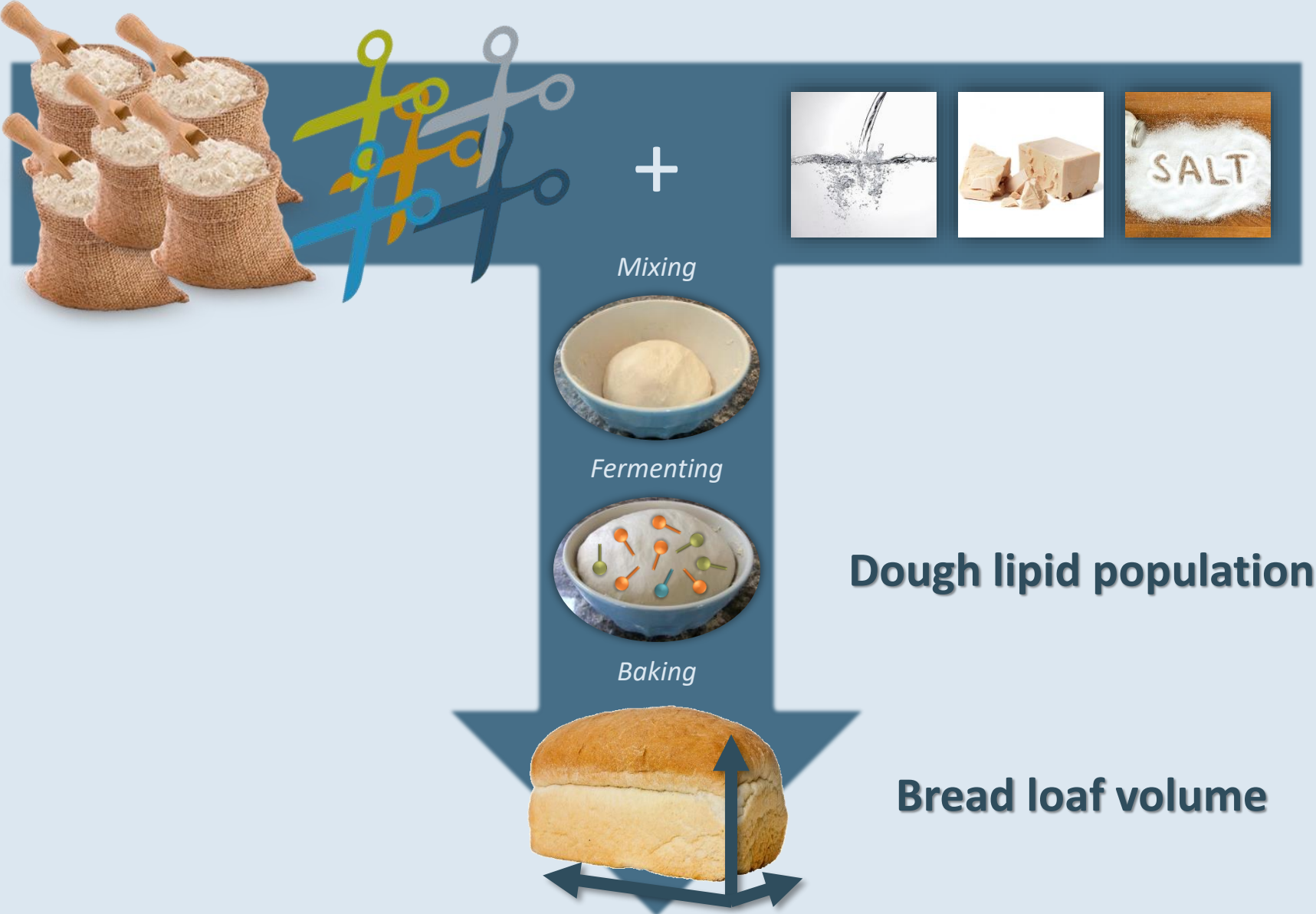


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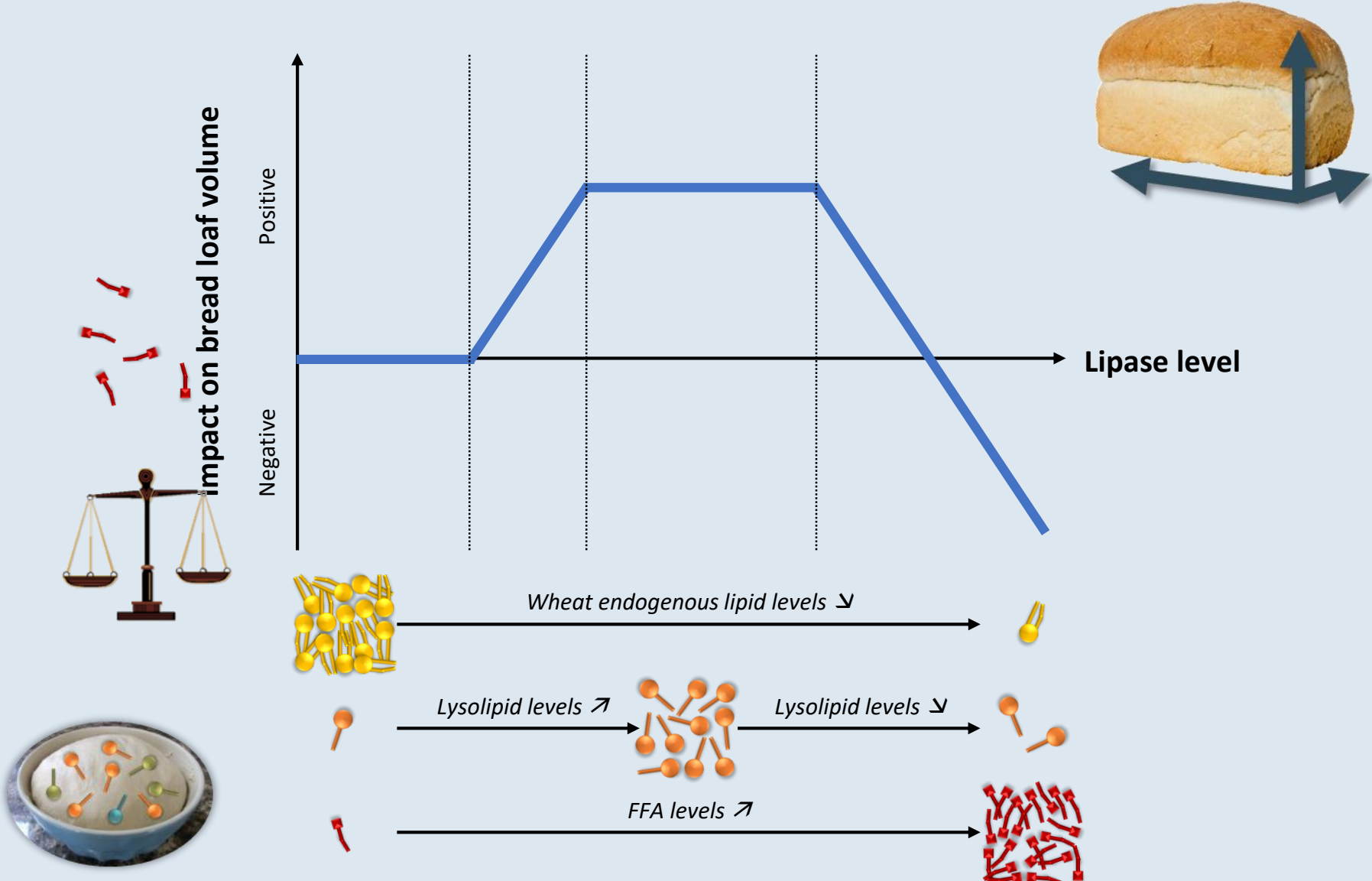
# Results and discussion

