Crosslinking of fava bean protein and pectin: Effect of pH, concentration and temperature

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Medway Food Innovation Centre

Product development kitchen

Food processing & analysis lab

Algae Biotechnology

Under-utilized legume

Fava bean

Lupin

Improving key functionalities using low-emission technologies

RuBisCo

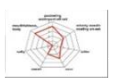
Duckweed

Improved extraction & assess bioavailability

Red clover

Novel extraction & texturization

Microalgae



ValgOrise: Algae for taste

PROFILE: Off-flavour masking

Imperial College London

Upcycled plant proteins

IPSUS: Upcycled plant proteins for meat and cheese alternatives

RESEARCH GRANTS (2020-)

£ 1.5M

Develop **user-inspired research** program

FOOD ACCELERATOR

100+ Companies

Offer state-of-the-art **technical facilities**

TOTAL INVESTMENT

£ 2.1M

Bridge the gap between **science and application**

MARKET TO REACH

£ 1B

Provide training to build **skilled workforce** for industry

TY OF VICH

Natural Resources Institute

defra Department for Environment Food and Rural Affairs

GCRF Global Challenges Research Fund

European Commission | Horizon 2020 European Union funding for Research & Innovation

STFC Food Network+

UKRI UK Research and Innovation

SUSFOOD2

eat it up

STARBUCKS

Study background

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Study background

- Growing demand for plant protein from different sources

- However, there is low consumption of Fava bean protein isolate (FBPI) due to its lack of heat stability, protein delivery, and poor texture; these factors can be improved by crosslinking with pectin (Khan et al., 2015).

- Pectin can stabilize emulsions both below and above the isoelectric point of the protein (Qu et al., 2018).

Image source: Hitchcock Farms

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Study background…

- Protein functionality is greatly affected by cross-linking, which improves the emulsifying, gelling, heat stability, foaming and thermo-rheological properties (Buchert et al., 2010).

- Cross-linking or chemical crosslinking is a chemical process in which a protein and a polysaccharide are bonded through covalent bonds, which improves their properties.

- Covalent bonds between pectin and plant protein may be formed by a chemical reaction generally referred to as conjugation; a glycosylation/glycation process, based on a Maillard-type reaction (Sedaghat Doost et al., 2019; Einhorn-Stoll et al., 2021).

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Research question

- Does pH, concentration and temperature conditions show any effect on cross-linking of faba bean protein isolate and pectin?

Research approach

Methodology

- The mixture of FBPI and pectin were made into a solution of ratios 1:0.1 (10%), 1:0.05 (5%), and 1:0.03 (3%).
- The solutions were stirred at 1000 rpm for 45 min using VELP Overhead Stirrer.
- With a total biopolymer concentration of 2% (w/v), the pH was adjusted to pH 5 , 7 and 9 using 2 M NaCl and 2 M HCl.

- Degree of graft
- Emulsifying properties

Ma et al. (2020)

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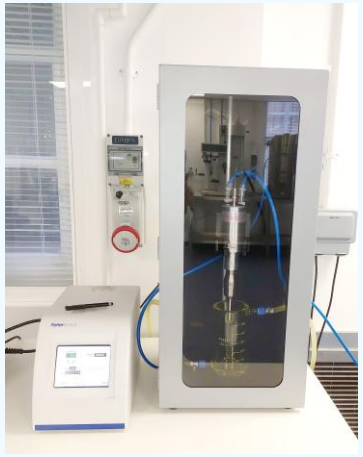
Treatments

Stirring and homogenising



Ultrasound Treatment

Heat Treatment

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Degree of graft

Degree of graft

- Degree of graft (DG) determined by modified OPA assay method
- Absorbance at 320 nm

$$DG = \frac{A_0 - A_t}{A_0} \times 100\%$$

- Different treatments
  - Ultrasound
  - Heat treatment
  - No treatment

- *A*0 = free amino group content of the mixtures of FBPI and pectin; *At* = free amino group content of FBPI-pectin conjugates prepared with ultrasound or Heat treatment.

