

Sansaneeayorn Kittikajhon 2010: Purification of Curcin from Seed Kernel of Physic Nut and Its Antibacterial and Anticancer Activity. Master of Science (Biochemistry), Major Field: Biochemistry, Department of Biochemistry. Thesis Advisor: Associate Professor Sunanta Ratanapo, Ph.D. 138 pages.

A toxin, curcin was simply purified from seed kernel of *Jatropha curcas* L. (KUBP 33). The crude curcin extract was prepared by stirring the grounded seed kernel in Tris buffer, pH 7.5 for 24 h. The fat in the upper layer was removed after standing at 4 °C and the extract was dialyzed against sodium acetate buffer, pH 4.5, followed by centrifugation to remove the pellet. The soluble purified curcin showed a major band of relative molecular weight of 29 kDa by SDS-PAGE. The purified curcin is a single polypeptide chain of molecular mass 28.4 kDa by MALDI-TOF MS. The toxicity on protein synthesis of the purified protein was shown by its rRNA *N*-glycosidase activity towards RNA from rabbit reticulocyte lysate. The LC-MS/MS analysis result revealed that the isolated protein was curcin. In this study the antimicrobial effect of the purified curcin was investigated by agar dilution technique. The result demonstrated inhibitory effect of curcin against 17 strains of the human-pathogenic bacteria with most potent to *Staphylococcus epidermidis* ATCC12228 and *Bacillus subtilis* ATCC6633 with MIC value at 78.1 µg/ml. The cytotoxic effects of the purified curcin to cell lines of oral cavity cancer (KB, ATCC CCL-17), breast cancer (MCF7, ATCC HTB-22), small cell lung cancer (NCI-H187, ATCC CRL-5804) and human colon cancer (SW620, ATCC CCL-28) were evaluated. The result showed that the purified curcin was not toxic to any cancer cell lines. Curcin in seed kernel of *J. curcas* of the total 157 accessions from various regions (38 provinces) of Thailand were evaluated by Western blotting probed with polyclonal antibody to curcin. The curcin content of each accession was calculated from its intensity by using the calibration curve of the purified curcin. The *J. curcas* accessions from various regions were classified into four groups based on the percentage of curcin content. The seed kernel of the three accessions including KUBP 19, 169 and 187 did not have any curcin. The highest curcin content of 9.09% (of kernel dry weight) was found in KUBP 75.

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Student's signature

Thesis Advisor's signature