## IMPACT OF LIPIDS ON BREAD QUALITY



# Results and discussion

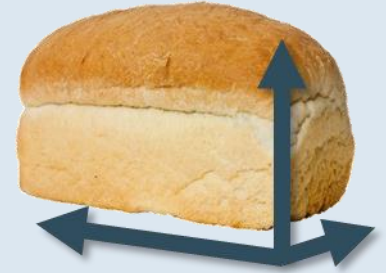
## IMPACT OF LIPIDS ON BREAD QUALITY



# Results and discussion

## IMPACT OF LIPIDS ON BREAD QUALITY

loaf volume  
Positive  
Negative



The positive effect of lipases in bread making is restricted by the unavoidable release of detrimental FFAs

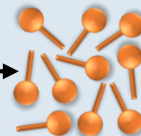


An appropriate balance between different types of lipids is crucial in bread making

Lysolipid levels ↗

Lysolipid levels ↘

FFA levels ↗





# Results and discussion

## IMPACT OF LIPIDS ON BREAD QUALITY

- Perfect bread making lipases preferably



MGDGs and NAPEs

↗ highly beneficial MGMGs

↘ detrimental NAPEs



DGDGs

- DGDGs:DGMGs  $\approx$  1:1 (synergistic effects)



TAGs and PCs

- Limit release of detrimental FFAs



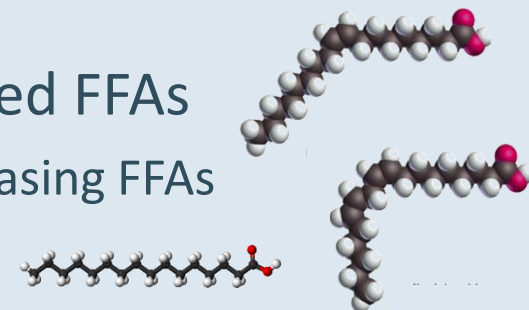
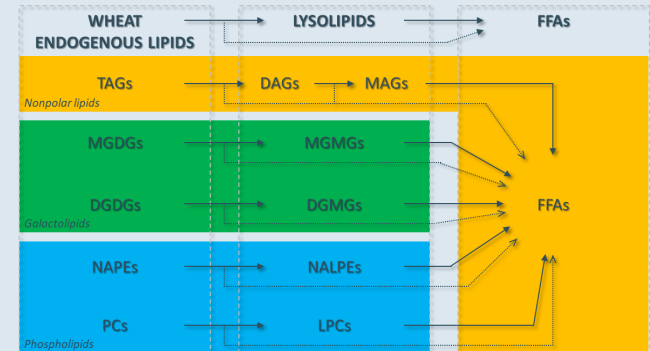
Lysolipids

- More beneficial than their precursors



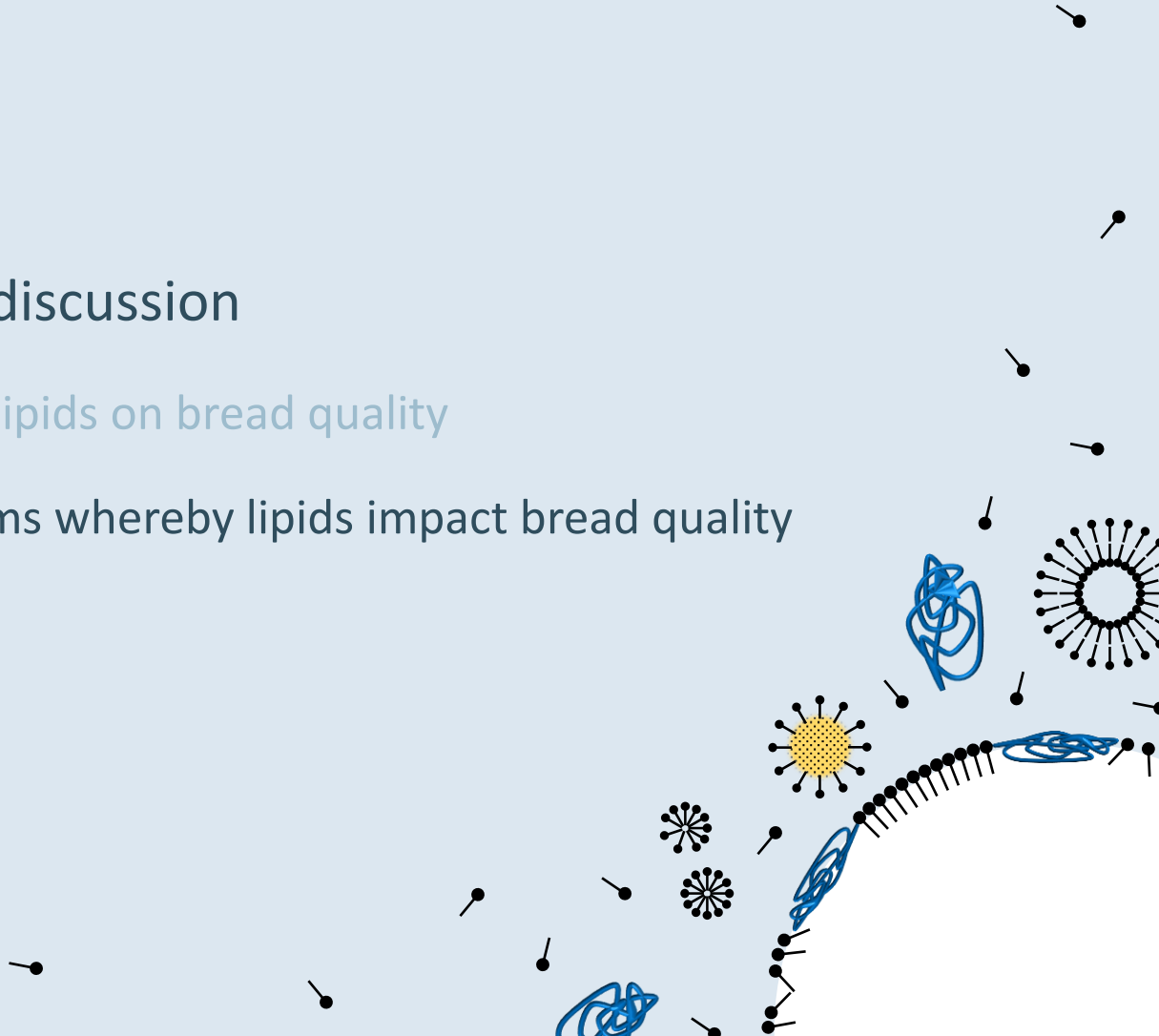
Release saturated rather than unsaturated FFAs