Vigo et al. (1992); Ma et al. (2020)

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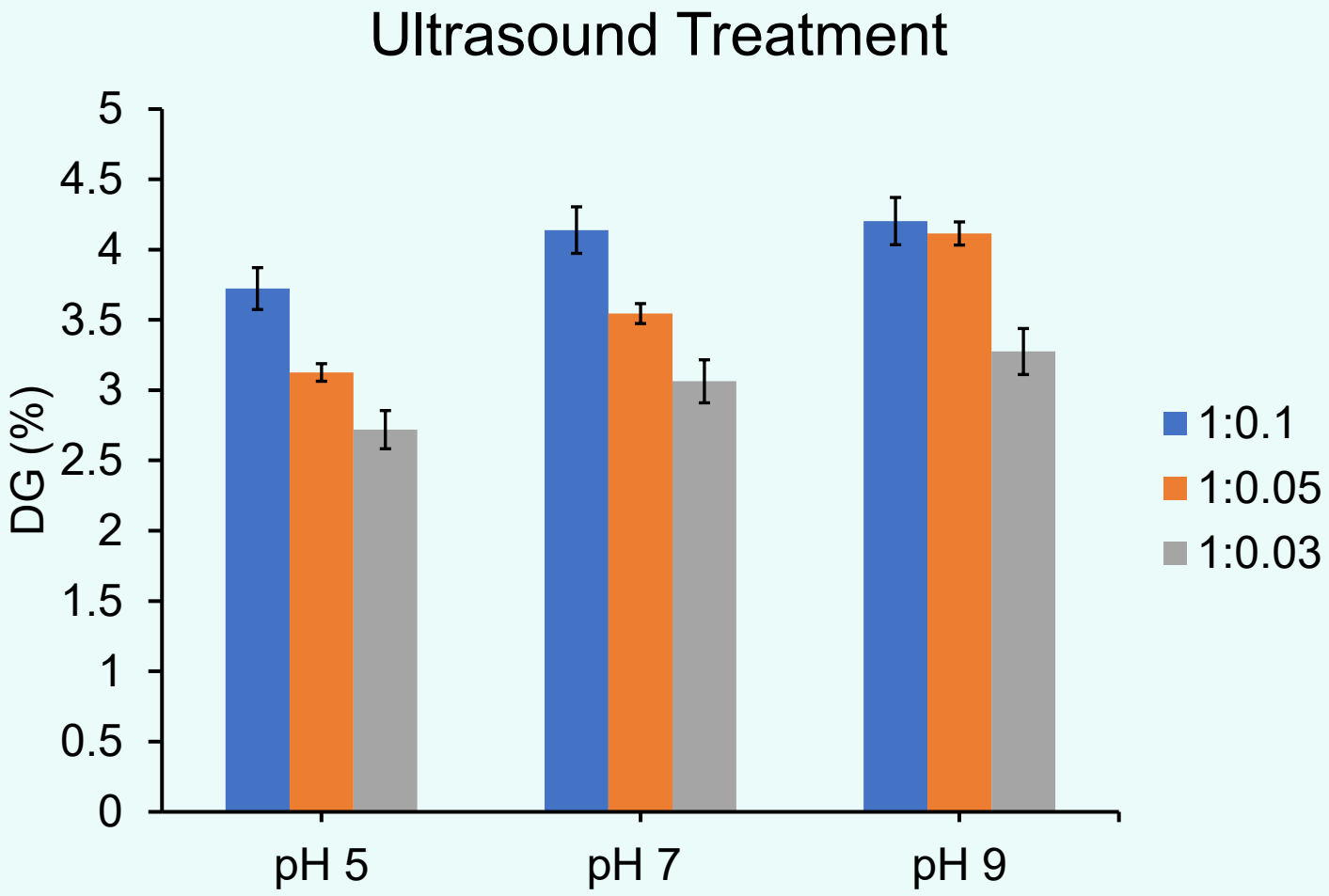
Degree of graft



- 10% pectin showed the greatest effect on DG
- Increase across pH due to formation of Maillard reaction from US treatment.

