Whey protein emulsions : how to control texture in a large range of protein concentration.

Thomas Croguennec, Marie Chevallier, Thibault Loiseleux, Catherine Garnier, Alain Riaublanc, Christelle Lopez

Joint Research Unit

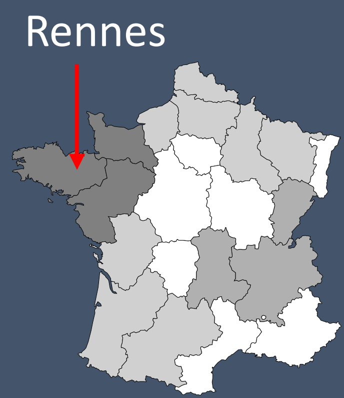
STLO

AGROCAMPUS Ouest–INRA

Science and Technology of Milk and Eggs (STLO)

Whey protein emulsions: how to control texture in a large range of protein concentration ?

CHEVALLIER M, LOISELEUX T, LOPEZ C, GARNIER C, ANTON M, RIAUBLANC A, **CROGUENNEC Thomas**



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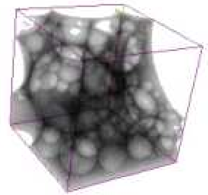
STLO: main research areas

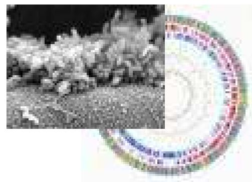
Increase the knowledge on **molecular and supramolecular structures** of milk and egg components

Understand protein-protein, protein-lipid, protein-mineral **interactions** leading to expected technological and bioactivity **functionalities** and understand their **digestion**



Analyze **transfers** during **technological processes**

Characterize **interactions between bacteria/environment** in relationship with product quality, food safety and probiotic activity



Whey protein emulsions: how to control texture in a large range of protein concentrations ?

Introduction

1. Design of fluid whey protein emulsions after heating at high whey protein concentrations
2. Preparation of texturized (gelled) whey protein emulsions at low whey protein concentrations

Conclusion

Acknowledgments

Introduction

Whey proteins : Large amount available around the world

MILK

CHEESE MANUFACTURE

CHEESE
(20 millions tonnes)

- ♦ 10% of the milk volume
- ♦ >80% of the milk proteins (caseins)

WHEY

- ♦ 90% of the milk volume
- ♦ <20% of the milk proteins (**whey proteins**)

From waste stream to value-added,
« gutter-to-gold » *Smithers, 2008*

- Exceptional biological value (amino acid composition)
- Ligand carriers (vitamins, minerals, fatty acids)
- Bioactive proteins and peptides

Excellent for human nutrition

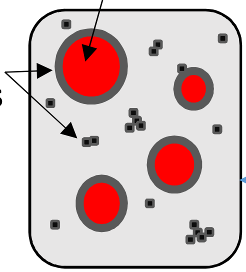
Dairy manufacturers:

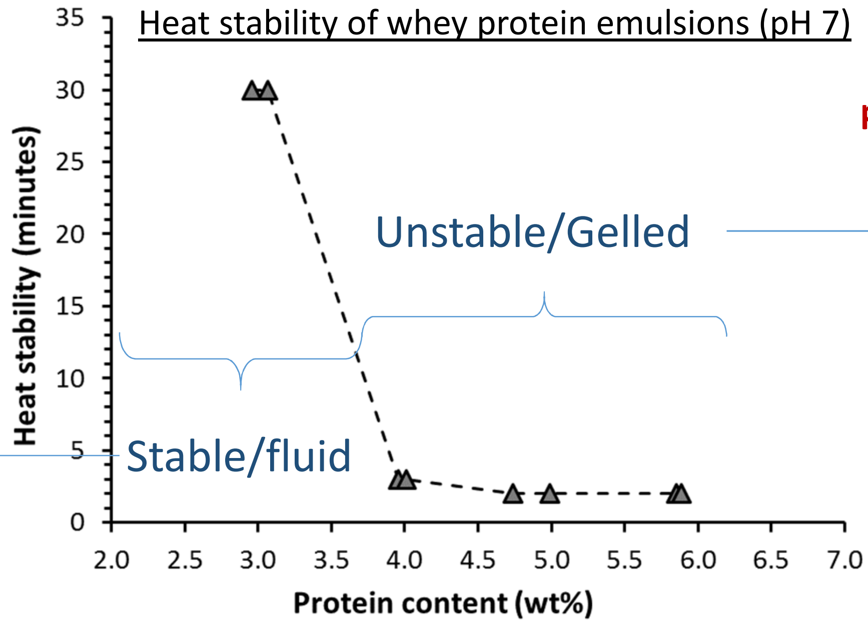
- **add value** to dairy compounds (*e.g. whey proteins*)

Introduction

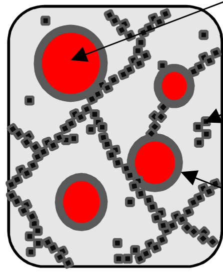
- Whey proteins :
  - natural emulsifiers (*adsorb to oil/water interface*)
  - heat-sensitive proteins (*heat treatments are required to extent food product shelf life*)
    - → heat sensitive emulsions
    - → texturized (gelled) emulsions

Fluid emulsions: Protein amount too low to build a continuous network





Gelled emulsions : Network of proteins entrapping oil droplets



Gelled emulsions obtained by using non-dairy **gelling agents**

Dairy industries

fluid emulsions obtained by using non-dairy **stabilizing agents**

Introduction

- New trends in Europe driven by consumer's expectations

  - **More natural and healthier food products**
  - **100% milk products** (without non-dairy additives)
  - **Innovative products** (new uses and consumption habits)

Research Questions

- How to design whey protein emulsions at high protein concentrations that are fluid after heating without non-dairy additives?
- How to obtain gelled whey protein emulsions at low protein concentrations without non-dairy additives ?

Whey protein emulsions: how to control texture in a large range of protein concentration ?

Introduction

1. Design of fluid whey protein emulsions after heating at high whey protein concentrations
2. Preparation of texturized (gelled) whey protein emulsions at low whey protein concentrations

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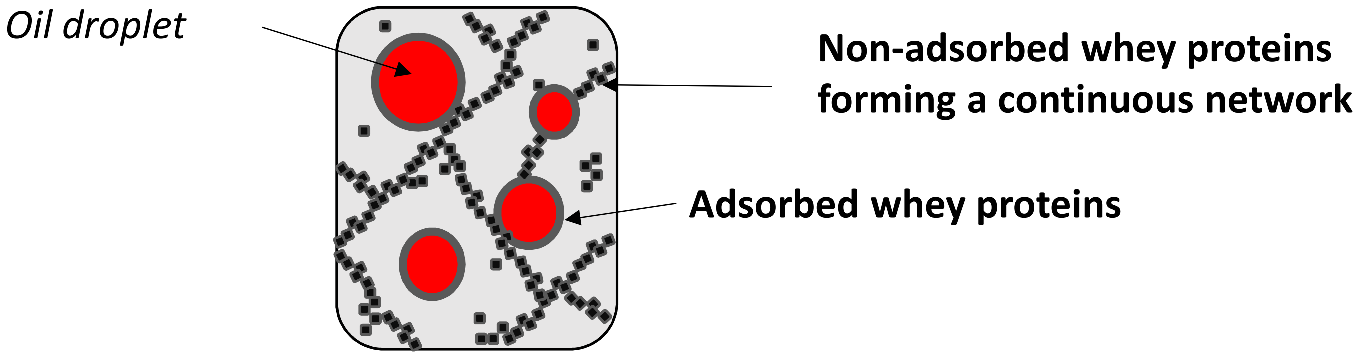
1. Design of fluid whey protein emulsions after heating

Emulsion gelation results from the establishment of **interactions between whey proteins**

- Whey proteins denature and aggregate during heating
- A gel is formed when the whey protein concentration is enough (critical gelation concentration)



To prevent emulsion gelation:

- Limit the interactions between proteins at the interface of oil droplets and proteins in the continuous phase
- Reduce the interactions between the proteins in the continuous phase

Objective : designing fluid emulsions at protein concentration higher than 4%

1. Design of fluid whey protein emulsions after heating

a. Have a look on scientific bibliography

- Whey protein aggregates are more heat stable in solution than native whey proteins (number and reactivity) *Ryan et al., 2012* (+)
- A combination of whey protein aggregates and native whey proteins improves emulsion heat-stability Çakır-Fuller, 2015 (+)
- Emulsifier properties of whey protein aggregates are reduced, compared to native proteins Kiokias et al., 2006 (−)
- Interfacial whey protein aggregates induce oil droplet flocculation on heating Sarkar et al., 2016 (−)

Whey protein aggregates are heat stable in solution (i.e. continuous phase of emulsion), but destabilize oil droplets (poor emulsifiers, bridging between droplets)

1. Design of fluid whey protein emulsions after heating

a. Have a look on scientific bibliography

- Caseins are heat stable and gives heat stable emulsions

(Hunt & Dalgleish, 1995; Srinivasan et al, 2002)

- Caseins adsorbed preferentially at fat droplet surface, compared to whey proteins.