- Presumably delays detrimental effect of releasing FFAs



# Overview

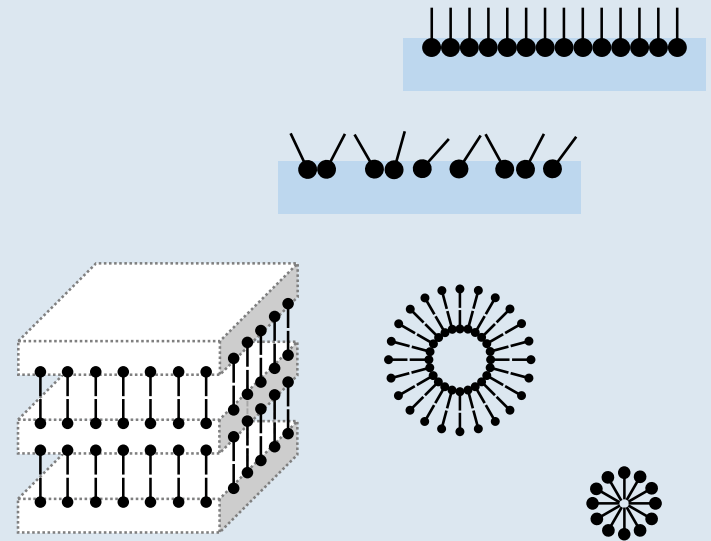
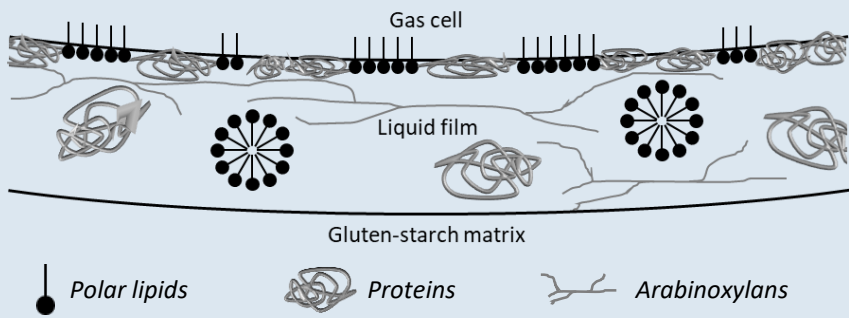
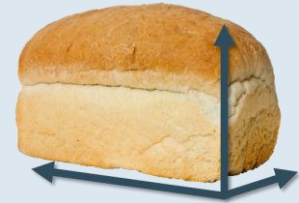
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  - Mechanisms whereby lipids impact bread quality
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# Results and discussion

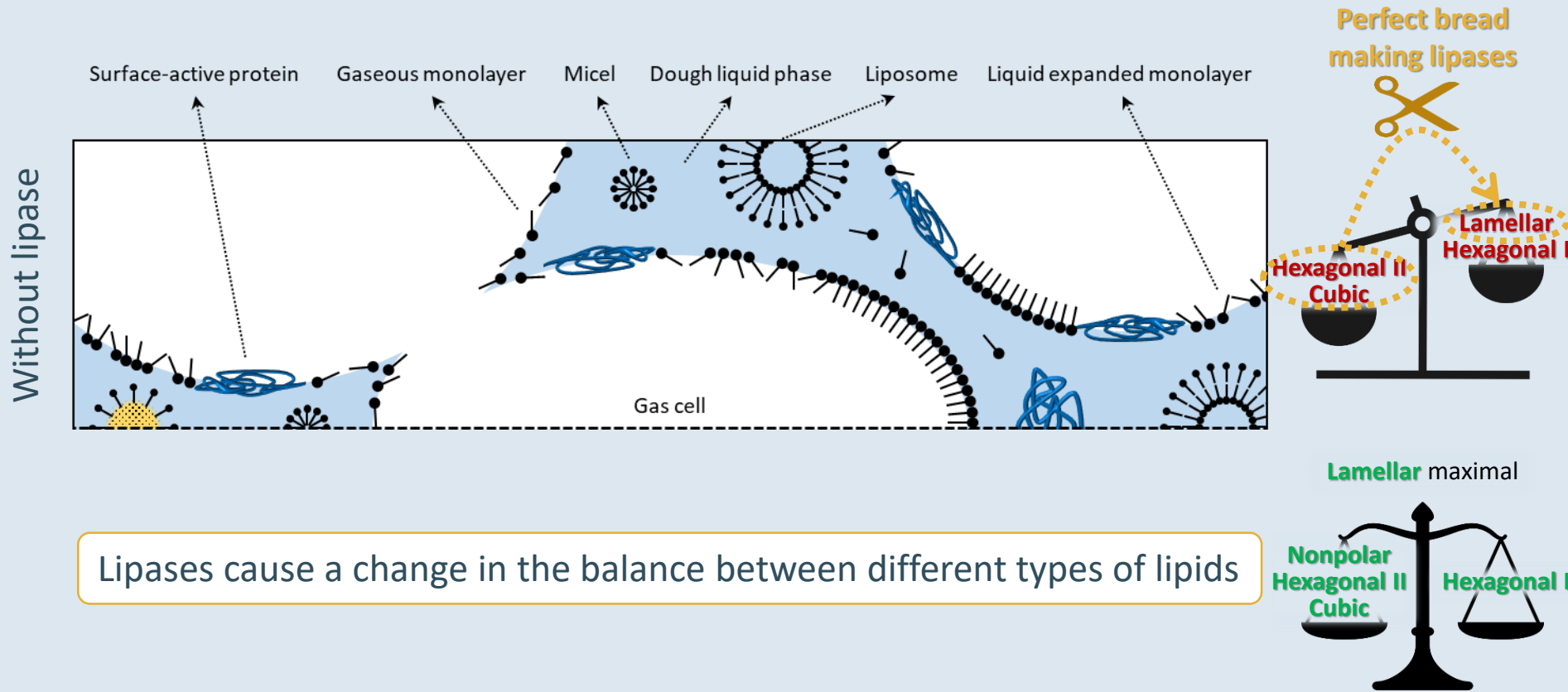
## *MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY*

- A **mechanism** whereby wheat endogenous lipids and their enzymatically released hydrolysis products affect bread loaf volume was proposed
  - Entirely relies on the ability of lipids to **directly** stabilize gas cells in dough



# Results and discussion

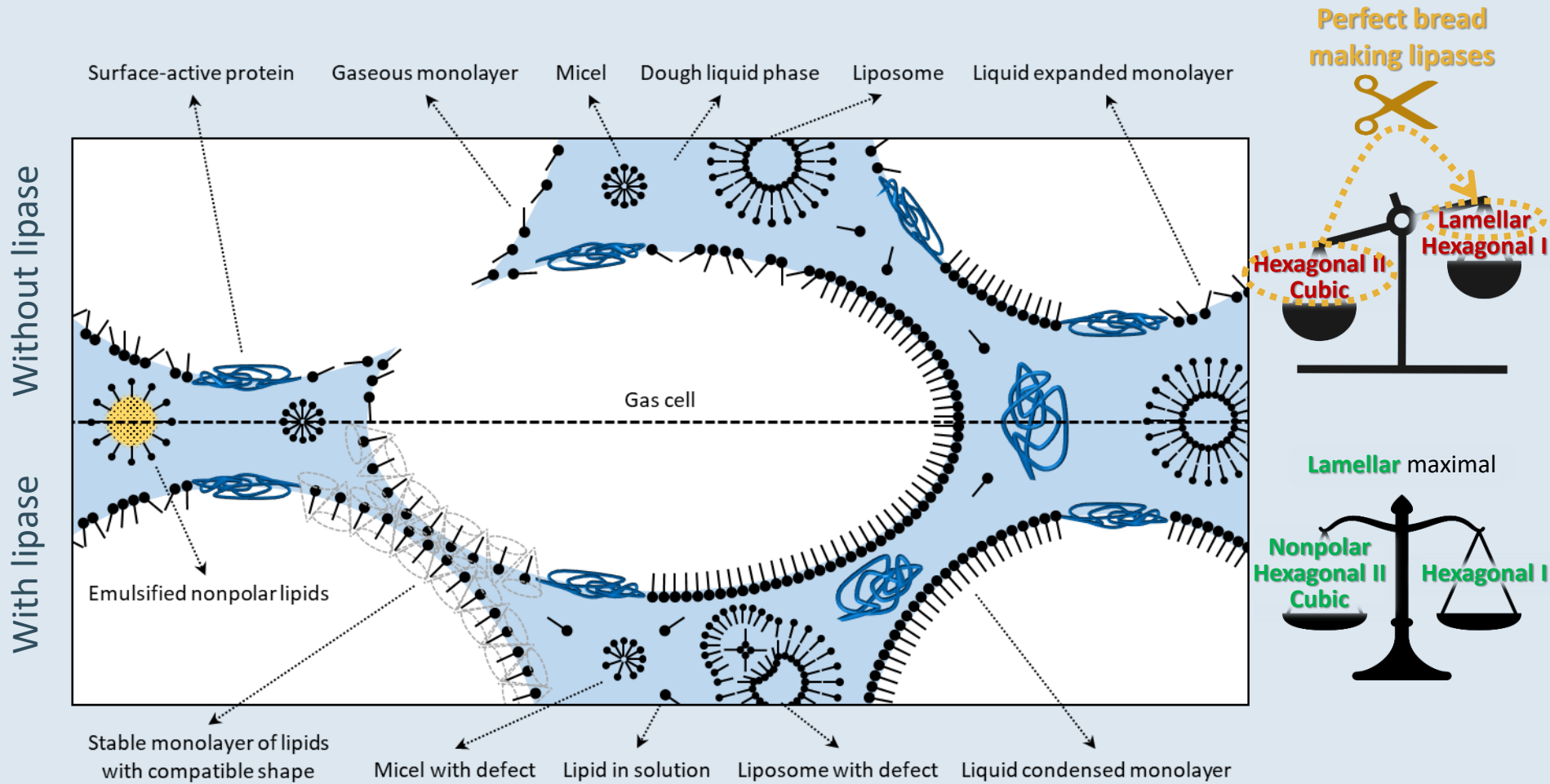
## MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY



Lipases cause a change in the balance between different types of lipids

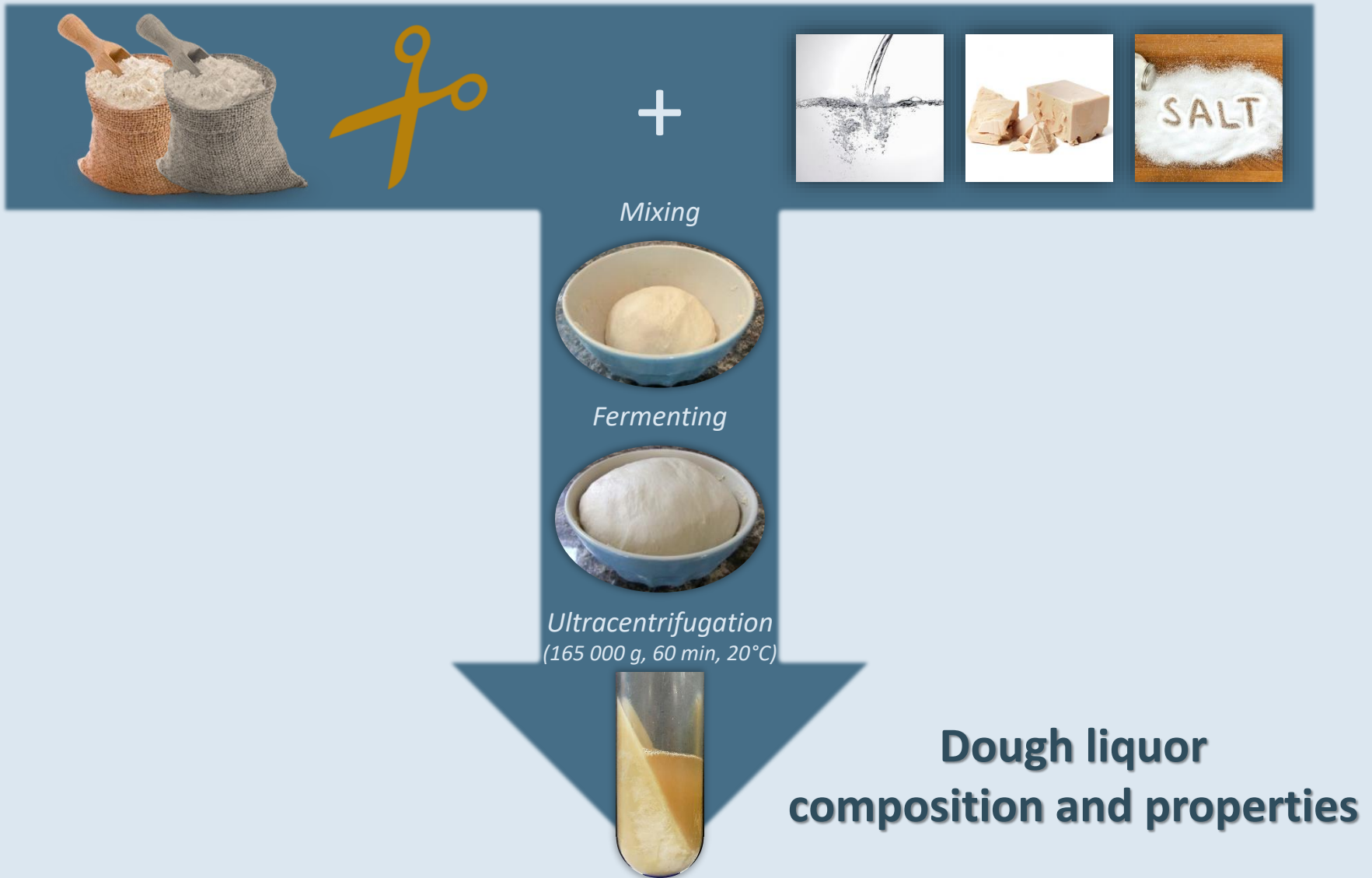
# Results and discussion

## MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY



# Results and discussion

## *MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY*



# Results and discussion

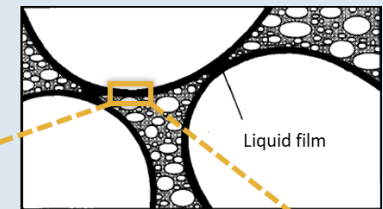
## *MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY*

### ■ Dough liquor

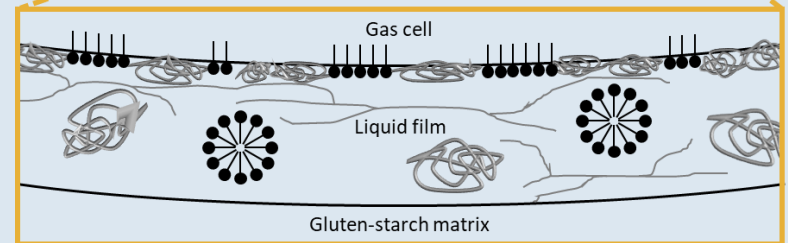
~ Fraction representative for liquid films surrounding gas cells in dough

### ■ Composition and properties

→ information about (components responsible for) **direct** gas cell stabilization in dough