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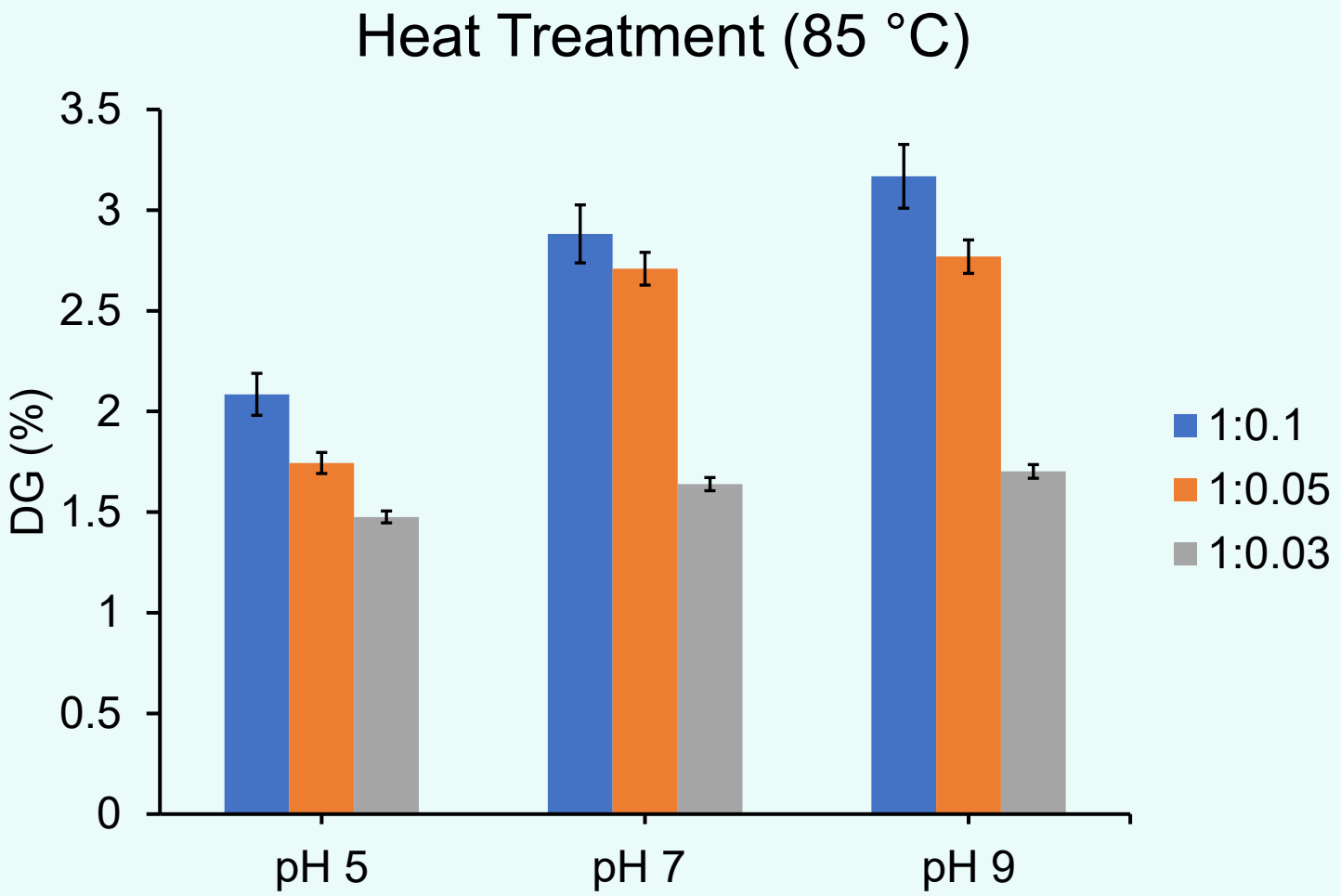

- The DG was less in heat treatment
- This could be due to temperature of heating which might have denatured the proteins.