Srinivasan et al., 1996

- Caseins improve the heat stability of whey protein emulsions (casein covered oil droplets do not contribute to whey protein network)

Dickinson & Parkinson, 2004; Parkinson & Dickinson, 2004

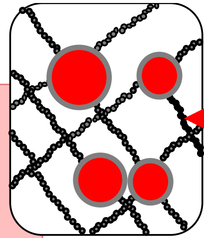
Caseins are good protector for oil droplets

1. Design of fluid whey protein emulsions after heating

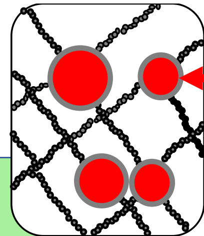
b. Strategy

Whey protein-rich emulsion stability depends on:

1. Stability of the **continuous phase** (replace native whey proteins by aggregated whey proteins)



2. Stability of the **oil droplets** (replace native whey proteins by caseins)

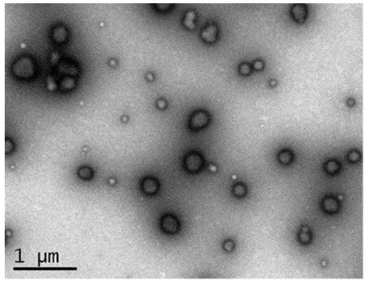


3. **Competition** for the oil droplet surface

1. Design of fluid whey protein emulsions after heating

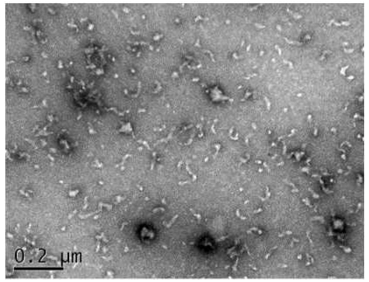
b. Strategy

What type of whey protein aggregates?

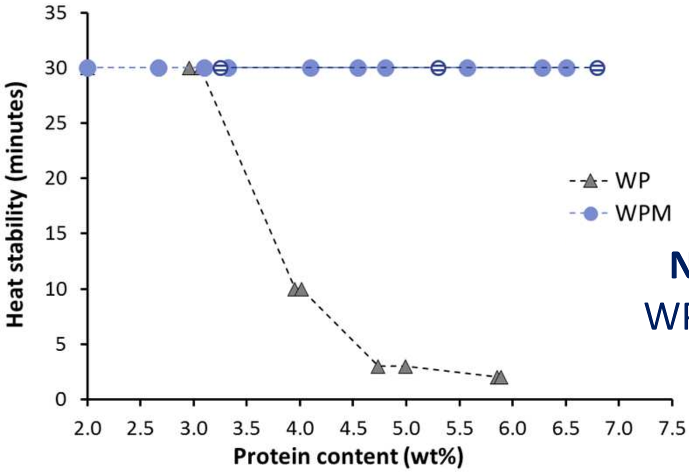


Whey protein microgels
(dense aggregates of ϕ~300nm)

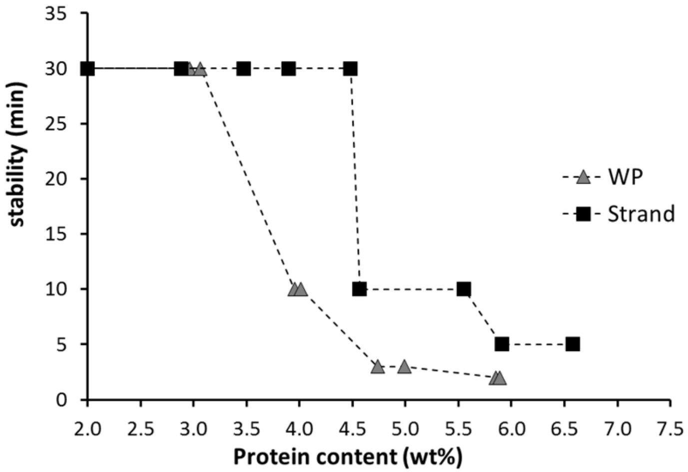
Whey protein **strands**
(low density aggregates of ϕ~70nm)



Heat stability of the native whey proteins (WP), whey protein microgels (WPM) and whey protein strands in solution (= continuous phase of the emulsion)



Number/reactivity:
WP >> stands >> WPM



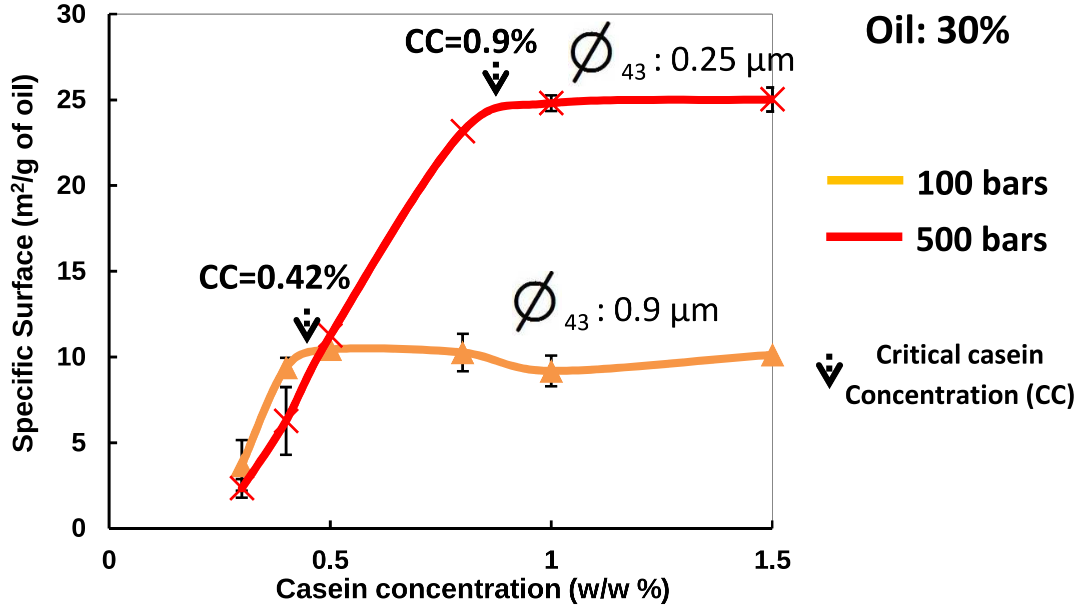
Whey protein microgels should be extremely stable in the continuous phase of the emulsion

1. Design of fluid whey protein emulsions after heating

b. Strategy

What amount of caseins in the emulsion ? (depend on the surface of the oil droplets)

Surface of the oil droplets depends on
- amount of oil in the emulsions
- size of the oil droplet (homogeneization pressure)



To have oil droplet fully covered by caseins:
Casein concentration > Critical casein concentration