Advanced stages of fermentation to early stages of baking

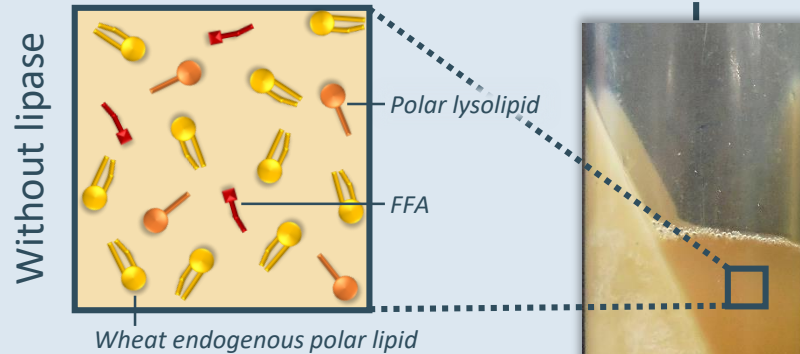


# Results and discussion

## *MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY*

- **Dough liquor** → **Direct** impact on gas cell stability

Lipid composition



Interfacial properties

**Practically no polar lipids recovered!**

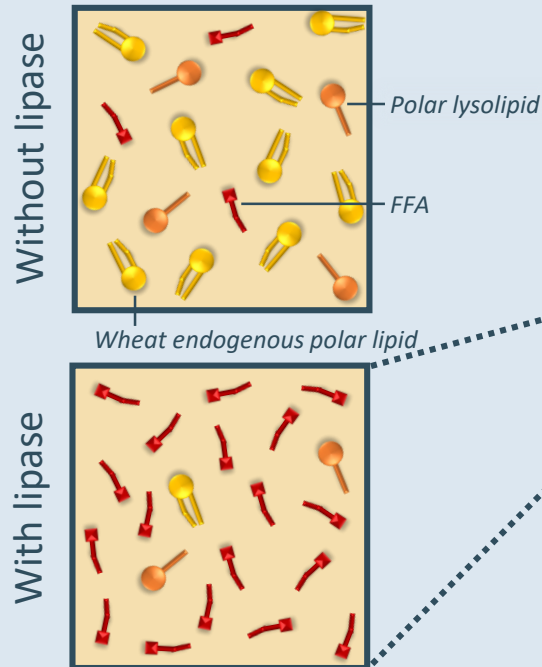


# Results and discussion

## MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY

- **Dough liquor** → **Direct** impact on gas cell stability

Lipid composition



Interfacial properties



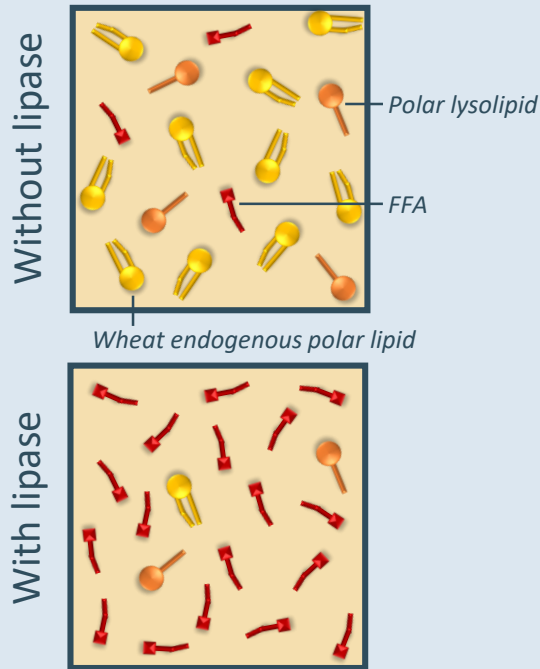
**Practically no polar lipids recovered!**

# Results and discussion

## MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY

- **Dough liquor** → **Direct** impact on gas cell stability

Lipid composition





Practically no polar lipids recovered!



Interfacial properties

- **Surface tension ( $\gamma$ )**
- **Surface dilatational modulus (E)**

Lipase impact on...

	Dough liquor		Bread
	$\gamma$	E	LV
	↘	≈	+ 30%
	↘↘	↗	+ 30%

No relation!

# Results and discussion

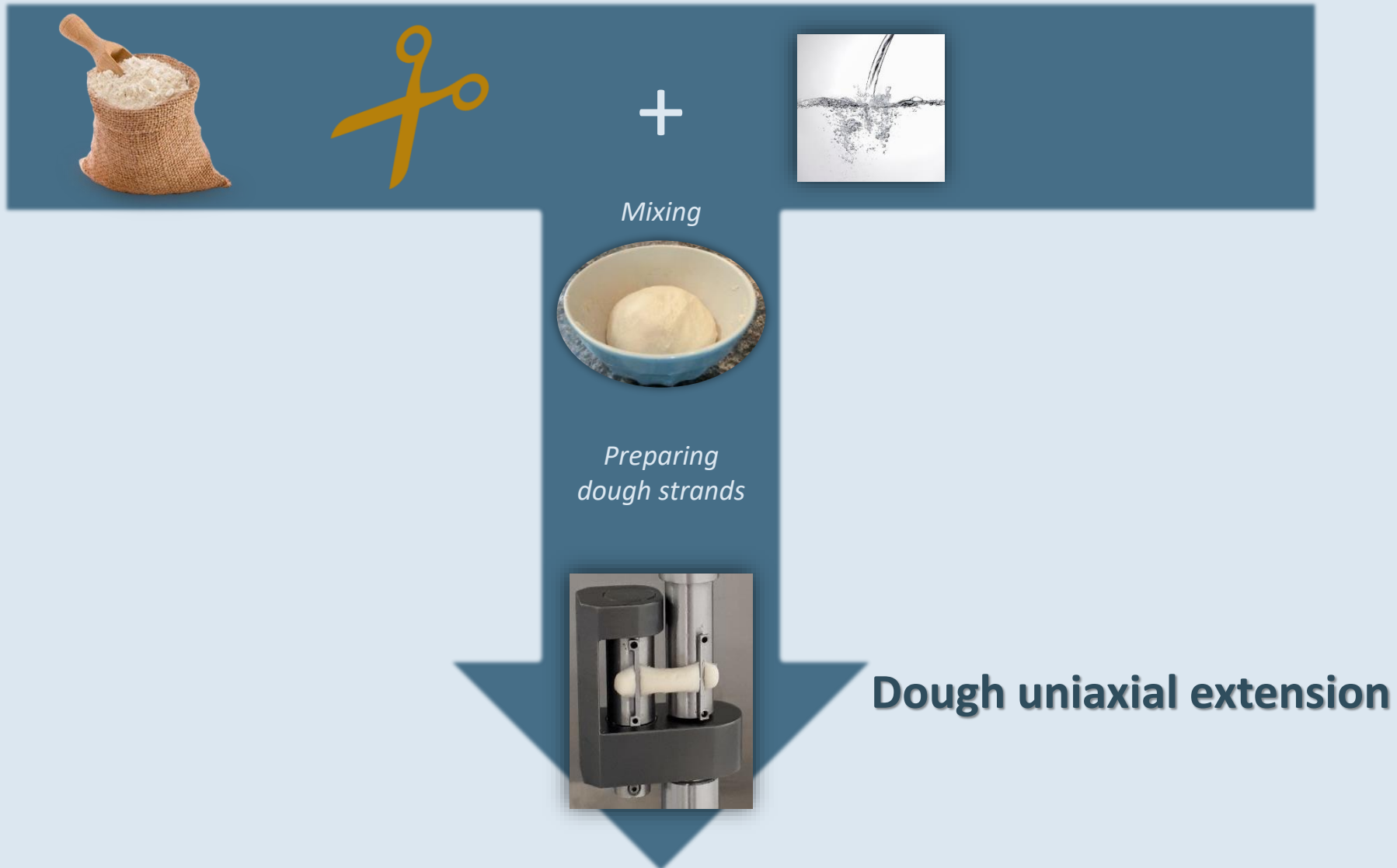
## *MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY*

- **Dough liquor** → **Direct** impact on gas cell stability
  - No relation between lipase-induced changes in...
    - Dough liquor composition and interfacial properties
    - Bread loaf volume
- Lipase impact on loaf volume not due to altered dough liquor composition and/or interfacial properties
- Unclear whether this is due to
  - Lipases having no positive impact on interfacial gas cell stability during bread making
  - Dough liquor not being an appropriate model system for gas cell interfaces in dough



