Enzymatic cross-linking of food proteins

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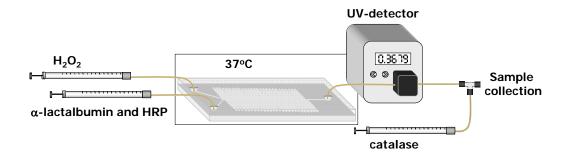
Laboratory of Food Chemistry

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The cross-linking of food proteins is a tool to form potential new food ingredients. A quite novel approach in food protein cross-linking is enzymatic oxidation. By catalyzing the conversion of tyrosine residues to phenolic radicals, peroxidase (EC 1.11.1.7) can initiate the formation of (iso)dityrosine bonds.

Previously, we studied the importance of the protein conformation of α -lactalbumin for the availability of tyrosine residues and the formation of dityrosine protein cross-links. Using calcium-depleted α -lactalbumin (apo-form), the tyrosine residues became more exposed, resulting in a range of cross-linked protein oligomers and polymers. Within food chemistry more and more, attention shifts towards functional molecules instead of components. For this a functionality profile of more specific products, on the molecular level, is required. To direct the formation of specific products with certain functionality, more control is required over the peroxidase-mediated protein cross-linking reaction than is currently achieved at lab scale reactions.



Microreactors are ideal for directing complex enzymatic synthesis, like multienzyme catalysis and cascade reactions. In this research, we study the peroxidase-catalyzed cross-linking of α -lactalbumin in a microfluidic system. To quantify the progress of the reaction, the absorbance increase at 318 nm due to dityrosine formation is monitored on-line and compared with the amount of reacted monomeric α -lactalbumin as determined with size-exclusion chromatography (SEC) at various residence times. The increase in absorbance at 318 nm shows a logarithmic relation with the extent of reacted monomer as described by a reaction model (see reference). With the reproducible cross-linking products being formed, a fast in-line screening method for the peroxidase-mediated cross-linking of α -lactalbumin has been obtained.

WH Heijnis, PA Wierenga, AEM Janssen, WJH van Berkel & H Gruppen In-line quantification of peroxidase-catalyzed cross-linking of α -lactalbumin in a microreactor Chem Eng J **157**, 189-193 (2010)

Available projects

<u>Biochemistry</u>: Identification of α -lactalbumin protein cross-links using a proteomics type of approach. Purification of oligomeric products using fast protein liquid chromatography (FPLC), directed proteolysis and mass spec analysis of purified peptides.

<u>Food Chemistry:</u> Determination of techno-functional properties (gelling, foaming etc) of purified α -lactalbumin oligomers.

BSc and MSc students are encouraged to discuss the possibilities for a research project on this topic. Please contact Dr. Willem van Berkel (willem.vanberkel@wur.nl).