PhD course (1.1 ECTS + 0.9 ECTS optional assignment): November, 2018 (week 46) Location: WICC, the Netherlands Draft Programme rev. 24 Sept, 2018/PW

2018: 1.1 ECTS (Mon 9:00 – Thu 15:00) (+ 0.9 ECTS for report)

		Monday Nov 12	**Tuesday Nov 13**	**Wednesday Nov 14**	**Thursday Nov 15**
Preparation of presentation and reading of course literature		**Proteins: Sourecce, structures and functionality**	**Physical changes and techno-functionality**	**Chemical changes and analytical methods**	**Enzymatic hydrolysis and digestibility**
	9:00- 10:00	**Welcome** and introduction to the course / *Peter Wierenga* **Food proteins:** An introduction to food proteins, their sources and structures / *Peter Wierenga*	**Protein denaturation, gelation and how to evaluate it** /*Richard Ipsen*	**Enzymatic and chemical oxidation of proteins** *Willem van Berkel*	**In vivo perspective on hydrolysis (human)** Evan Abrahamse
	10:00-10:45	**Analysis of proteins** *Peter Wierenga*	***Assessing foam and emulsion properties of food proteins*** / Roy Delahaije/ *Peter Wierenga*	**Maillard reaction in proteins** *J. Otte*	**Enzymatic hydrolysis of food proteins** / *Peter Wierenga*
	10:45-11:00	*Coffee break*	*Coffee break*	*Coffee break*	*Coffee break*
	11.00-11.45	**Milk proteins:** structure and (food) functionality / *Richard Ipsen*	**How functionality is affected by protein aggregation /** *Richard Ipsen*	**Meat proteins** ... Rene Lametch	**Effects of systems conditions/modifications** on protein hydrolysis *Yuxi Deng/Peter wierenga*
	12:00-13:00	*Lunch break*	*Lunch break*	*Lunch break*	*Lunch break*
	13:00-14:45	**Legume proteins** – isolation / structure and functionality / *Peter Wierenga*	**Effects of protein modifications/plant proteins on foam and emulsion properties /** *Peter Wierenga*	***Proteomics*** / *Rene Lametch*	**Bioactive peptides from food proteins** *Jeanette Otte*
	14:00-14:45	Presentations of participants / *Students*	**Combinations of milk proteins and vegetable proteins /** *Richard Ipsen*	Students presentations: 5 students	Introduction to the assignment /P. Wierenga/ *Jeanette Otte* Preparation of outline / *Students and teachers*
	15:00-15:15	*Coffee break*	*Coffee break*	*Coffee break*	*Coffee break*
	15:15-16:00	***Industrial perspective (FrieslandCampina?)***	Students presentations: 5 students	Student presentations: 5 students	**Evaluation of the course** End of course **Writing of report during the next week** **Deadline: December 13**
	16:00-17:00	Student discussions of selected challenges / *Richard and Peter*	Student discussions of selected challenges / *Richard and Peter*	Student discussions of challenges/*Marianne and Peter*	
	17:30-21:00		**Dinner?**		

The aim of the course: The course will provide basic knowledge of selected food proteins and their reactions during processing and influence on structure and functionality. In addition, advanced methods used to reveal the proteins and chemical/structural changes will be presented, and participants challenges discussed?.

Student discussion topics:

Monday: *using more purified materials / using commercial preparations; isolation challenges/analytical challenges*

Tuesday: *Why combinations of plant / dairy proteins are better than the pure systems.*

Wednesday:

- *About protein modifications and suitable methods to detect,*
- *Can we see glycation by SDS-PAGE? What are the advantages and limitations of using intact proteins? Same for the method where hydrolysis is performed first. Perhaps discuss a case from one of the students.*
- *Glycation: Challenges in quantifying extent of glycation /Maillard in industrial samples (mixtures of proteins, no 'blanks'/untreated samples available, possibility that secondary reactions already occurred)*

Monday: Proteins: Sources, Structures and functionality

- ✓ Introduction to protein basic structure / basic chemical properties
- ✓ Introduction Analytical techniques for protein research
- ✓ Specific details on proteins from :
  - **Milk**:
    - composition, major proteins and their typical properties (..and...?)
  - **Legumes**:
    - Isolation, influence /issues with non-protein compounds (PPO)
    -
  - **Meat** (Wednesday)
- ✓ Industrial perspective: Variations in properties

Tuesday: Physical changes and techno-functionality:

- Processing induced changes of proteins (unfolding/aggregation; covalent modifications) and methods to analyse /characterise these changes
  - CD/Fluorescence/DSC
  - OPA/ LC-MS etc (covalent modifications)
  - Light scattering etc (aggregates)
- Methods to analyse foam/emulsions:
  - Existing and new concepts/insights
  - Interfacial properties and links to foam/emulsions
  - Experimental techniques

- Protein aggregation and effects on functionality
- Examples of foam/emulsion properties: Extend to plant proteins, influence of modifications and non-protein compounds
- Combinations of milk proteins and vegetable proteins.

Wednesday: Chemical changes and analysis/or analytical methods

- **Oxidation**: PPO, crosslinking enzymes (oxidative enzymes), transglutaminase
- **Protein glycation** (the Maillard reaction) and methods to detect this / ***J. Otte (prepared with Peter Wierenga/YuxiDeng)***
  1) Occurence of Maillard glycation? Why is it important? E.g. Examples with milk proteins and changes in functionality,
  2) Overview of stages of the Maillard reaction with proteins (glucose, lactose etc), addition, rearrangement (= glycation), further reactions and AGE.
  3) Principles of analysis: How can we analyse this modification? In principle either A) on intact proteins by OPA/TNBS/Ninhydrin or LC-MS, or B) after enzymatic hydrolysis of the proteins, (LC-MS), C) after acid hydrolysis (6M HCl, 23 h, 100 °C) (analysis of CML/furosine, LC-UV or LC-MS)
  4) Examples:
    - Analysis of **intact proteins** - I have examples with glycation of b-Lg detected by LC-MS, I think up to 11 lactose units can be seen on the deconvoluted mass spectrum, and I can explain deconvolution, and they can calculated the expected ions for b-Lg with varying number of lactose units attached
    - Maybe there is also the option to analyse modifications of tryptic peptides? From the glycated proteins, e.g. intermediate between intact proteins and free amino acids (then this should be method type no. 2)
    - Analysis of glycated and modified amino acids by LC-MS after hydrolysis of the proteins. I know that colleagues in Mariannes group have a good method to detect maybe 12 modifications, which they are going to publish. Perhaps I can get some examples from them.

- ✓ Meat proteins (basic/ chemistry/analysis)
- ✓ Proteomics

Thursday:

- *In vivo* protein hydrolysis; human perspective
  - Known concepts
  - Gastric emptying
  - Kinetics/extent of digestion
  - Post-prandial amino acid responses
- *In vitro* protein hydrolysis
  - Known concepts / principles
  - How to analyse peptides (including principles MS annotation*)*
  - Is protein hydrolysis predictable?
- Effects of system conditions: Choice of enzyme, enzyme: substrate ratios, pH/T/I, protein modifications
- Bio-active peptides