1. Design of fluid whey protein emulsions after heating

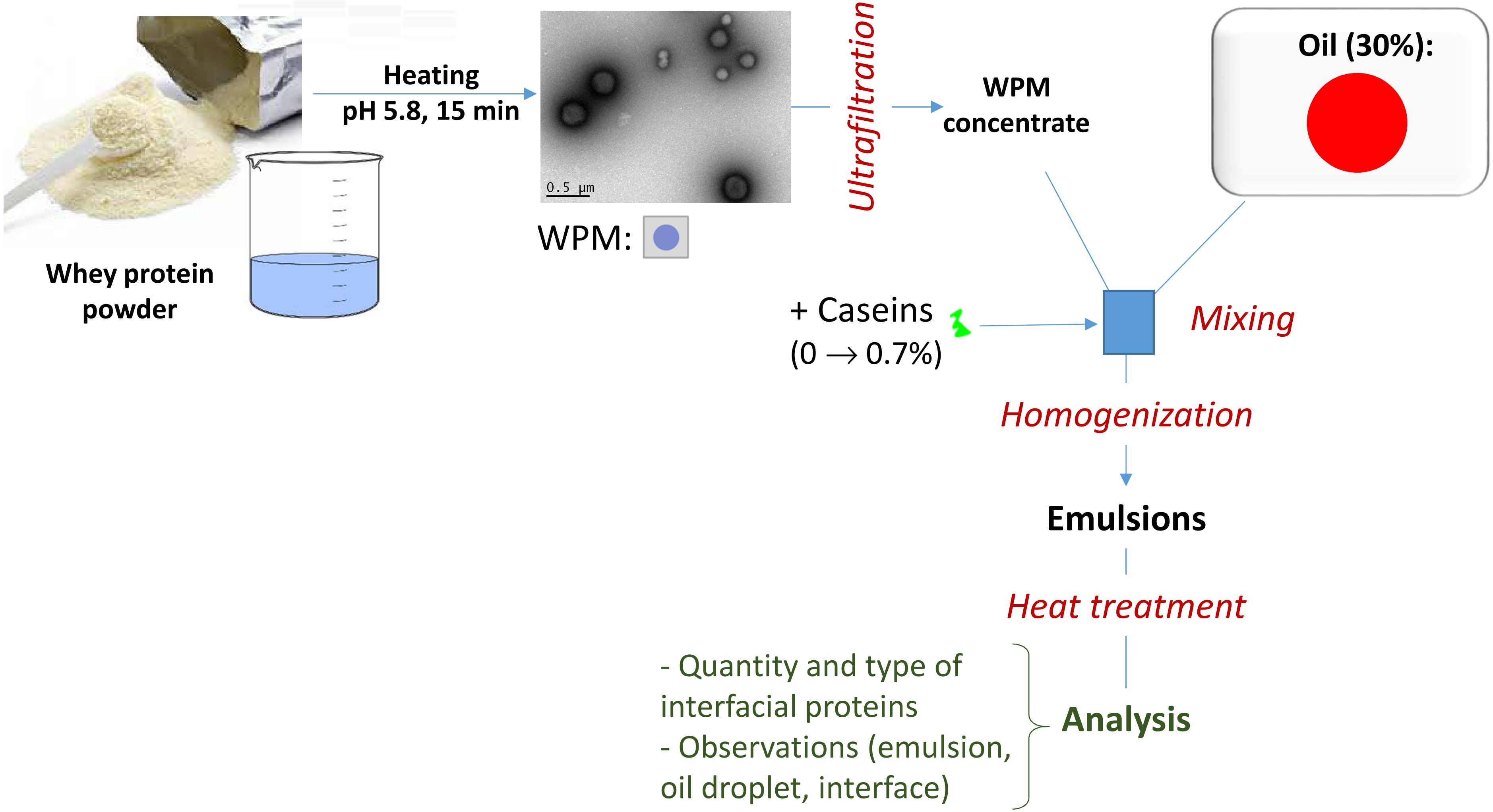
c. Hypothesis to test

♦ **Above the critical casein concentration**, the whey protein aggregates are released in the continuous phase, the oil droplets are covered exclusively by caseins and the **emulsions are stable** on heating

♦ **Below the critical casein concentration**, some whey protein aggregates adsorb at oil droplet surface and **destabilize the emulsions** on heating (casein amount is not enough to fully cover the oil droplet surface)

1. Design of fluid whey protein emulsions after heating

d. Materials and methods



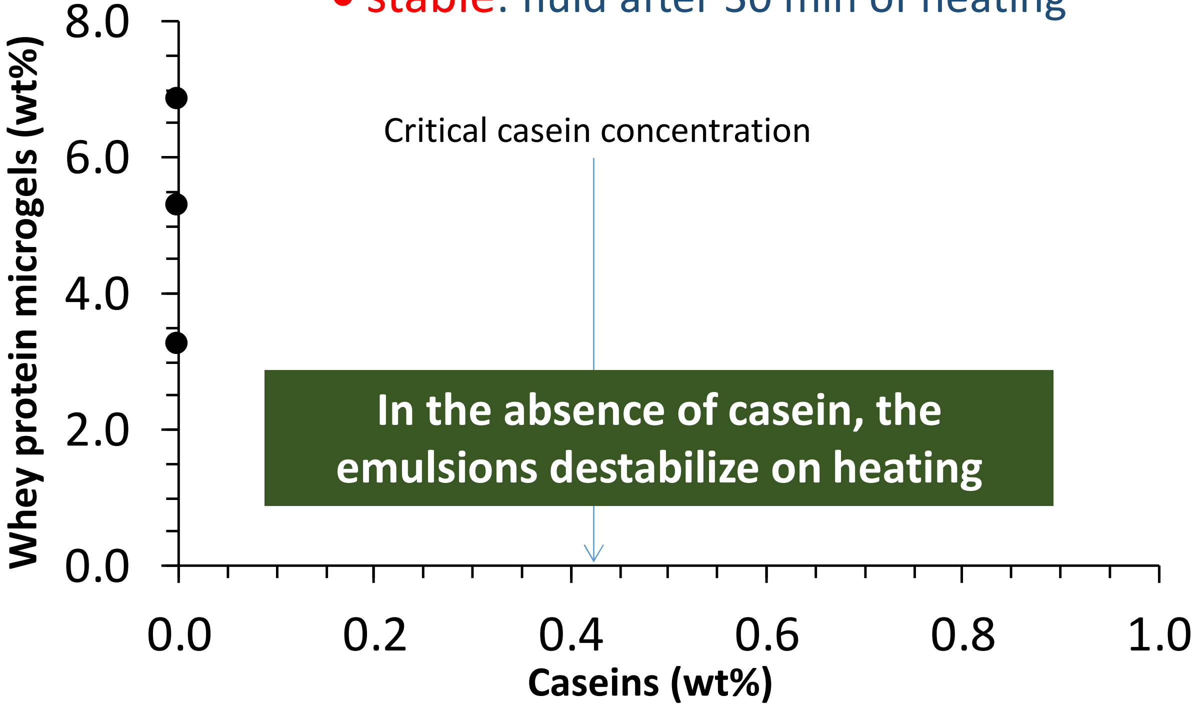
1. Design of fluid whey protein emulsions after heating

e. Results

- Emulsions **below the critical casein concentration** at different whey protein microgel concentrations

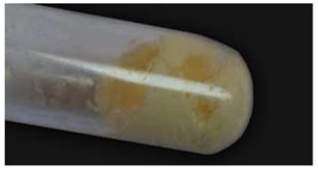
Test of heat stability: heating at 120°C up to 30 min

- unstable: gelled before 5 min of heating
- stable: fluid after 30 min of heating

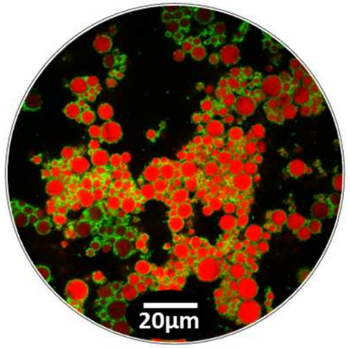


In the absence of casein, the emulsions destabilize on heating

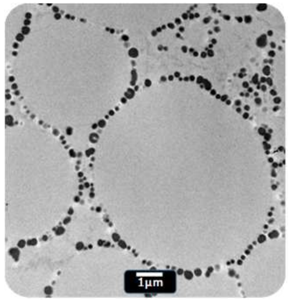
Observations (after 1 min of heating)



Emulsion scale



Oil droplet scale



Oil droplet surface scale

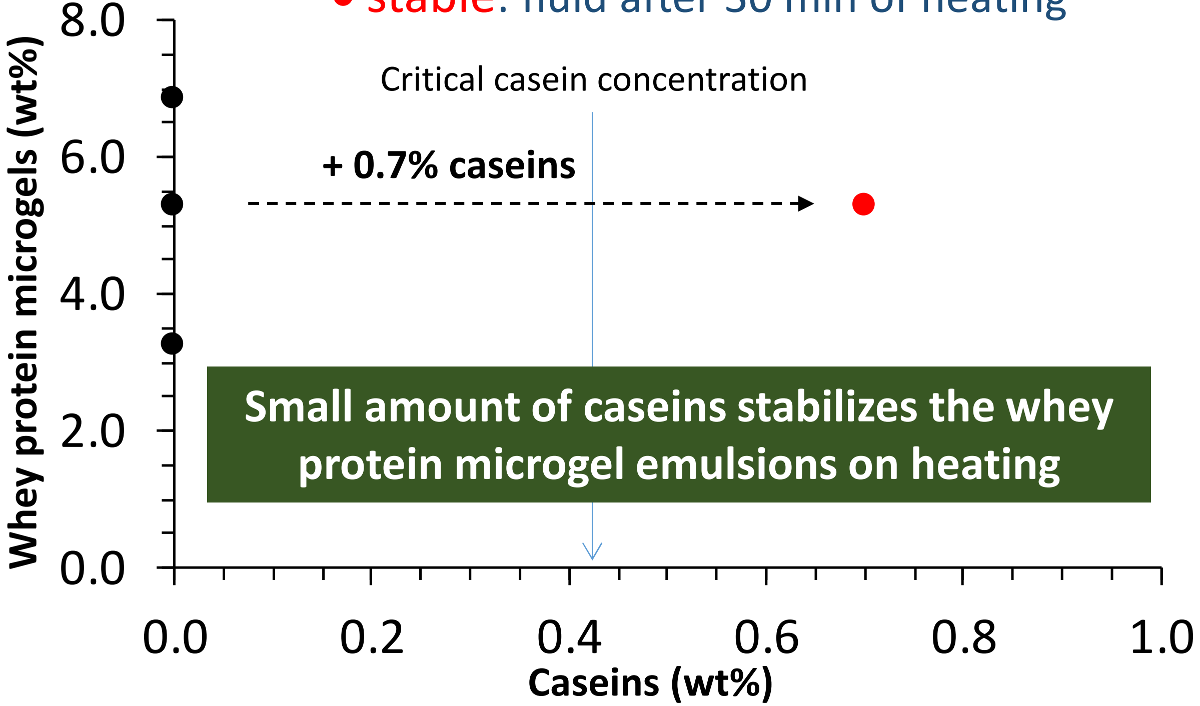
1. Design of fluid whey protein emulsions after heating

e. Results

- Emulsions **above the critical casein concentration**: +0.7% caseins

Test of heat stability: heating at 120°C up to 30 min

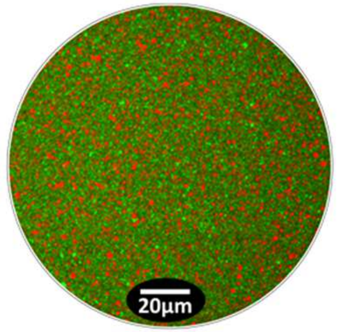
- unstable: gelled before 5 min of heating
- stable: fluid after 30 min of heating



