Obtaining of Bioactive Hydrolysates from Protein of Californian Red Worm (Eisenia fetida) Through Enzymatic Hydrolysis and Crossflow Filtration.

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ABSTRACT

The California red earthworm (*Eisenia Fetida*) plays an important role in the decomposition of garbage and organic waste. However, although they have been used for thousands of years for their therapeutic benefits, the scientific evidence for the use of bioactive compounds has not been well proven in practice. The objective of this study was to optimize the enzymatic hydrolysis of California red earthworm meat to obtain peptides with antioxidant capacity and it's scaled up from laboratory scale to bench scale, and then to separate it in 3 and 1 kDa fractions in a membrane system of cross-flow filtration. The worms were manually separated, washed with water, purged for 4 hours with 4% sodium bicarbonate, then sacrificed using 7% saline solution, and finally the worms were washed with drinking water. The optimization of the hydrolysis used a spherical composite central response surface design with five points at the center using 4 factors, pH (7-9), temperature (40-60 °C), substrate (100-200 g) and alkalase enzyme (500-1500 uL) and as response variables, soluble protein percentage (PP), degree of hydrolysis (DH), antioxidant capacity (AC) ABTS and FRAP, implementing a 7.5 L reactor. The procedure to achieve the dimensional analysis consists of three steps: Listing important variables, verifying dimensional homogeneity by transferring it to a dimensionless form and determining dimensionless Pi numbers from a transformation matrix, "Pi theorem". Fractionation was performed using 7-channel membranes of 0.013 m² area and 250 mm length (Tami inc., France) with molecular weight cut-off of 3 and 1 kDa, using a transmembrane pressure of 4 bar and a retention flow rate of 450 L/h at 20°C and pH 8.5. The optimal hydrolysis conditions are pH 8.5, temperature 45°C, with 125.01g substrate and 1243 uL of enzyme, obtaining DH of 16.52%, PP of 3.38% and AC of 2055 and 170 umol-equ trolox/g protein for ABTS and FRAP, respectively. Additionally, the optimal hydrolysate has an ORAC of 823 umol-equ trolox/g protein and iron chelation with IC50 at 150 ppm. The dimensional analysis of the hydrolysis process from 0.5 L to 7.5 L showed that the dimensionless number for the scale-up is the Reynolds, the scaling was performed with geometric similarity modifying the impeller speed which went from 240 rpm in 0.5L to 122.45 rpm in 7.5L. Additionally, the verification of the response variables was carried out without obtaining statistically significant differences between both ($P<0.05$). The purification of the peptides by means of the membrane system concentrated the proteins of the retained with respect to the initial fluid, while the concentration of the permeate is significantly lower compared to the original fluid, both in the 3 and 1 KDa membrane. Additionally, the ABTS and ORAC activities in the permeates with respect to the retained are significantly higher for both membranes. It is concluded that the enzymatic hydrolysate of Californian red worm has a high AC and a low IC50 in iron chelation, making it a substrate of interest for application in different industries.

Keywords: Californian red earthworm, degree of hydrolysis, scaled up, antioxidant capacity

Biography:

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