On improving the stability of binary protein-stabilised emulsions

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Introduction

- Binary dairy/plant protein blends are gaining interest due to their functionalities, nutritional aspects, sustainability, and masking off-flavour of plant protein.
- Polysaccharide incorporation to protein-stabilised emulsions can improve the stability of emulsions.
- Legume protein-polysaccharide complexes possess unique structures in food colloids, while protein hydrolysates, or soluble fractions of proteins were used in most cases.
- Employing entire legume proteins for emulsion stabilisation would expand the use of plant protein.
- Whey and pea protein were selected as representative dairy and plant proteins and sodium alginate was applied as a model polysaccharide.
- Binary whey/pea protein-stabilised emulsions were produced with/without sodium alginate at pH 6.6, 8.0, and 11.0.
- This research provides a potential strategy to address the limited emulsifying properties of pea protein.

Aims and objectives

- This study aims **to improve the stability of binary dairy/plant protein-stabilised emulsions by adding polysaccharide** and **to understand the effect of pH in promoting protein adsorption at the interface.**
- We hypothesised that by adding polysaccharide the protein adsorption can be improved and both proteins could coexist at the interface.

Evaluation

- **Physical stability**
  The creaming index was calculated from the percentage of the height of the cream layer over the height of the total emulsion sample at storage day 0, 1, 7, 14, 21.
- **Droplet surface charge and size distribution**
  The droplet charge and size distribution of the samples were evaluated using a Malvern Zetasizer Nano and a Mastersizer 3000.
- **Interfacial properties**
  The cream phase and clear serum phase were obtained by centrifugation at 14,000 × g for 90 min. The serum phase was determined by the Lowry method and the interfacial protein composition of the cream phase was determined by SDS-PAGE.
- **Microstructural elucidation**
  Confocal laser scanning microscopy was carried out using a Zeiss LSM 800. The water phase and the oil phase were dyed with Nile blue A and Nile red.

Sample preparation

Pea protein pre-treatment
↓
Pea protein + Whey protein
↓
Binary protein (BP) + Sodium alginate (SA)

pH control to pH 7	**pH control to pH 8, 11**
↓ **Canola oil addition**	↓ **Canola oil addition**
↓ **High-shear mixer (7,000 rpm, 5 min)**	↓ **High-shear mixer (7,000 rpm, 5 min)**
↓ **High-pressure homogenisation (15 MPa, 3 passes)**	↓ **High-pressure homogenisation (15 MPa, 3 passes)**
↓ **pH control with HCl to pH 6.6**	

- **Concentration**
: 2% binary whey/pea protein (1:1, w/w), 0.4% sodium alginate, 20% canola oil.

