

Chanthakan Nuchasuk 2012: Purification and Characterization of a Ribosome-Inactivating Protein from Seed Coat of *Jatropha curcas*. Doctor of Philosophy (Biochemistry), Major Field: Biochemistry, Department of Biochemistry.

Thesis Advisor: Associate Professor Sunanta Ratanapo, Ph.D. 172 pages.

A new type 1 ribosome-inactivating protein, which demonstrated lectin-like hemagglutination activity, was first purified from the seed coat of *J. curcas* Linn. by ammonium sulfate precipitation and chromatography on DEAE-Sephacel™ and CM-cellulose columns. It was designated as “Jc-SCRIP”. Purification fold of Jc-SCRIP increased 113.5 times in final step with 1.12% yield of the total protein. It was a monomeric glycoprotein of a molecular mass of 38,938 Da, as determined by MALDI-TOF/MS. Structural analysis of Jc-SCRIP indicated that its major structure was β -sheet with N-terminal amino acid sequence: AINGGVA. The neutral sugar content of Jc-SCRIP was about 4.80% (w/w). It possessed strong *N*-glycosidase activity that released an RNA fragment of approximately 560 nucleotides from the rabbit reticulocyte rRNA after acidic aniline treatment. The positive antimicrobial effect of Jc-SCRIP was tested by agar dilution technique against 9 human-pathogenic bacteria and 1 fungus. Its most potent inhibitory activity was against *Staphylococcus epidermidis* ATCC 12228, with MIC value of 7.81 μ g/ml. Jc-SCRIP showed the cytotoxic effects to cell lines of a human breast adenocarcinoma (MCF-7), a colon adenocarcinoma (SW620), and a liver carcinoma (HepG2), with IC₅₀ values of 0.15, 0.25 and 0.40 μ M, respectively. Jc-SCRIP had the larvicidal effects to the third instars larvae of mosquitoes, *Aedes aegypti* Linn. and *Culex quinquefasciatus* Say with LC₅₀ values of 1.44 and 0.0303 mg protein/ml, respectively. It also showed the larvicidal effects to the second instars larvae of *Spodoptera litura* and *Spodoptera exigua* with LC₅₀ values of 0.0525 and 0.0629 mg protein/ml, respectively. The potent larvicidal activity of Jc-SCRIP suggests that it may be used as a low cost natural agent to control both mosquitoes and crop pests. Further evaluation of biosafety to human and its toxic stability should be done before the application use.

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