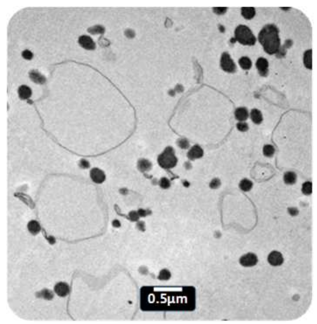
Small amount of caseins stabilizes the whey protein microgel emulsions on heating

Observations (after 30 min of heating)

Emulsion scale



Oil droplet scale

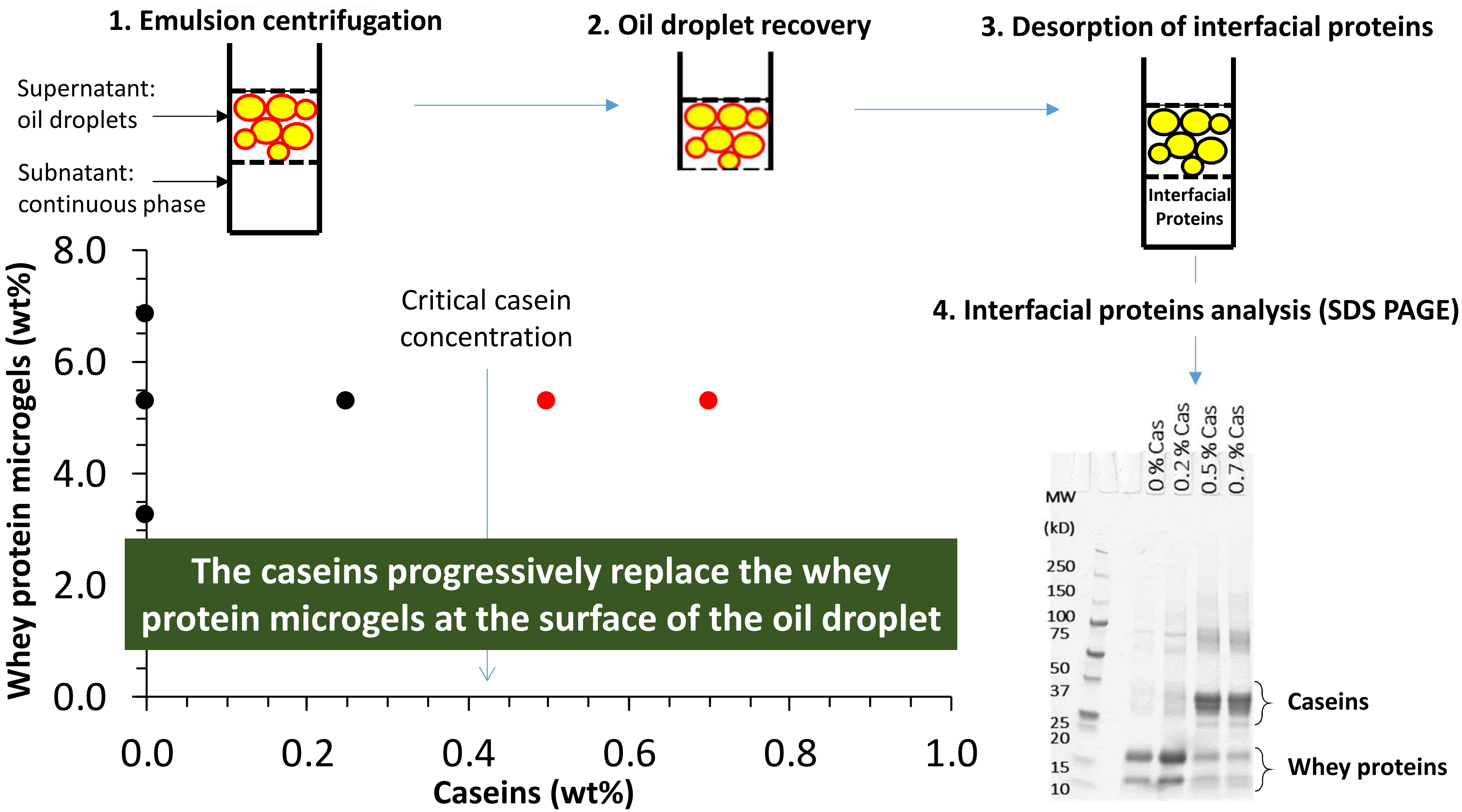


Oil droplet surface scale

1. Design of fluid whey protein emulsions after heating

e. Results

- Emulsions at different casein concentrations: **analysis of the fat droplet surface composition**



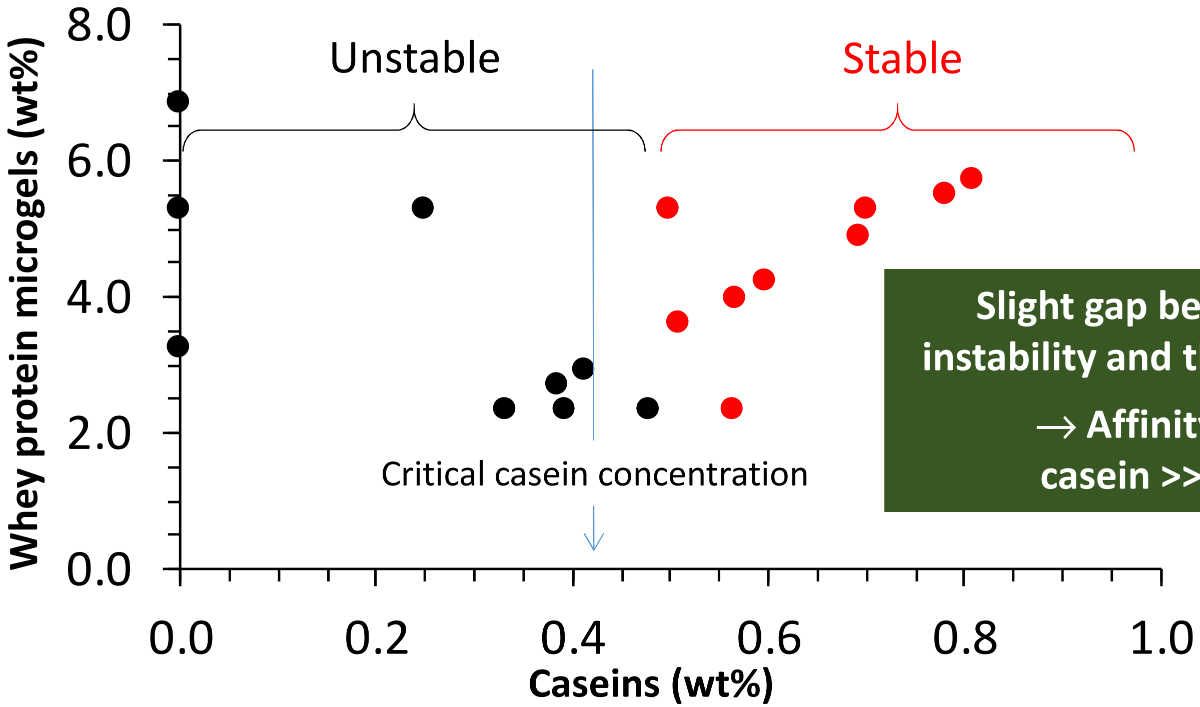
The caseins progressively replace the whey protein microgels at the surface of the oil droplet

1. Design of fluid whey protein emulsions after heating

e. Results

• Generalization for emulsions at different concentration of caseins and whey protein microgels in order to support the hypothesis