Results

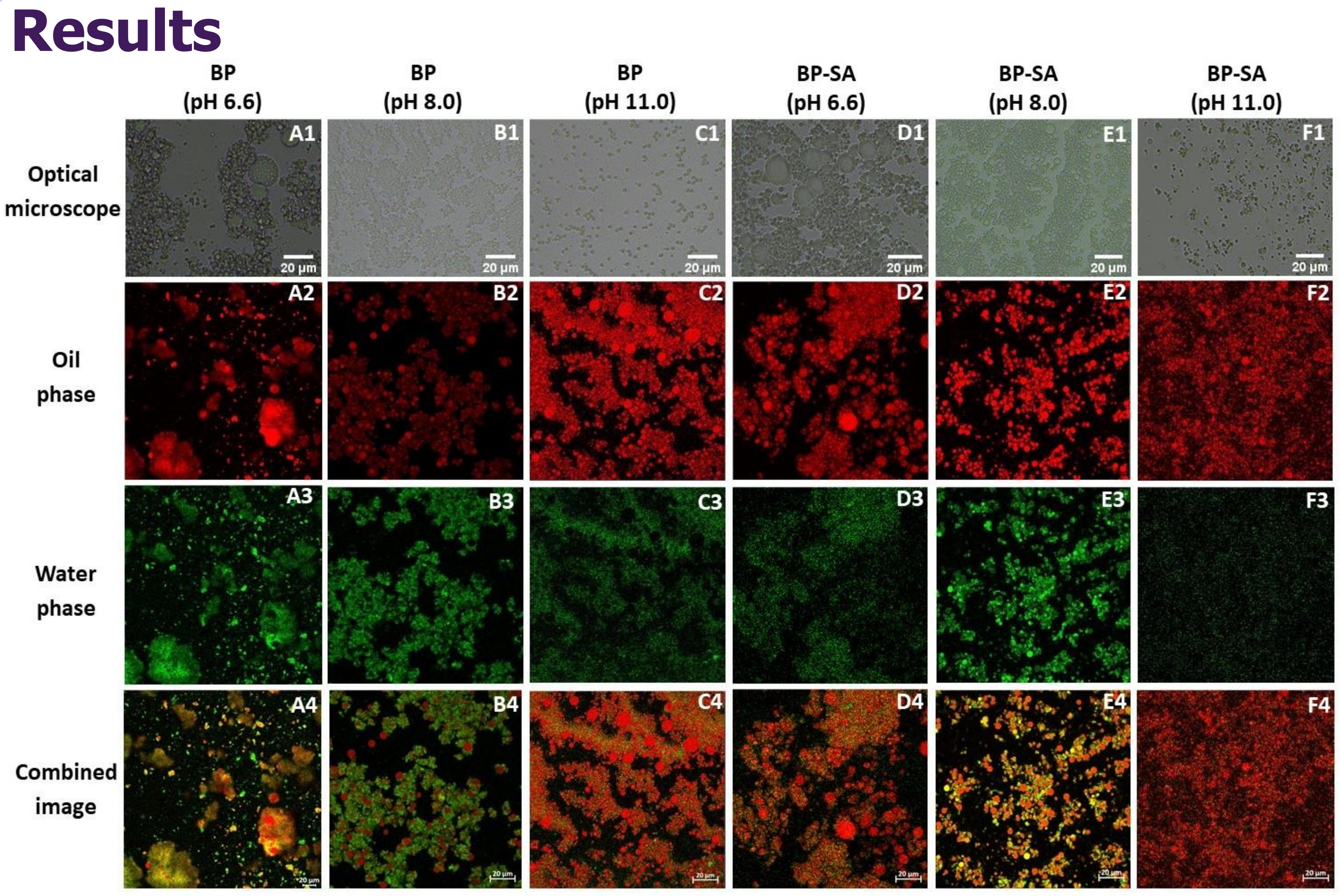

Figure 1. Microstructural images of BP_pH6.6 (A); BP_pH8.0 (B); BP_pH11.0 (C); BP-SA_pH6.6 (D); BP-SA_pH8.0 (E); BP-SA_pH11.0 (F) using optical microscope 40× objective (1) and confocal microscope with 20× and 40× objectives stained with Nile blue A (protein in the water phase) and Nile red (oil phase). (2) water phase, (3) oil phase, and (4) combined image of oil and water phase. The scale bars in (1) and (4) represent 20 μm.
BP: binary protein-stabilised emulsions; BP-SA binary protein-stabilised emulsions with sodium alginate.

