A method for upgrading porcine blood into a decolourized and tasteful protein ingredient

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Introduction: Porcine blood from Danish abattoirs has mainly been used for animal feed of mink, before the COVID-19 outbreak led to a total lockdown. If the great nutritional and economic potential of blood protein is to be exploited, the blood proteins should be upgraded for human nutrition. The aim of this study is to develop an enzymatic method to produce a decolourized and tasteful protein ingredient by identifying the "window of success", where blood protein is hydrolysed sufficiently to remove the red colour, but without evolving bitter taste.

Materials and methods: Red blood cell concentrate (RBC) was collected from a Danish pig abattoir and stored at 4°C.

Enzymatic hydrolysis of RBC was done using "papain" (Performase® batch no. PSM48000-4517/2 from Enzybel®International Belgium) and "Protease A+B" derived from Aspergillus niger (SHAANXI BETTERING biotechnology, China). RBC was diluted 1:4 (w/w), temperature and pH were adjusted and an enzyme-substrate conc. of 0.35% (w/w) for papain and 0.25% (w/w) for Protease A+B was added. Stirring (200 rpm/min) was applied for 1, 2, 3, 4 and 5 h during hydrolysis, and stopped by heating to 80°C for 15 min. The samples were adjusted to pH 5.0 ± 0.1, centrifuged at 7000 g for 45 min.

Decolouration (DD%). Supernatant was diluted 1:20, and the absorbance at 405 nm was measured using a Lambda 25 UV-VIS Spectrophotometer (Perkin Elmer, USA). The DD% was calculated according to Shi et al., 2015.

Degree of Hydrolysis (DH%). The o-phthaldialdehyde (OPA) test was applied to determine the DH% (Nielsen et al., 2001). The success criterion was defined as average scores < 15%.

Protein recovery (%) was calculated from the protein content (6.25*N%). The success criterion was defined as recovery > 55%.

Sensory evaluation. Five untrained assessors evaluated off-flavours, using a 4-point scale 1 = no off-flavour, 2 = low, 3 = medium and 4 = high intensity. The success criterion was defined as average scores ≤2.

The study was conducted as two experiments.

Results: Experiment 1 - DH% and sensory evaluation:

The DH% increased for both enzymes as the treatment time increased. For papain, DH% increased from 0.7 to 11.8, whereas Protease A+B increased from 3.6 to 8.8. The sensory evaluation indicated a high intensity of bloody off-flavour early in the treatment period, which decreased over time, whereas bitterness became more intense. An acceptable degree of off-flavour was found after 4 hours treatment for both enzymes.

Experiment 2 - DD% and recovery: The degree of decolouration (DD%) increased during time. For both enzymes, samples with DD%-values of 91-93% had a light rose colour, while a DD% of 96-100% was light yellow. DD%-values >96% were obtained after 1 hour for papain, and after 3 hours for Protease A+B.

Protein recovery increased over time for papain and decreased for Protease A+B. A satisfying colour along with a recovery of ≥ 55% was obtained after 4 hours for papain and after 3 hours for Protease A+B.

Conclusions: This study indicates that the method can be used to identify a "window of success" for enzymatic hydrolysis of porcine blood

Future studies will focus on size exclusion chromatography (SEC) and relating it to DH% and the intensity of off-flavours

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Literature:

Nielsen et. al. (2001). Improved Method for Determining Food Protein Degree of Hydrolysis, Journal of Food Science, vol 66, No 5, 642-646.

Shi, J. et. al. (2015) Properties of Hemoglobin Decolorized with a Histidine-Specific Protease. Journal of Food Science, vol 80, No 6, E1202-E1208.