Thermal Stability of the emulsions



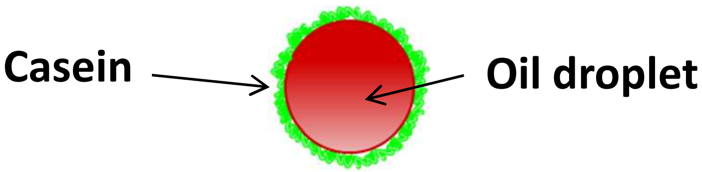
Slight gap between the limit of emulsion instability and the critical casein concentration

→ Affinity for oil droplet surface : casein >> whey protein microgels

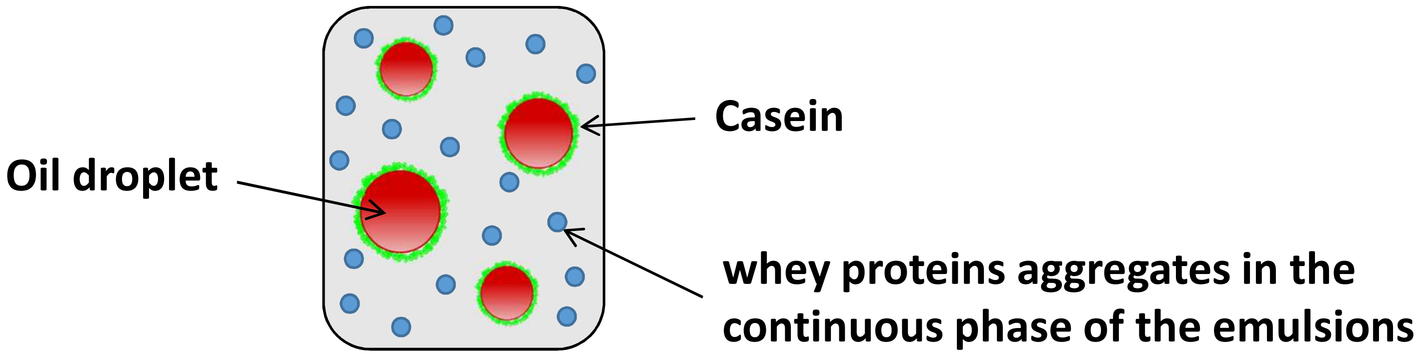
1. Design of fluid whey protein emulsions after heating

How to design whey protein emulsions at high protein concentrations that are fluid after heating in the absence of non-dairy additives?

- by adding sufficient amount of caseins to cover oil droplet surface
  → estimated value: mass of oil (g) × specific surface (m^2/g of oil) × protein interfacial load (g/m^2)
  (protein interfacial load ~ 2mg/m^2 for caseins)



- By selecting large and dense whey protein aggregates (*small number and low reactivity on heating*)



Whey protein emulsions: how to control texture in a large range of protein concentration ?

Introduction

1. Design of fluid whey protein emulsions after heating at high whey protein concentrations
2. Preparation of texturized (gelled) whey protein emulsions at low whey protein concentrations

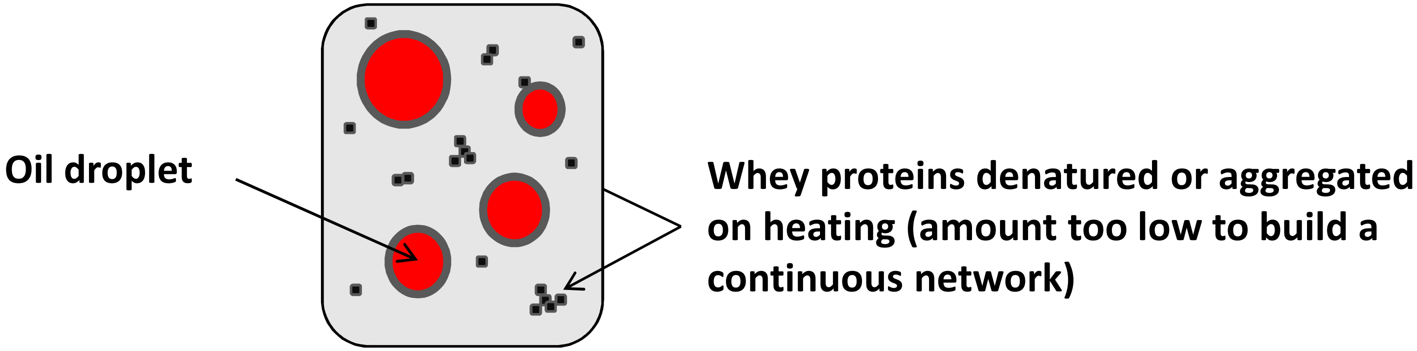
Conclusion

Acknowledgments

2. Design of texturized whey protein emulsions

♦ At **high protein concentration** → emulsions of native whey proteins gelled on heating

♦ At **low protein concentration** → not enough whey proteins to form a continuous network



Objective : designing texturized emulsions at protein concentration lower than 4%

2. Design of texturized whey protein emulsions

a. Literature review

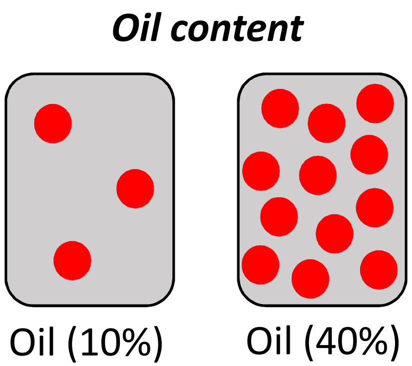
- Protein aggregates are able to connect adjacent oil droplets

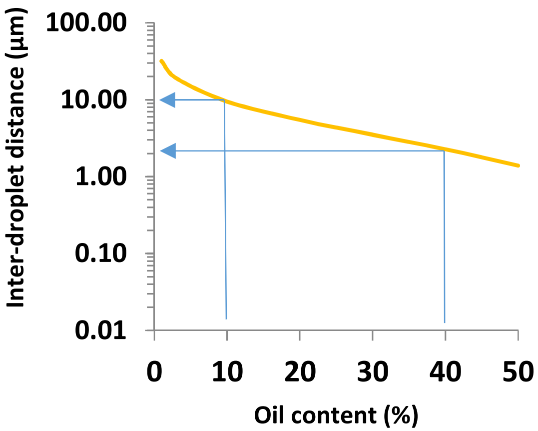
Surel et al., 2014

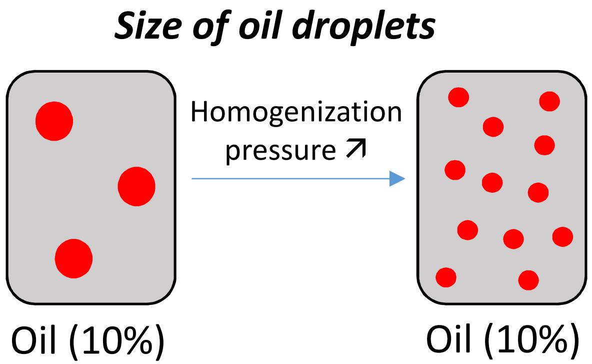
→ Distance between oil droplets is of tremendous importance … (the size of the aggregates also!)

- Distances between oil droplets depend on **oil content** and the **size of the oil droplets**

Jost, 2006; Dickinson, 2003; Jafari, 2008; Dickinson, 2010



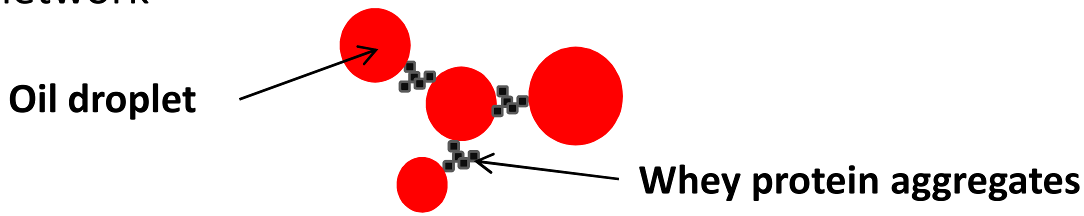


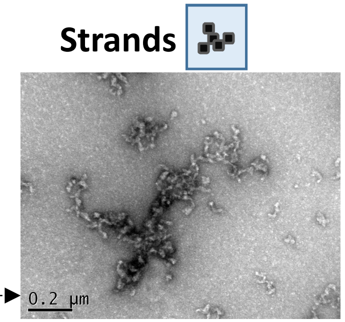


2. Design of texturized whey protein emulsions
b. Strategy

Connect the oil droplets in the emulsion in order to obtain a **continuous network** (gel) **at low protein concentrations** without non-dairy gelling agents