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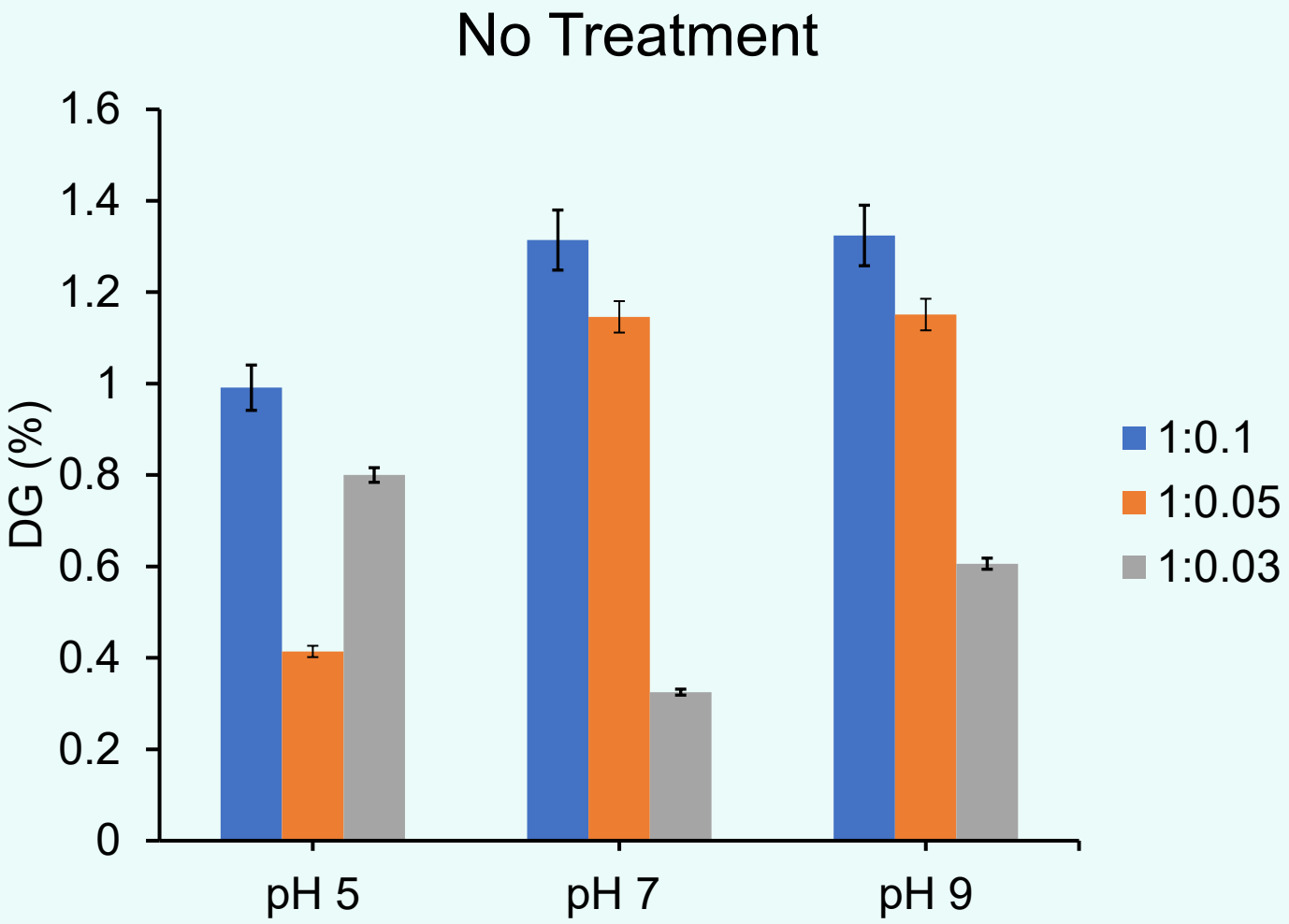

- Reduced levels of DG in conjugates without treatment
- A lack of treatment might have led to inability of Maillard reaction taking place

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Emulsifying properties

Emulsifying Properties

- Emulsifying activity index (EAI)

Absorbance at 500 nm

$$EAI\left(m^2/g\right) = \frac{2 \times 2.303 \times A_0 \times DF}{10000 \times \theta \times L \times C}$$

- DF = dilution factor (100); C = protein concentration (g/ml); L = optical path (1 cm); θ = oil volume fraction (0.25); A_0 = absorbance at 0 min; and A_{10} = absorbance at 10 min.