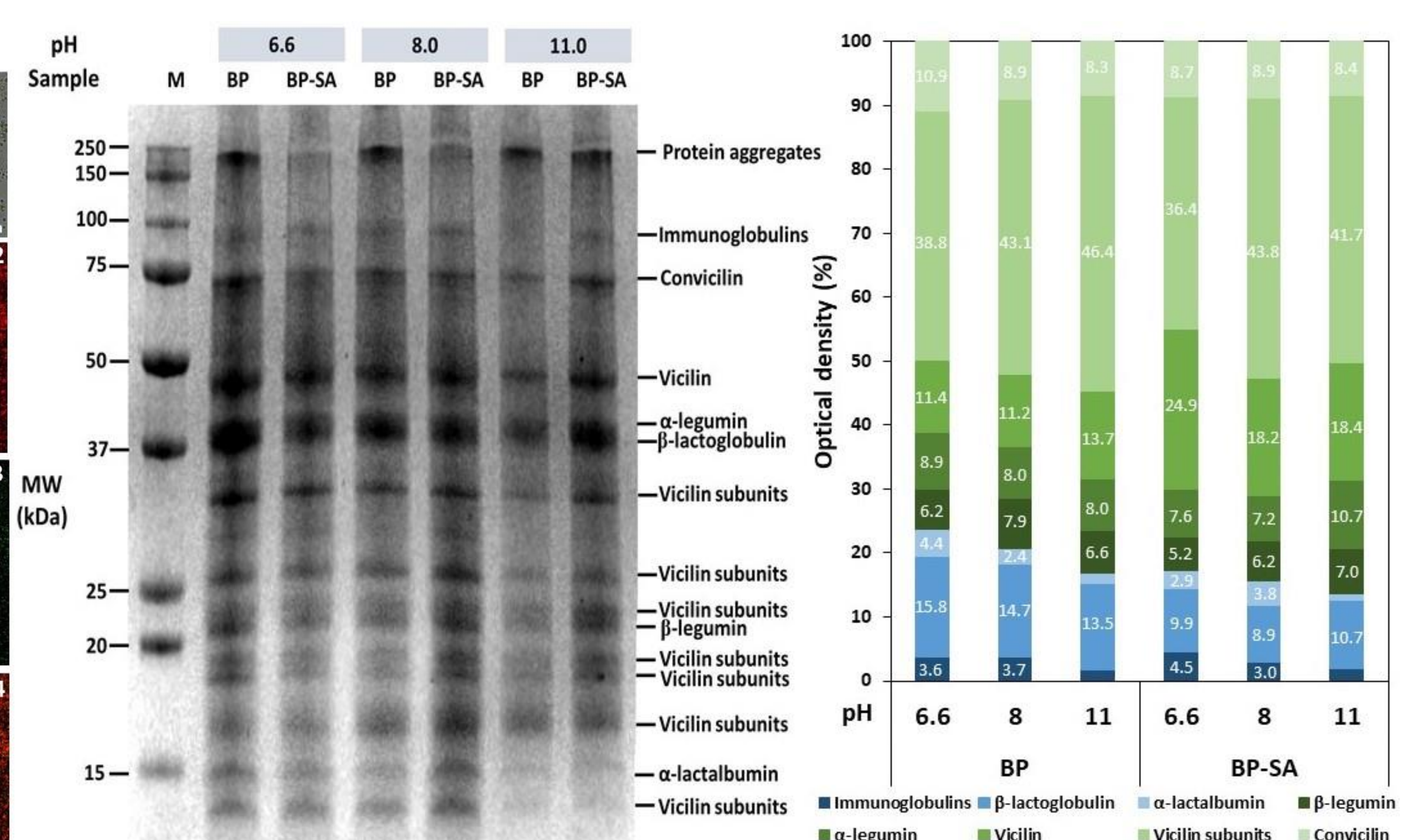

Figure 2. Interfacial composition of binary protein-stabilised emulsions with/without sodium alginate by SDS-PAGE under reducing conditions.

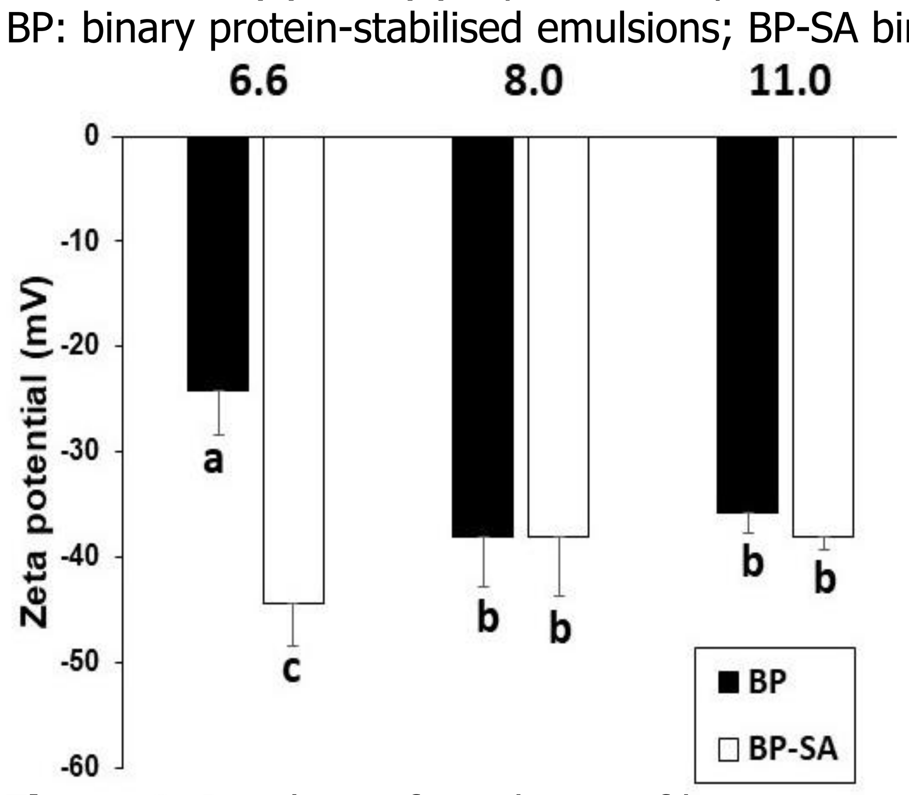

Figure 3. Droplet surface charge of binary protein-stabilised emulsions with/without sodium alginate.