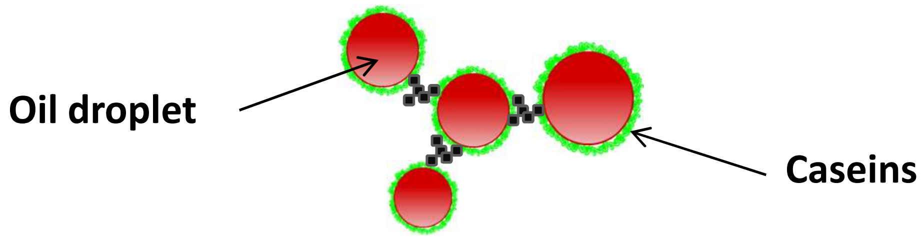
- Using **whey protein aggregates** to connect the oil droplets for obtaining à continuous network





For structuring whey protein emulsions at low protein concentrations the aggregates have to be of low density → **whey protein strands**

- Control of the size (distance between oil droplets) of the oil droplets by using caseins



Casein amount < critical casein concentration in order to allow the **adsorption of whey protein strands at oil droplet interface**

2. Design of texturized whey protein emulsions

c. Hypothesis to test

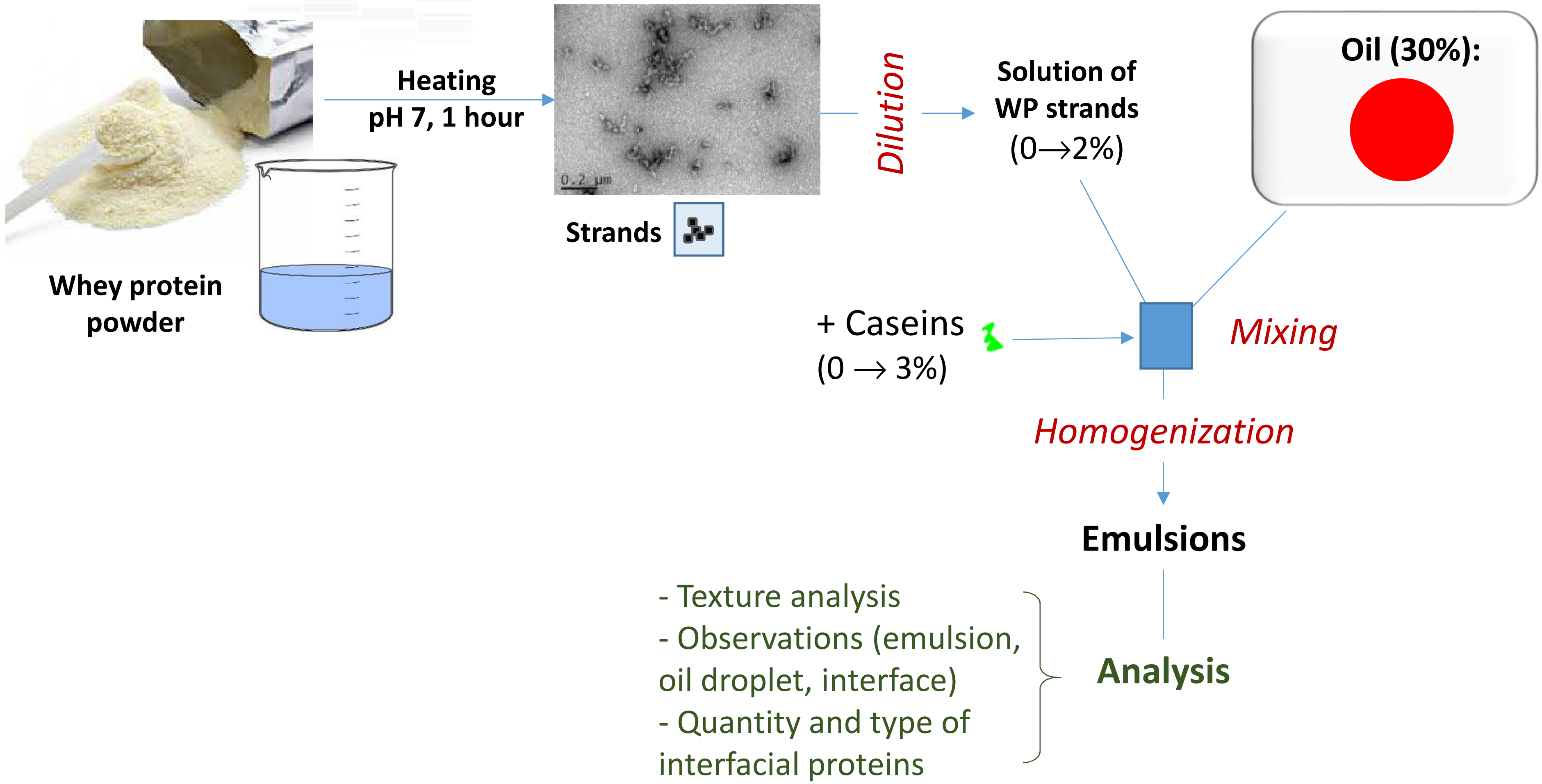
♦ The **whey protein strands** can **connect the oil droplets** in order to obtain a continuous network (as whey protein microgels can do below the critical casein concentration, *see section 1*.)

♦ The **whey protein emulsions will be texturized** (gelled) only if the whey protein strands are adsorbed at the oil droplet surface (**below the critical casein concentration**)

♦ **Above the critical casein concentration** the whey protein strands are discharged in the continuous phase of the emulsions and are not able to connect oil droplets → **fluid emulsions**

2. Design of texturized whey protein emulsions

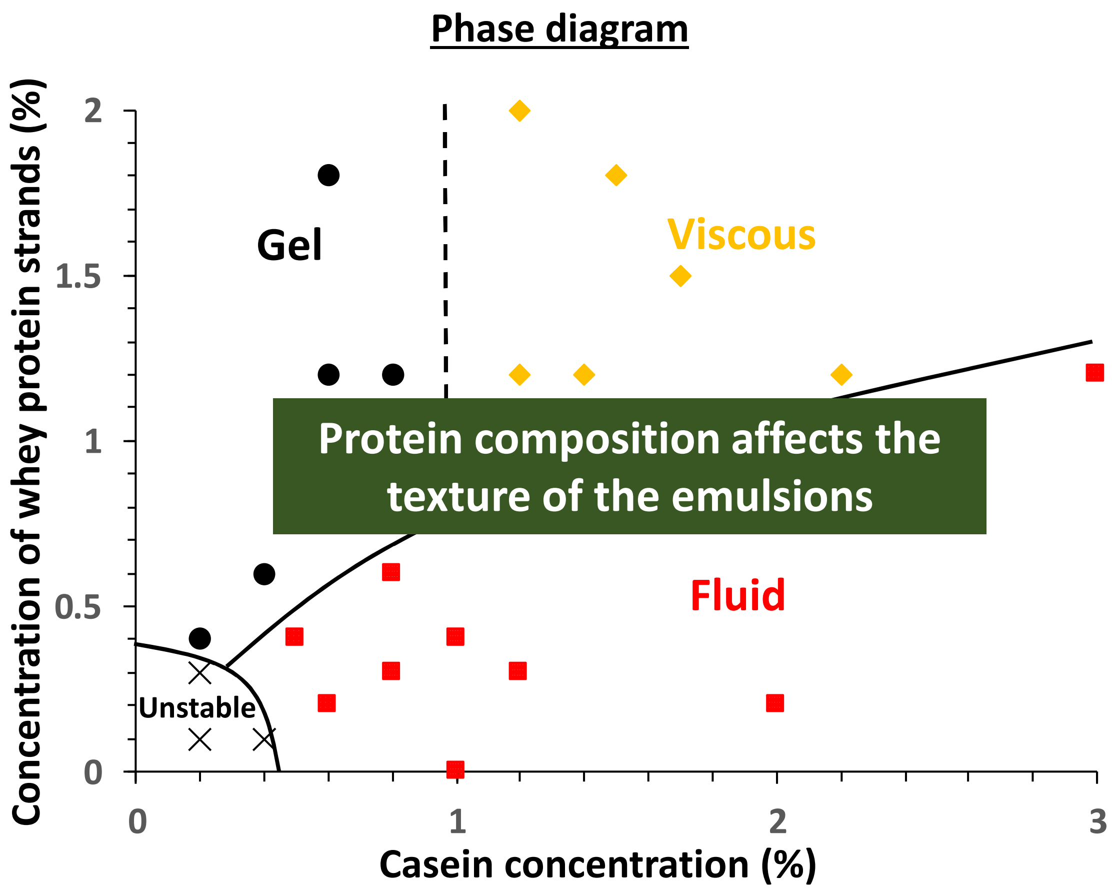
d. Materials and methods



2. Design of texturized whey protein emulsions

e. Results

Screening of the emulsion texture according to protein composition in the emulsions