(Li et al., 2020)

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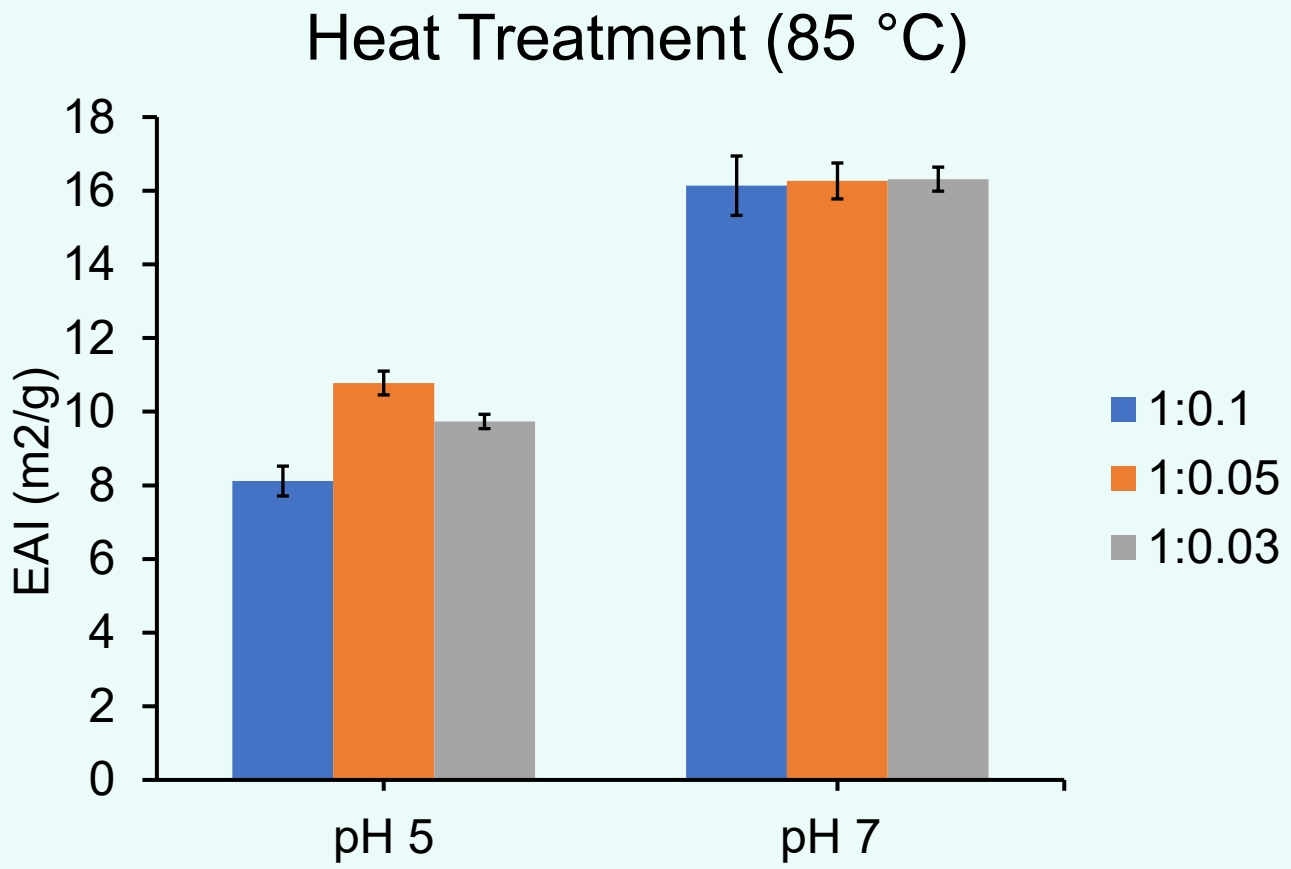
Emulsifying Activity Index (EAI)



- The EAI increased with increase in pH across concentrations
- Combined properties of FBPI and pectin led to an increased emulsion activity

