HIDROLISIS PROTEIN DARI BIJI JARAK KEPYAR *(Ricinus communis L.)* DENGAN ENZIM TRIPSIN USP DAN UJI AKTIVITAS ANTIBAKTERI PEPTIDA YANG DIHASILKAN

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INTISARI

Telah dilakukan penelitian hidrolisis protein biji jarak kepyar dengan enzim tripsin *grade* USP dan uji aktivitas peptida yang dihasilkan sebagai antibakteri. Tujuan utama penelitian ini adalah mengidentifikasi peptida aktif antibakteri dari hidrolisat protein biji jarak kepyar. Ekstraksi protein biji jarak kepyar dengan pengadukan 12 jam menggunakan sodium dodesil sulfat (SDS) 0,01%. Protein hasil ekstraksi dihidrolisis enzimatis menggunakan enzim tripsin. Hidrolisat protein difraksinasi menggunakan kolom *solid-phase extraction* (SPE) penukar kation dengan dielusi pada pH 4, 5, 6, 7, dan 8. Masing-masing fraksi peptida diuji aktivitas antibakterinya dengan metode difusi cakram terhadap bakteri *S.aureus* dan *E.coli* untuk mengetahui aktivitas antibakterinya. Fraksi peptida antibakteri paling tinggi selanjutnya ditentukan peptidanya berikut urutan asam aminonya dengan menggunakan HRMS.

Ekstraksi protein biji jarak kepyar dengan larutan SDS berhasil dilakukan, diperoleh rendemen protein mencapai 41,35%. Enzim tripsin USP mampu menghidrolisis protein dengan derajat hidrolisis mencapai 82,15%. Fraksinasi hidrolisat protein penukar kation menghasilkan fraksi peptida pada pH elusi 4-8 masing-masing sebesar 9,14%; 6,57%; 4,42%; 2,51%; dan 1,29% dari total peptida.. Fraksi peptida yang memiliki aktivitas antibakteri, yaitu fraksi peptida pH 4 dan pH 5 dengan diameter zona inhibisi terbesar dimiliki fraksi peptida pH 4 dengan zona penghambatan sebesar 4-4,5 mm. Identifikasi peptida fraksi pH 4 dengan HRMS menghasilkan 5 peptida spesifik yang terdeteksi dengan perangkat lunak *online* MASCOT, yaitu THIALLQTK + *Acetyl* (N-term), *Phospho* (ST); IEGLLPPGTK + *Acetyl* (N-term), *Phospho* (ST); ARGPLQMMAR + *Oxidation* (M); LGGTMIAPQDK + *Oxidation* (M); VAGGRTPVGPR + *Phospho* (ST).

Kata kunci: antibakteri, biji jarak kepyar, peptida, tripsin

HYDROLYSIS OF PROTEIN FROM CASTOR BEAN (Ricinus communis L.) BY USP GRADE TRYPSIN AND ANTIBACTERIAL ACTIVITY ASSAY OF THE RESULTED PEPTIDES

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ABSTRACT

A research on hydrolysis of the castor bean protein has been carried out using USP grade trypsin followed by antibacterial activity test of resulted peptides. The main objective of this study was to identify antibacterial active peptides from the castor bean protein hydrolyzate. Protein of castor bean was extracted with sodium dodecyl sulfate (SDS) 0.01% using stirring for 12 hours. The extracted protein was hydrolyzed enzimatically using trypsin enzyme. Protein hydrolyzate was fractionated using a cation exchange solid-phase extraction (SPE) coloumn eluted at pH 4, 5, 6, 7, and 8. Each peptide fraction was tested for its antibacterial activity by the disc diffusion method against S.aureus and E.coli. the peptides of highest antibacterial fraction then was determined for their amino acid sequence using HRMS.

The extraction of castor bean seed protein with SDS solution was successfully carried out with a yield reaching 41.35%. The USP trypsin enzyme is able to hydrolyze proteins with the degree of hydrolysis reaching 82.15%. The fractionation of cation exchange protein hydrolyzate produced a peptide fraction at elution pH 4-8 each 9.14%; 6.57%; 4.42%; 2.51%; and 1.29% of total peptide. The peptide fraction that has antibacterial activity are the peptide fraction pH 4 and pH 5 with the largest activity observed at the pH 4 peptide fraction with inhibition zone of 4-4.5 mm. The identification of peptide present at pH 4 fractions with HRMS resulted in 5 specific peptides detected by MASCOT online software, namely THIALLQTK + Acetyl (N-term), Phospho (ST); IEGLLPPGTK + Acetyl (N-term), Phospho (ST); ARGPLQMMAR + Oxidation (M); LGGTMIAPQDK + Oxidation (M); VAGGRTPVGPR + Phospho (ST).

Keywords: antibacterial, castor bean, peptide, trypsin