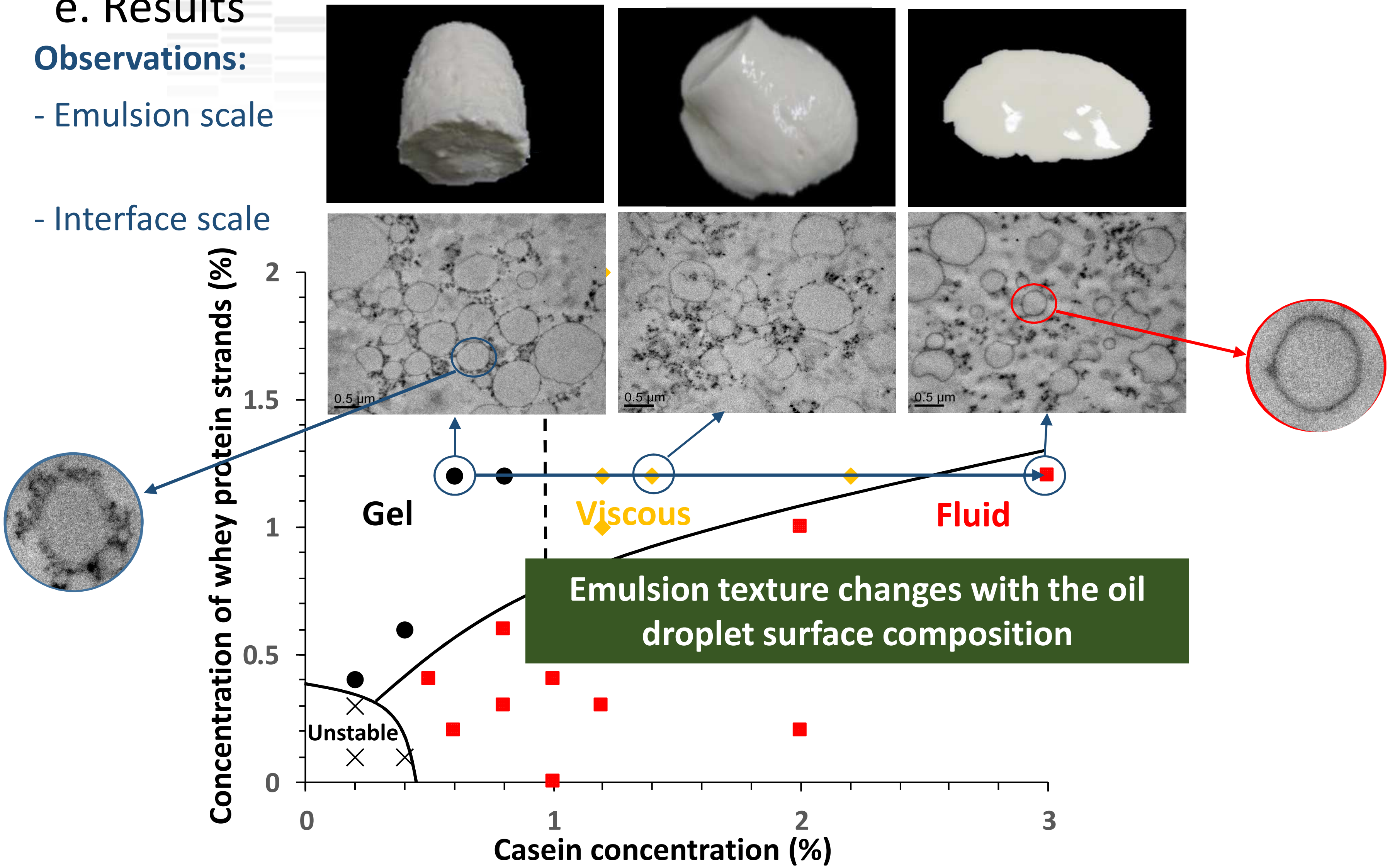
2. Design of texturized whey protein emulsions

e. Results

Observations:

- Emulsion scale

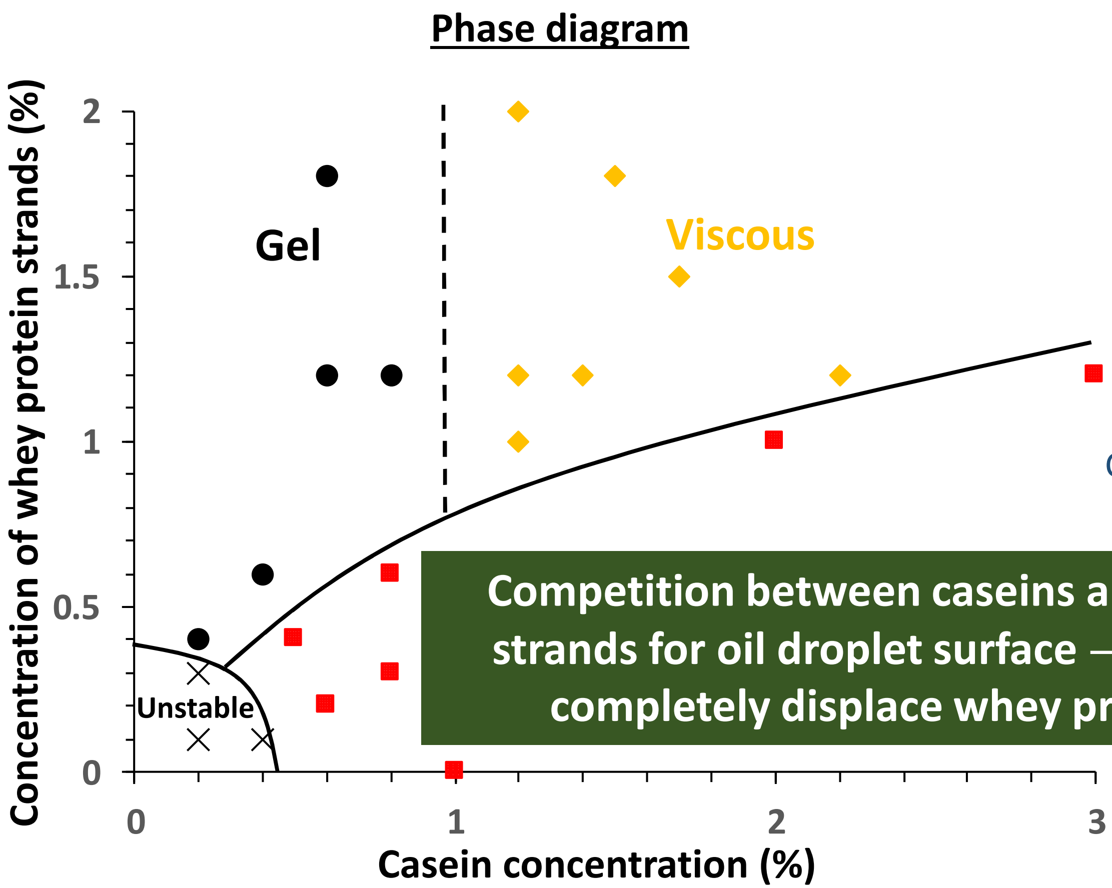
- Interface scale

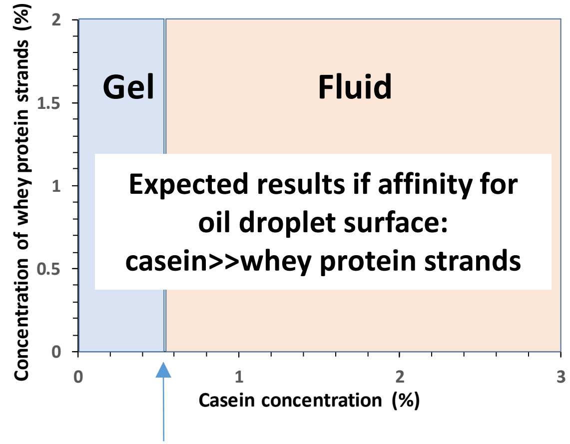


2. Design of texturized whey protein emulsions

e. Results

Results of emulsion texture were not obvious regarding the **critical casein concentration**