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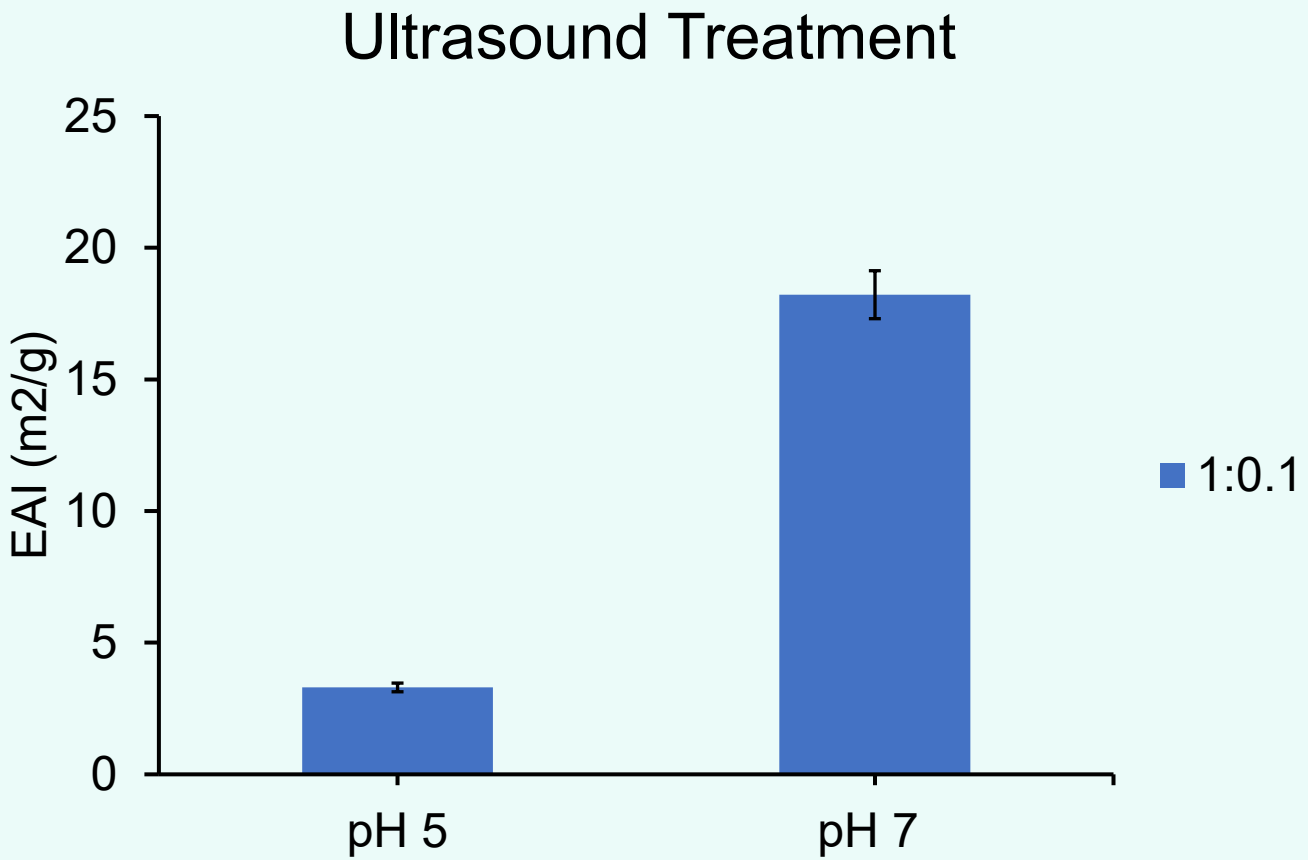

- Similar observation for US treatment.

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Emulsifying Stability Index

- Emulsifying stability index (ESI)

$$ESI\,(min) = \frac{A_0}{A_0 - A_{10}} \times (T_{10} - T_0)$$

- A_0 is the absorbance at 0 min, and A_{10} is the absorbance at 10 min. T_0 represents 0 min, and T_{10} represents 10 min (Li et al., 2020).