# Results and discussion

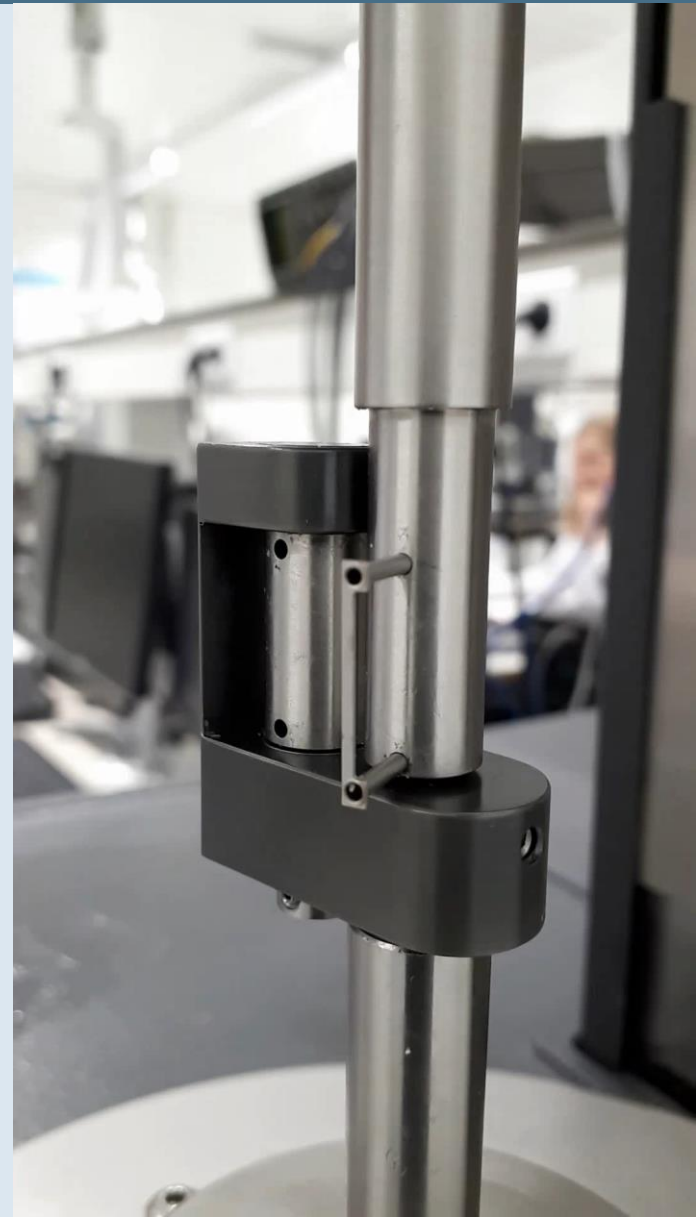
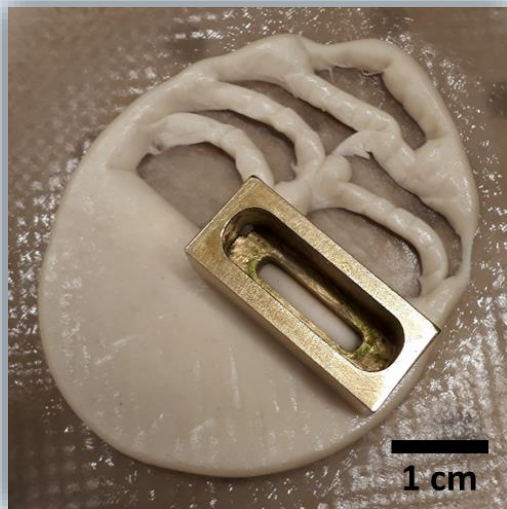
## *MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY*



# Results and discussion

## *MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY*

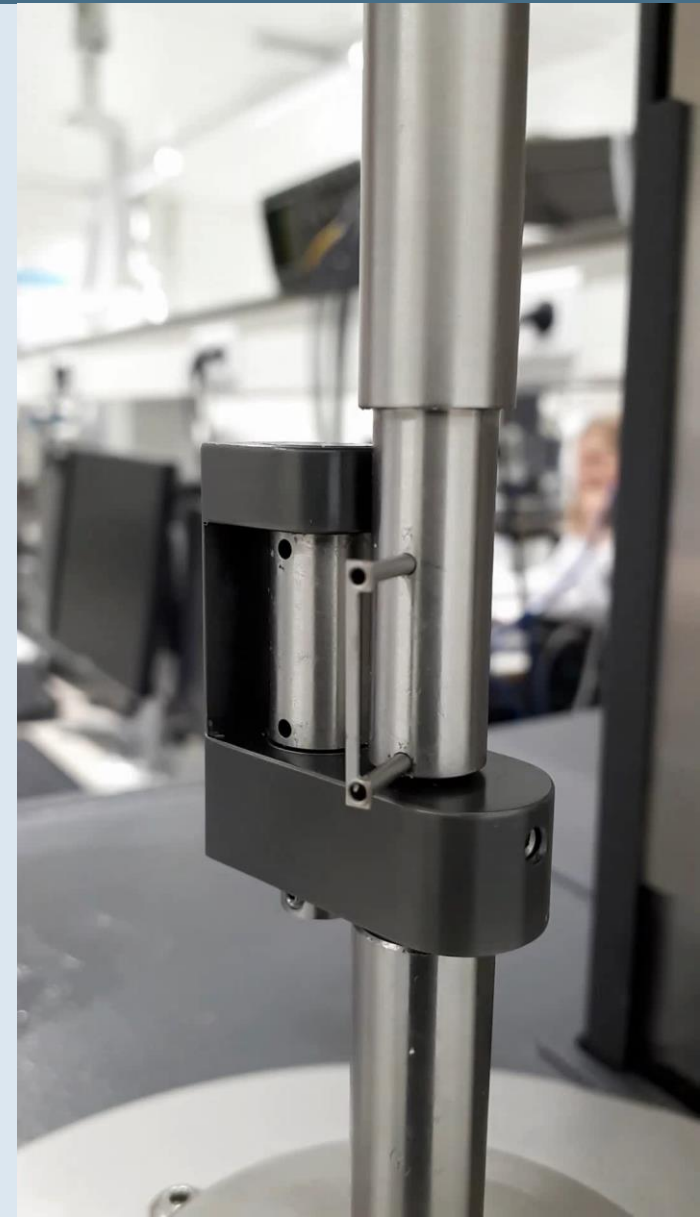
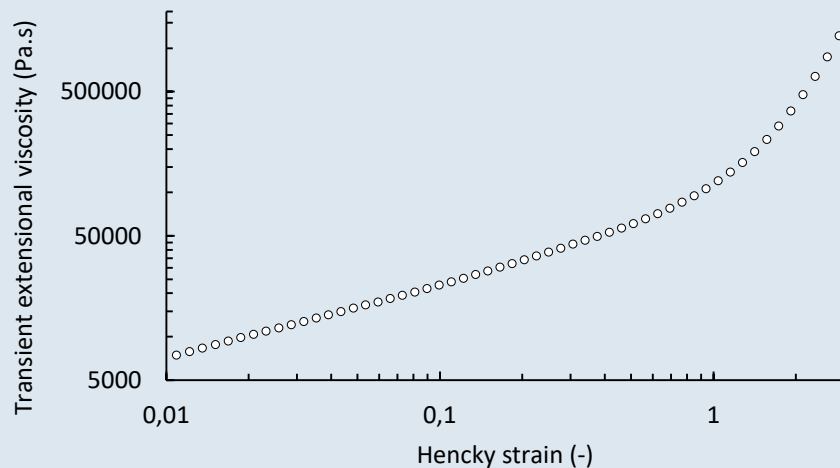
- **Dough uniaxial extension**
  - ~ Dough flow behavior or rheology
- Create dough strands



