

Poster 4

Generation of anti-inflammatory peptides from β -casein on pilot plant scale

Karina Altmann, Adam Wutkowski, Ingrid Clawin-Rädecker, Martin Klempt, Hans Meisel, Peter Chr. Lorenzen

Department of Safety and Quality of Milk and Fish Products, Max Rubner-Institut, Kiel, Germany

Milk provides a rich source of biologically active components. Growing attention is paid to bioactive peptides from milk proteins hidden within the sequence of native proteins, which can be released by enzymatic hydrolysis. By oral consumption of active peptides or liberation of these peptides during gastrointestinal digestion they may exert anti-inflammatory, antioxidative or antihypertensive activities [1-3].

The aim of the present work was to generate anti-inflammatory peptides from bovine β -casein by hydrolysis using different preparations of tryptic enzymes on pilot plant scale.

For preparation of the hydrolysates, proteolysis of a 5 % β -casein solution (w/w) was performed with tryptic enzymes for 3-4 h at 37°C under pH-stat conditions of 7,8. The total proteolysate was fractionated by ultrafiltration (NMWCO = 5 kDa). Freeze-dried peptide fractions were characterized by RP-HPLC. The molar mass and primary structure of peptides were identified by LC-MS and Proteome Discoverer 1.4. The anti-inflammatory activity of peptide fractions was examined by TNF- α mediated activation of the pro-inflammatory transcription factor NF- κ -B in HEK cells.

Peptide fractions > 5 kDa generated by a TPCK-treated trypsin preparation from pork pancreas showed a potential anti-inflammatory activity. In comparison to the control (HEK cells + TNF- α without hydrolysate), the addition of the hydrolysate resulted in a reduction of NF- κ -B activation in HEK cells over 50% with a value of $3,84 \pm 0,32$. The hydrolysate > 5 kDa produced by a food grade trypsin preparation from codfish showed almost the same NF- κ -B activation. Proteolysis with an enzyme preparation from pork pancreas, which exhibits trypsin activity as well as a small chymotrypsin activity, resulted in a lower anti-inflammatory activity. Peptides > 5 kDa generated with codfish enzymes, which have only a small trypsin but a high chymotrypsin activity, showed no anti-inflammatory activity with an NF- κ -B activation of $7,24 \pm 1,82$. The active components were identified as mainly hydrophobic peptides with molar masses between 2 and 7 kDa.

References:

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- [2] MILLS, S. *et al.*, International Dairy Journal 21 (6) 2011: 377-401
- [3] KORHONEN, H. *et al.*, International Dairy Journal 16 (9) 2006: 945-960