- Absorbance at 500 nm

(Li et al., 2020)

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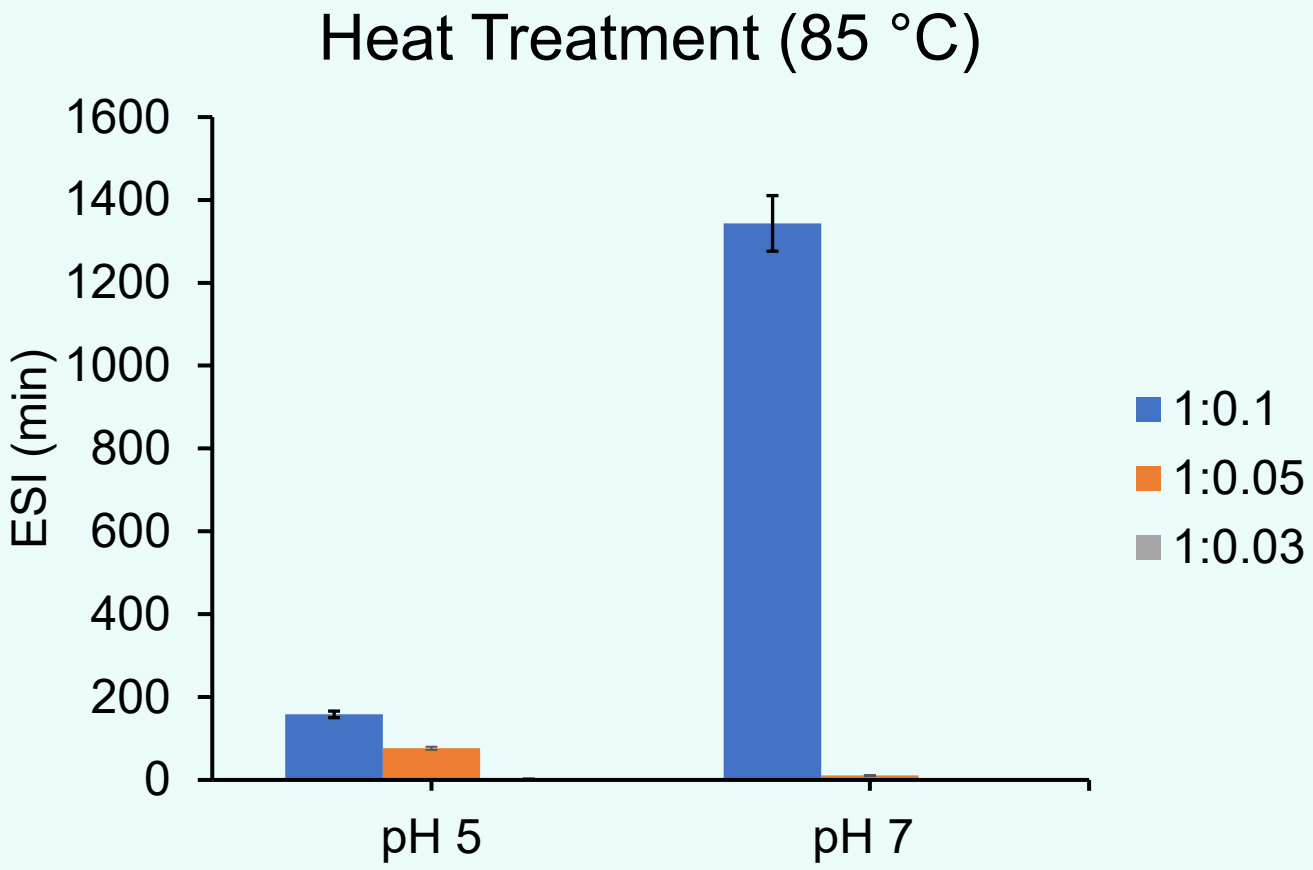
Emulsifying Stability Index



- Emulsion stability higher at pH 7 under wet heat conditions
- Crosslinking between the protein and polysaccharide led to more stable emulsion