# Results and discussion

## *MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY*

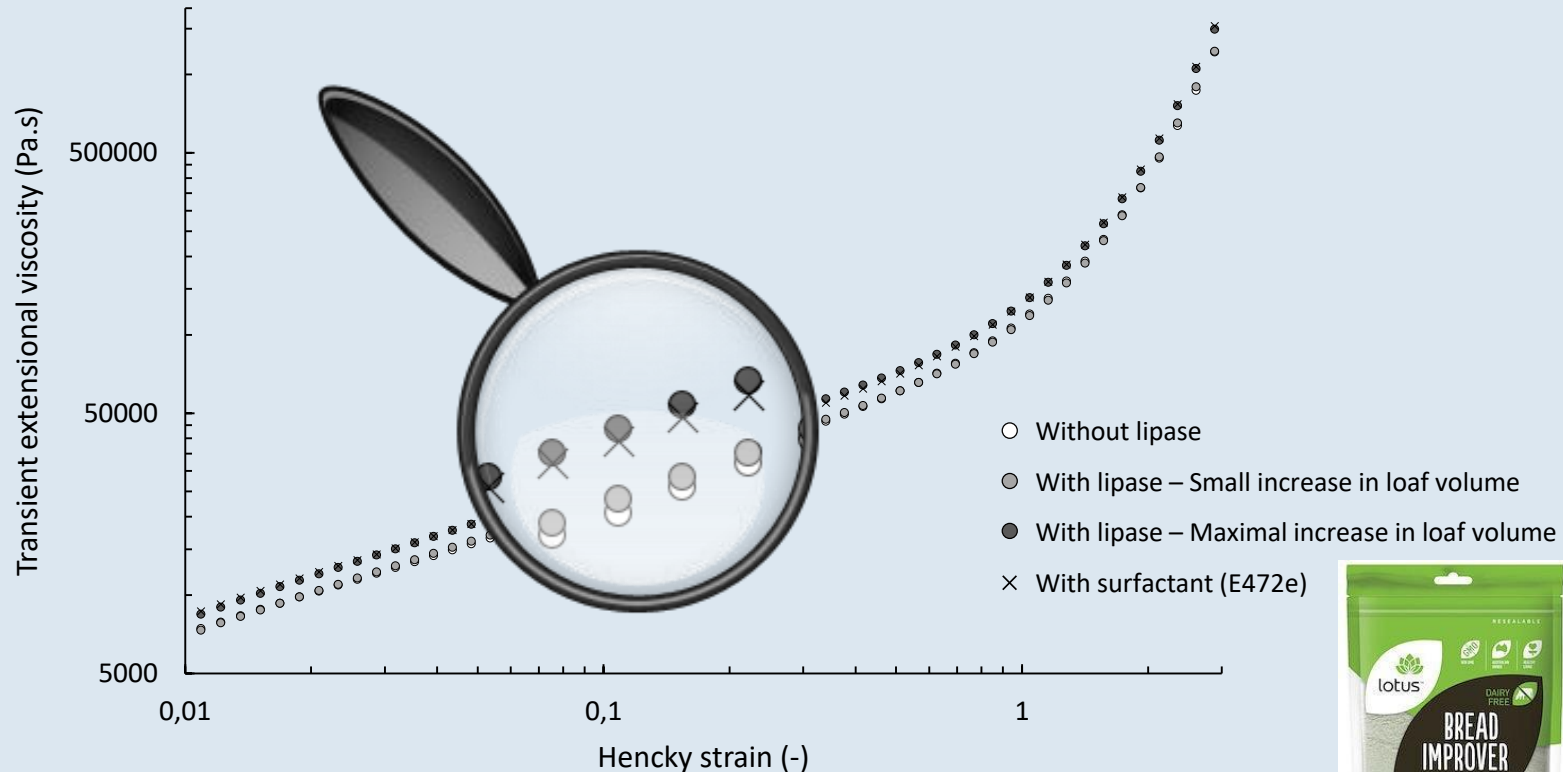
- **Dough uniaxial extension**
  - ~ Dough flow behavior or rheology
- Create dough strands
- Output



# Results and discussion

## MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY

- **Dough uniaxial extension**  **Indirect** impact on gas cell stability



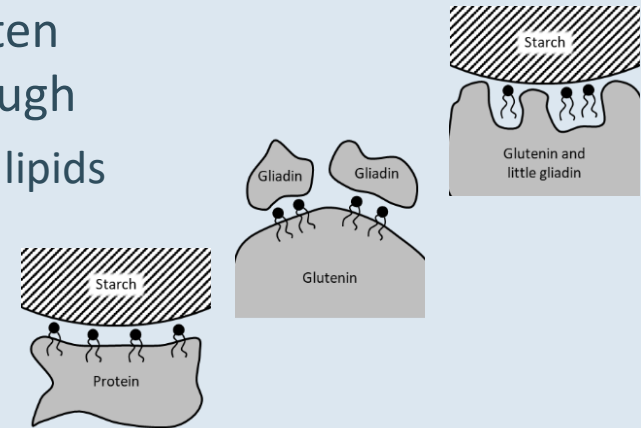
# Results and discussion

## MECHANISMS WHEREBY LIPIDS IMPACT BREAD QUALITY

- **Dough uniaxial extension** ☞ **Indirect** impact on gas cell stability

- Including lipase increased dough extensional viscosity

- Enhanced lipid-mediated starch-gluten and gluten-gluten interactions in dough
  - Due to increased polarity of dough lipids
  - ~ Lipids “bridge” starch and gluten
  - Improvement of
    - Dough strength
    - Dough gas holding capacity



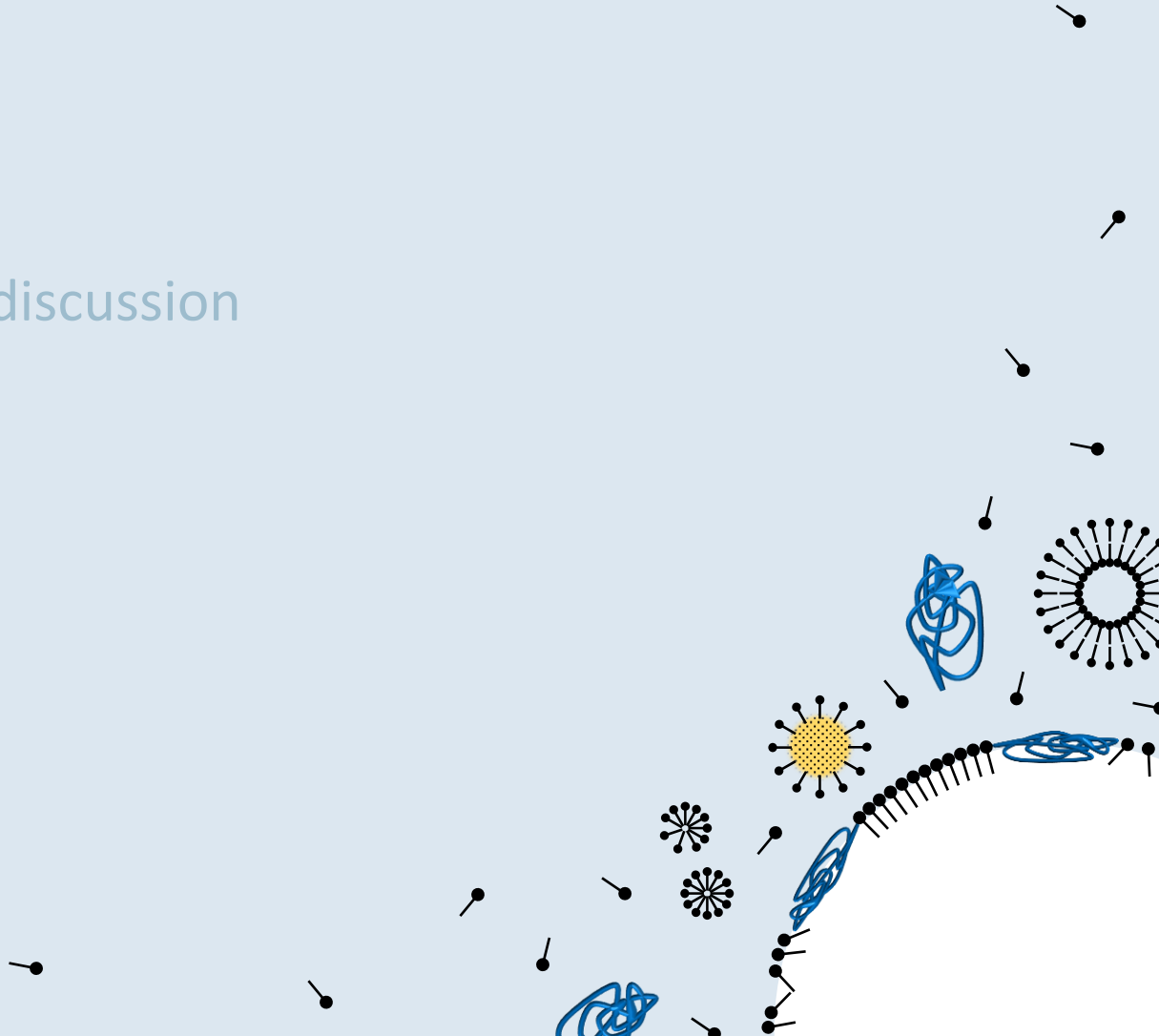
→ Lipids can contribute to the stability of gas cells in dough in an **indirect** way





