

## Peptide profile of whey protein hydrolysates from free and immobilised trypsin

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Whey proteins are widely used in food formulation due to their nutritional and functional properties. From a dietary point of view, enzymatic hydrolysis of whey protein concentrates is interesting e.g. to reduce allergenicity or to produce bioactive peptides. In fact, during recent years  $\alpha$ -lactalbumin and  $\beta$ -lactoglobulin, were shown to contain bioactive sequences. Pancreatic enzymes - preferably trypsin - as well as other enzymes including Alcalase® and pepsin have been used for identification of many known bioactive peptides.

The immobilisation of enzymes on solid carriers can offer several advantages over free enzymes including easy handling, recovery from the reaction medium, reuse and operation in continuous reactors. Traditional carriers include porous silica, porous glass and cellulose derivatives. Zeolites are porous aluminosilicates available in a wide range of particle size and porosity and can also be used as carriers. Spent grains are a brewing by-product with a high content in cellulose and can also be interesting as carriers for enzyme immobilisation because, besides having the necessary characteristics (e.g. stability, rigidity, low mass transfer limitations), they are cheap and food grade.

This work suggests the use of immobilised trypsin on spent grains and zeolite NaY to hydrolyse whey protein concentrates. Hydrolyses of a commercial spray dried whey protein concentrate with 80 wt % of protein were carried out at 37 °C in a 0.5 L stirred, tank-type, batch reactor equipped with pH and temperature control. The pH was kept constant using 0.5 mol.L<sup>-1</sup> NaOH and the degree of hydrolysis was monitored by the pH-stat method. Trypsin from porcine pancreas with an activity of 1020 BAEE/mg (Sigma Chemical Co) was used both in free and immobilised form. For each experiment, samples were collected at variable intervals of time and analysed by RP-HPLC to evaluate the degradation of native whey proteins ( $\alpha$ -lactalbumin and  $\beta$ -lactoglobulin) and the formation of peptides, which were separated according to their polarity.

Keywords: whey protein concentrate, trypsin immobilization, protein hydrolysis, peptide profile