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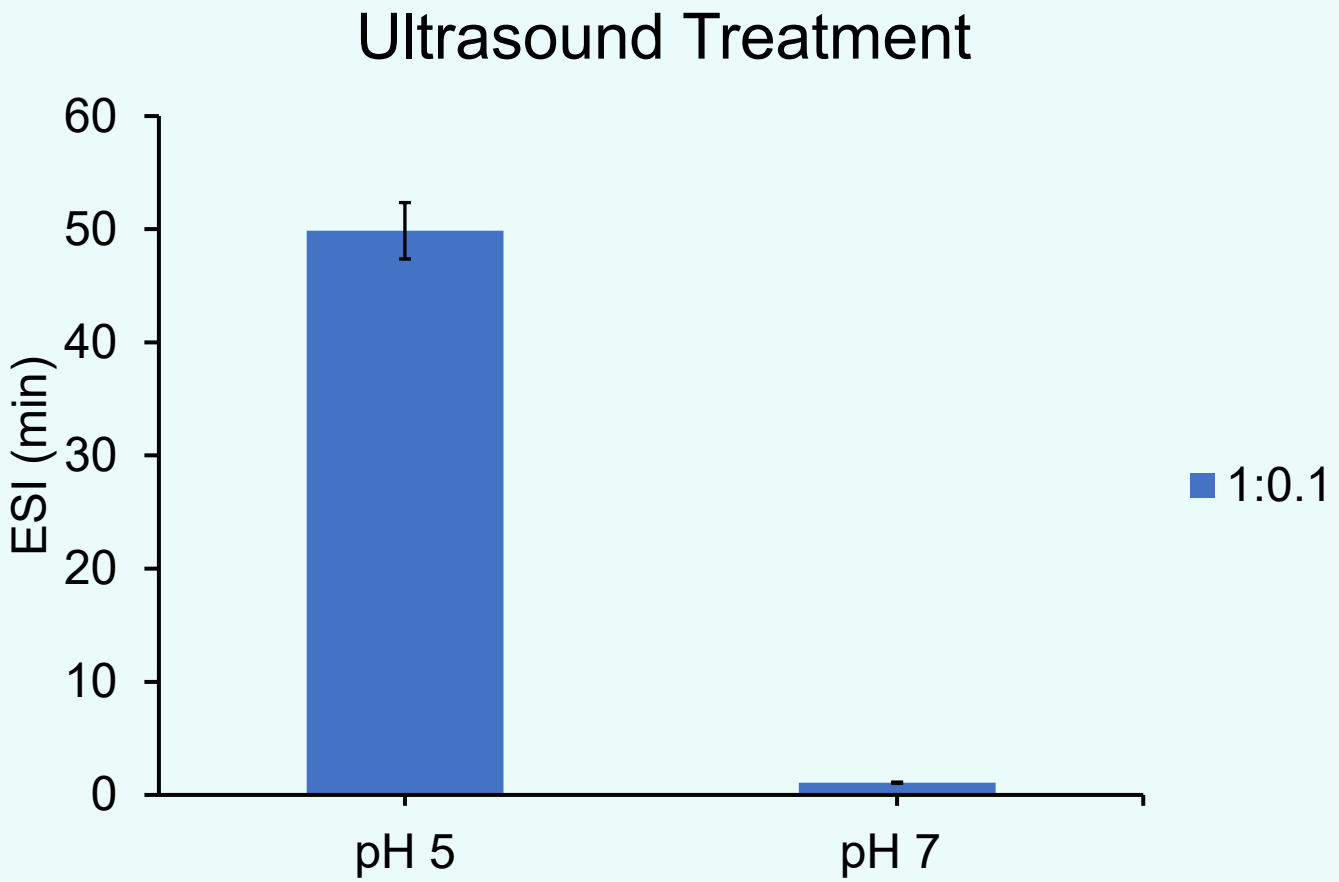

- Emulsion stability decreased using US
- Emulsion was more stable at pH 5 and less stable at pH 7

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Conclusion

- ❑ Ongoing studies on rheology, particle size and SDS-PAGE of conjugates
- ❑ Combined properties of conjugates leads to formation of more stable emulsion
- ❑ Emulsions display more stability under wet heat treatment than US
- ❑ Outcome important in the application, stability and functioning of protein-based products

Acknowledgement

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Thank you

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