

Purification and characterization of nitric oxide inhibitory peptides from *Actinopyga lecanora* through enzymatic hydrolysis

ABSTRACT

Actinopyga lecanora, commonly known as sea cucumber, is a rich protein source. This marine protein source was hydrolyzed using six proteases to generate anti-inflammatory hydrolysates and bioactive peptides. Bromelain hydrolysates after 1 h hydrolysis exhibited the highest nitric oxide (NO) inhibitory activity reflecting anti-inflammatory activity. A sequence of two fractionation methods was employed to fractionate the peptides based on their hydrophobicity using a semi-preparative RP-HPLC and isoelectric points using isoelectric focusing technique. Based on these fractionation methods, basic peptides with relatively higher hydrophobicity provided higher NO-inhibitory activity than did acidic peptides. Furthermore, using Q-TOF mass spectrometry; 12 peptide sequences were successfully identified. The inhibitory effect of the purified peptides from *A. lecanora* on NO production by lipopolysaccharide (LPS)-stimulated RAW 264.7 cells was investigated. The three identified bioactive peptides, namely LREMLSTMCTARGA, AVGPAGPRG and VAPAWGPWPKG, exhibited the highest NO-inhibitory activity with values of 76.3, 66.6 and 69.9%, respectively. These results revealed that *A. lecanora* could be used as an economical protein source for the production of high-value bioactive peptides with potent anti-inflammatory activity using RAW 264.7 cell lines as model. These peptides may be useful ingredients in food and pharmaceutical applications.

Keyword: *Actinopyga lecanora*; Bioactive peptides; Enzymatic hydrolysis; NO-inhibitory activity; Anti-inflammation