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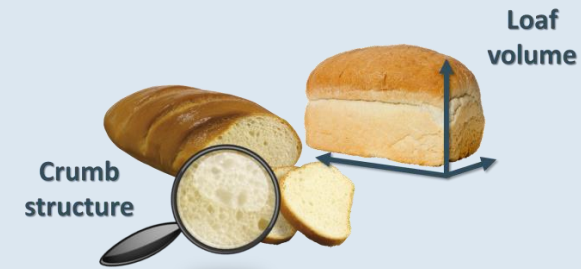


# Conclusions

*Wheat endogenous lipids and their enzymatically released hydrolysis products greatly impact bread loaf volume*

- Lipids —●

- Minor constituents in flour
- Excellent targets for improving bread quality



- Perfect bread making lipases preferably



MGDGs and NAPEs



DGDGs



TAGs and PCs



Lysolipids



Release saturated rather than unsaturated FFAs



# Conclusions

*Wheat endogenous lipids and their enzymatically released hydrolysis products greatly impact bread loaf volume*

- Appropriate balance between different types of lipids is crucial in bread making
  - Beneficial effects of enzymatically converting endogenous lipids into their corresponding lysolipids
  - Unavoidable release of detrimental FFAs

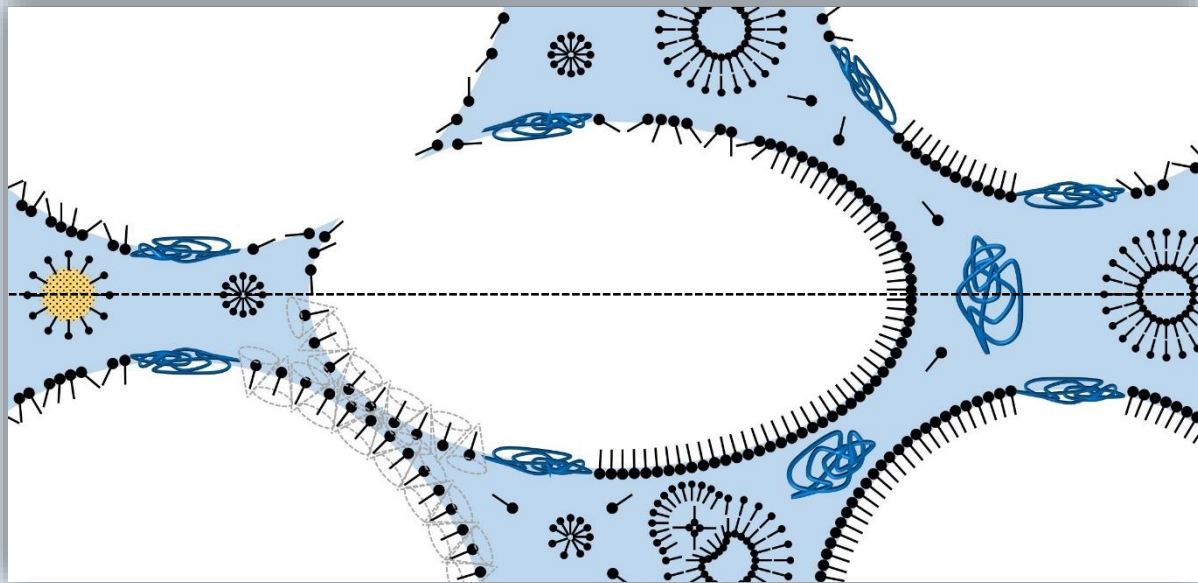


Findings aid in the development of new lipases and wheat varieties for bread making applications



# Conclusions

*A mechanism whereby wheat endogenous lipids and their enzymatically released hydrolysis products affect bread loaf volume was proposed*



- Completely relies on the ability of lipids to **directly** stabilize gas cells

# Conclusions

*A mechanism whereby wheat endogenous lipids and their enzymatically released hydrolysis products affect bread loaf volume was proposed*

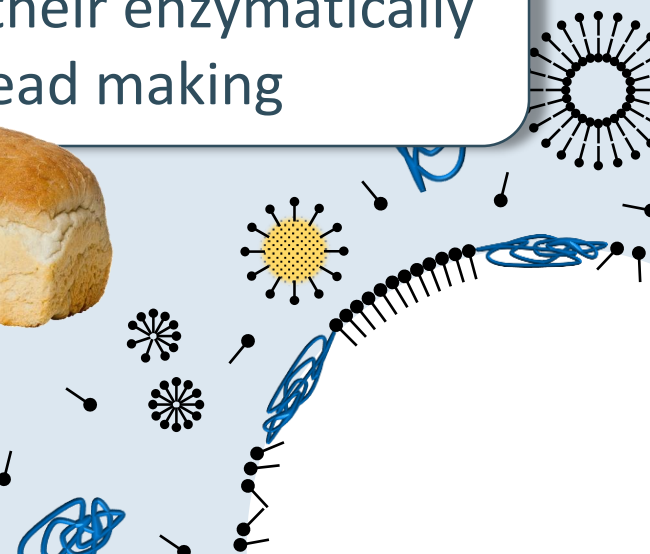
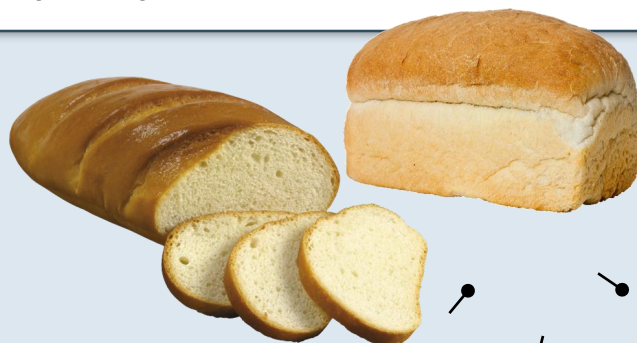
- Validity could not be proven
  - No relation between lipase-induced changes in...
    - Dough liquor composition and interfacial properties
    - Bread loaf volume
  - Lipases no positive impact on interfacial gas cell stability?
  - Dough liquor not an appropriate model system for gas cell interfaces?
- An **indirect** impact of lipids on the stability of gas cells in dough was demonstrated



# Conclusions

*Altogether, we suspect that the impact of wheat endogenous lipids and their enzymatically released hydrolysis products on bread loaf volume results from both their **indirect** and **direct** effect on gas cell stability*

Findings contribute to the development of a unified theory on the role of wheat endogenous lipids (and their enzymatically released hydrolysis products) in bread making





# The role of wheat endogenous lipids and their enzymatically released hydrolysis products in bread making

**Sara MELIS and Jan A. DELCOUR**

ICBC 2021 | 30 March 2021 | Online event

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