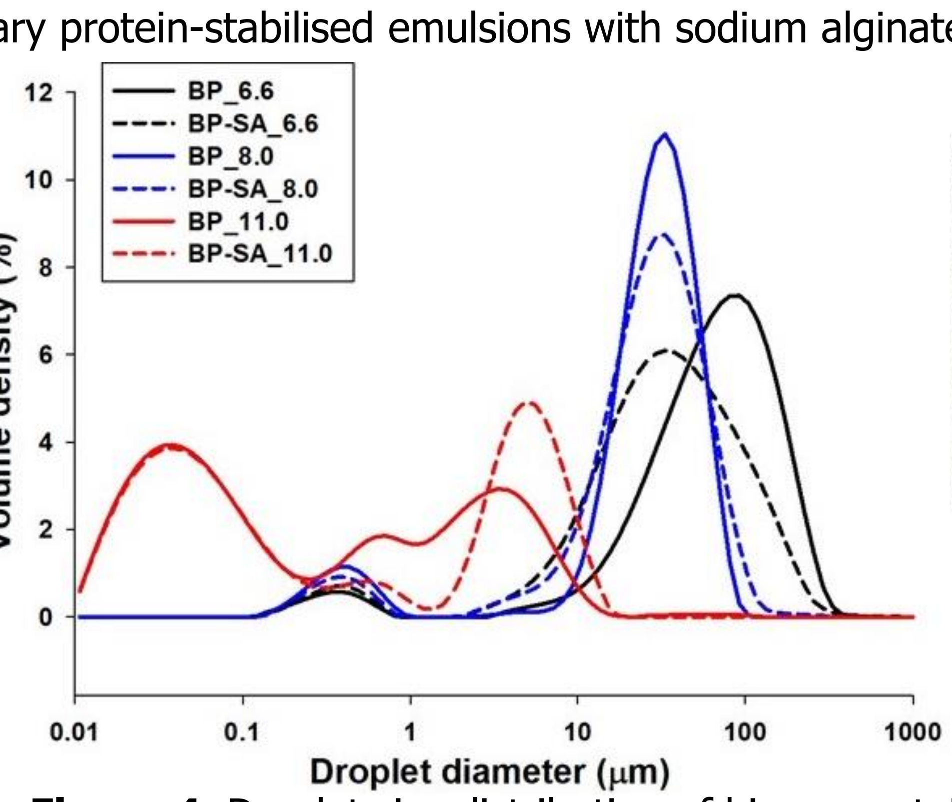

Figure 4. Droplet size distribution of binary protein-stabilised emulsions with/without sodium alginate.