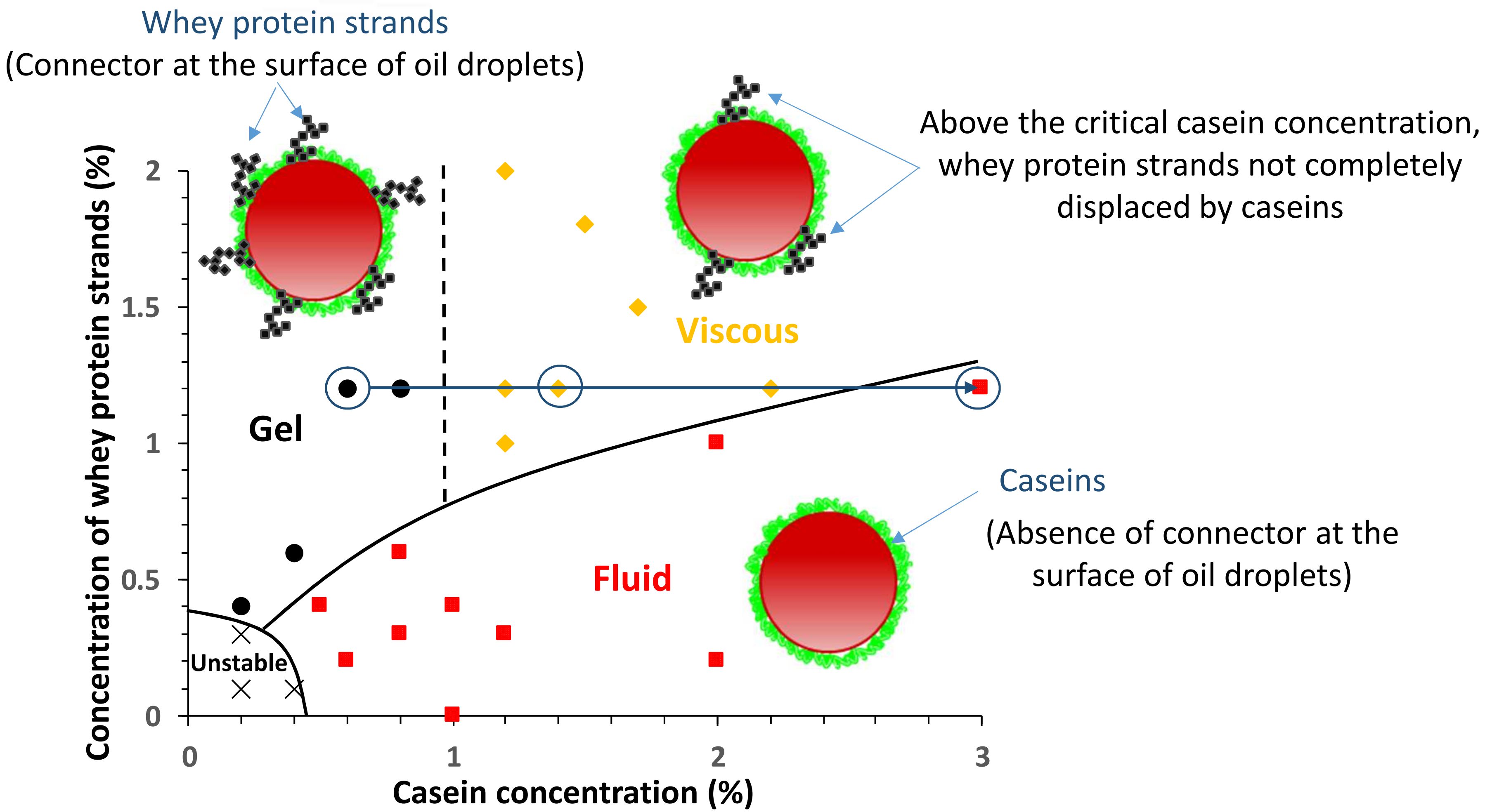


Competition between caseins and whey protein strands for oil droplet surface → caseins do not completely displace whey protein strands

2. Design of texturized whey protein emulsions

e. Results

Structure of the oil droplet surface (schematic representation)



2. Design of texturized whey protein emulsions

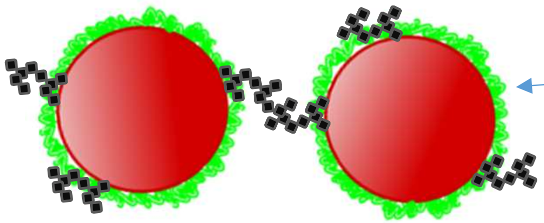
- How to obtain texturized (gelled) whey protein emulsions at low protein concentrations (below 4%) without non-dairy additives ?

- Select aggregates of low density (whey protein strands)

- Use whey protein strands as « connector » at the surface of the oil droplets (size of the aggregates ~ distance between oil droplets)

- In combination with the homogenizing pressure, use caseins as emulsifiers to control the size of the oil droplets (distance between oil droplets)

Whey protein strands (= connecting agent)



Caseins (with the homogenizing pressure, they control the oil droplet size and the distance between oil droplets)

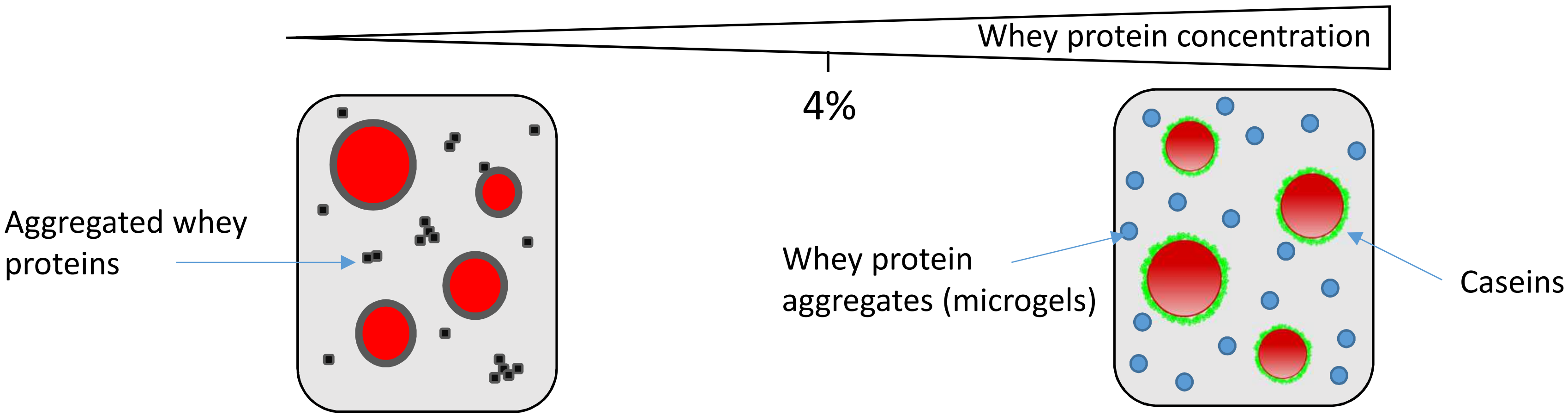
« connected » oil droplets

The number of « connectors » defines the texture of the emulsions (gel, viscous solution, fluid)

Conclusion

To design fluid heat stable emulsions:

- ♦ At low protein concentrations → **native whey proteins**
- ♦ At high protein concentrations → whey protein aggregates (**microgels**)
  + **casein > critical concentration** to cover the oil droplet surface



Whey protein concentration is too low to obtain a continuous network

Oil droplets are covered with caseins and whey proteins are structured in microgels (dense aggregates) in order to prevent the gelation of the continuous phase and the interaction with the oil droplet surface

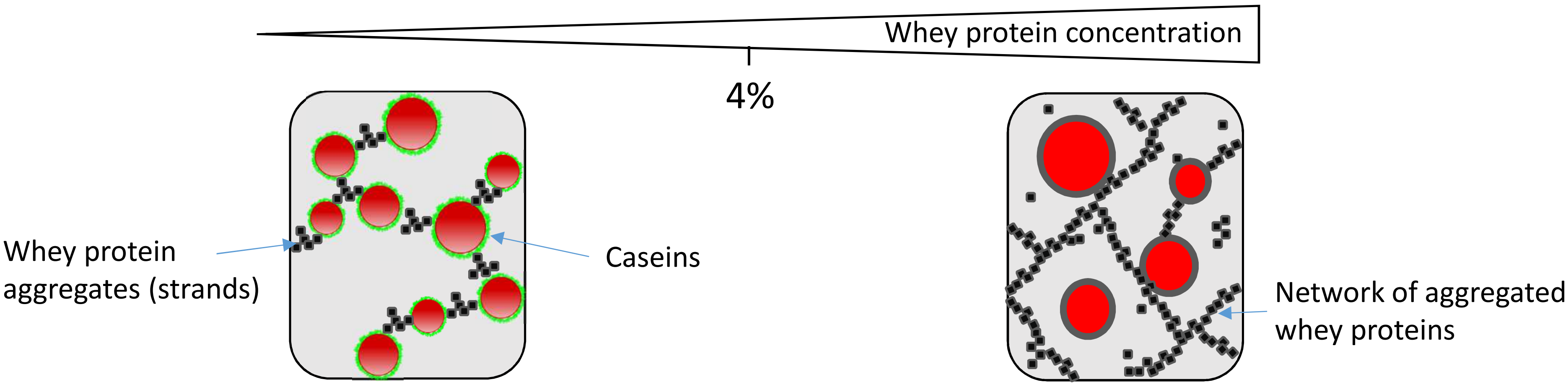
Conclusion

To design texturized (gelled) emulsions:

- ♦ At low protein concentration → whey protein aggregates (**strands**)

  + **casein < critical concentration** to cover the oil droplet surface

- ♦ At high protein concentration → **native whey proteins**



Use caseins as emulsifying agent and low density whey protein aggregates to connect the oil droplets

Native whey proteins gelled on heating

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