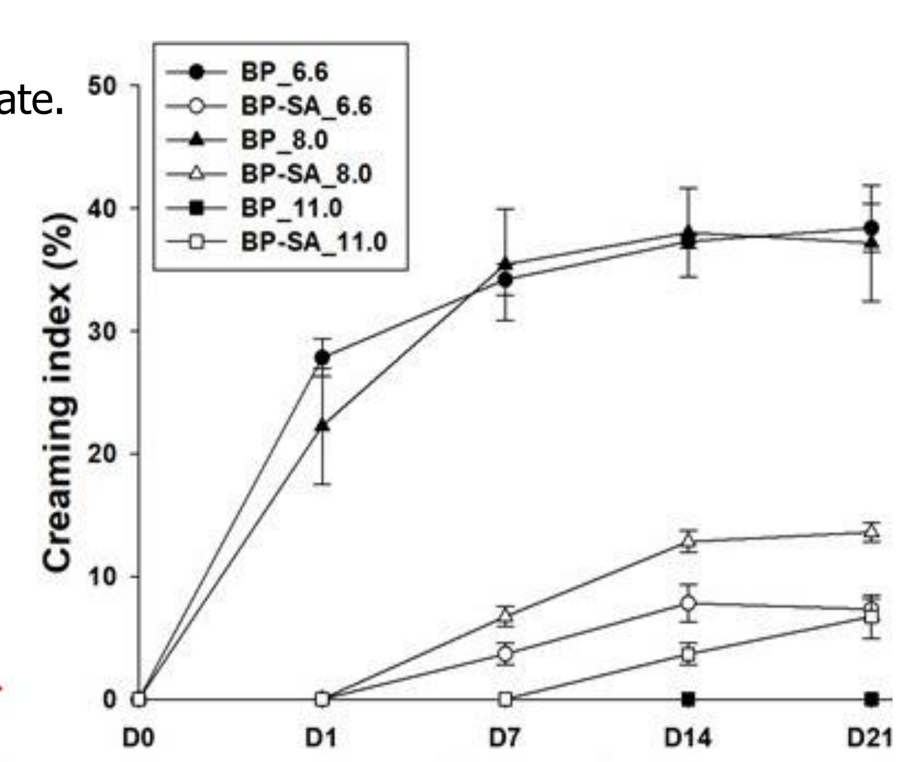

Figure 5. Creaming index of binary protein-stabilised emulsions with/without sodium alginate during storage.

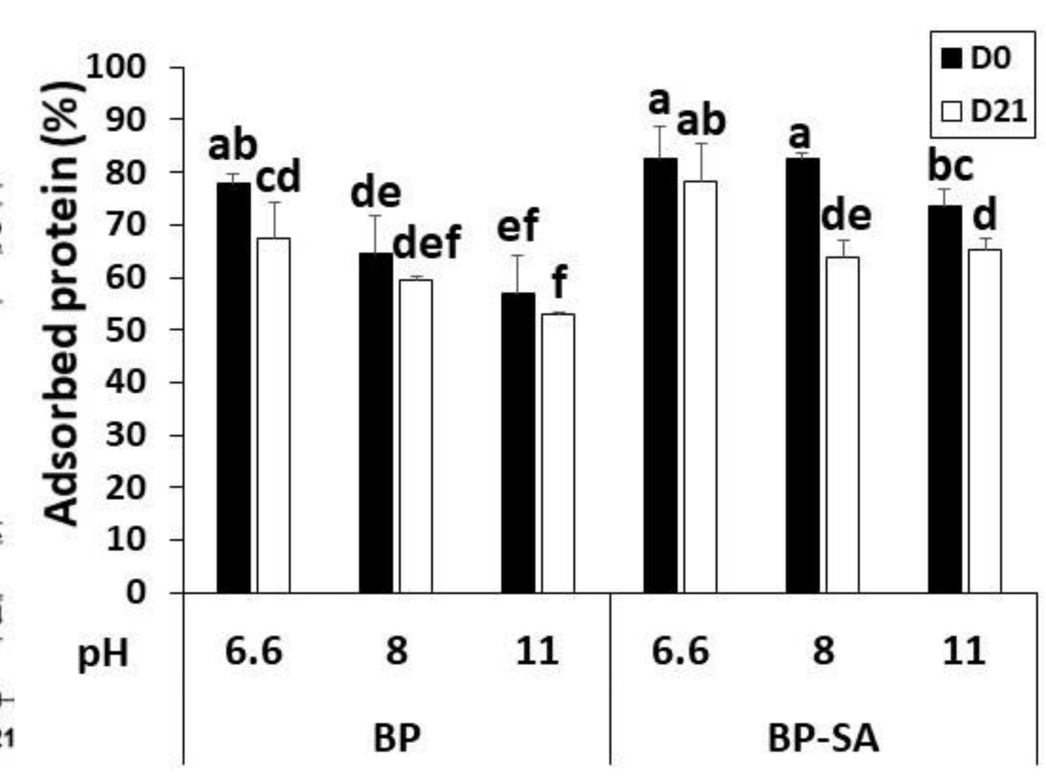

Figure 6. Interfacial protein composition of binary protein-stabilised emulsions with/without sodium alginate.

Conclusion

- At pH 6.6, **alginate addition prevents pea protein from particle formation** by forming the electrostatic complex.
- **Alginate addition** can significantly ($p < 0.05$) **reduce the droplet size at pH 6.6** (from 67.13 μm to 33.84 μm)
- The BP is most stable at pH 11.0, whereas alginate addition can enhance emulsion stability at pH 6.6 and 8.0.
- **An emulsion with small droplets (1-3 μm)** can be formed regardless of alginate addition **at pH 11.0**.
- Alginate addition can enhance protein adsorption at the interface, and the amount of protein adsorbed decreases with increasing pH.
- **Proteins remained attached at the interface at pH 6.6** on D21, while adsorbed protein promoted by alginate addition were displaced at pH 8.0 and 11.0.
- Both whey and pea protein are present at the interface, although **pea protein is dominant**, and **alginate addition promoted the adsorption of vicilin**.
- This demonstrates the beneficial effects of using dairy/plant protein blends in terms of functionality.

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