INTISARI

UJI AKTIVITAS SITOTOKSIK SENYAWA KALANON DAN 2,4-DINITROFENILHIDRAZON KALANON PADA SEL HELA DAN PENGARUHNYA TERHADAP EKSPRESI GEN P53 DAN PROTEIN P21

Kalanon adalah senyawa turunan kumarin yang diisolasi dari tumbuhan *Calophyllum sp.* Penelitian terhadap kalanon terus berkembang, hingga diperoleh senyawa turunannya yaitu 2,4-dinitrofenilhidrazon kalanon. Kalanon dan 2,4-dinitrofenilhidrazon kalanon menunjukkan aktivitas sitotoksik terhadap sel leukemia L1210 dengan IC_{50} berturut-turut 59,40 dan 47,09 µg/ml. Beberapa kumarin menunjukkan aktivitas sitotoksik dan apoptosis pada sel kanker. Di antara senyawa tersebut adalah *2-(8-Hydroxy-6-methoxy-1-oxo-1H-2-benzopyran-3-yl) Propionic Acid* (NM-3). Pemberian NM-3 pada beberapa sel menunjukkan peningkatan ekpresi p53. Pada kanker pada sel kanker payudara MCF-7 and ZR-75-1, NM-3 menginduksi protein p21. Kalanon dan 2,4-dinitrofenilhidrazon kalanon diduga mempunyai aktivitas yang sama pada sel kanker serviks HeLa.

Penelitian ini bertujuan untuk mengetahui aktivitas sitotoksik kalanon dan 2,4-dinitrofenilhidrazon kalanon pada sel kanker serviks HeLa serta mengetahui apakah aktivitas sitotoksik tersebut ditimbulkan melalui proses apoptosis, dan bagaimana pengaruhnya terhadap ekspresi gen p53 dan protein p21. Uji aktivitas sitotoksik kalanon dan 2,4-dinitrofenilhidrazon kalanon pada kultur sel kanker serviks HeLa dilakukan dengan metode *MTT assay*. Uji apoptosis dilakukan dengan pemberian fluorokrom etidium bromida dan *acridine orange*. Uji ekspresi p53 dilakukan dengan pemeriksaan PCR menggunakan primer p53 *wild type* dan uji ekspresi p21 dilakukan dengan pemeriksaan menggunakan imunohistokimia setelah inkubasi selama 24 jam.

Hasil penelitian menunjukkan kalanon dan 2,4-dinitrofenilhidrazon kalanon memiliki aktivitas sitotoksik dengan nilai IC_{50} sebesar berurut-turut 22,887 dan 61,893 µg/mL. Kedua senyawa menginduksi apoptosis, meningkatkan ekspresi gen p53, namun tidak meningkatkan ekspresi protein p21.

Kata-kata kunci : Kalanon, 2,4-dinitrofenilhidrazon kalanon, Sitotoksik, Gen p53, Protein p21, Karsinoma serviks

ABSTRACT

THE CYTOTOXIC ACTIVITY OF KALANON AND 2,4-DINITROFENILHIDRAZON KALANON ON HELA SERVICAL CANCER CELL LINE AND ITS EFFECT TO P53 GENE AND P21 PROTEIN EXPRESSION

Calanone (coumarin derivate compound), isolated from *Calophyllum sp.* and 2,4-dinitrophenilhidrazone calanone (calanone's derivate) had been shown to have cytotoxic activity on leukemia L1210 cell line with IC_{50} are 59.40 and 47.09 μg/ml respectively. Several coumarins had been shown caused apoptosis on cancer cell line and one of them, i.e. 2-(8-Hydroxy-6-methoxy-1-oxo-1*H*-2-benzopyran-3-yl) Propionic Acid (NM-3) . The exposure to NM-3 is associated with increases in expression of the p53 tumor suppressor. In human MCF-7 and ZR-75-1 breast cancer cells, NM-3 induces the p21 cyclin-dependent kinase inhibitor. Calanone and 2,4-dinitrophenilhidrazon calanone presumed to have the same activity on HeLa cervical carcinoma cell.

This studi was conducted to investigate the cytotoxic and apoptotic activity of calanone and 2,4-dinitrophenilhidrazone calanone and its effect on p53 and p21 expression on HeLa cervical carcinoma cell. Cytotoxic assay of calanone and 2,4-dinitrophenilhidrazone calanone was performed on HeLa cell line, using MTT assay. Apoptotic assay was performed on HeLa cell line incubated with calanone and 2,4-dinitrophenilhidrazon calanone for 24 hours, by immunofluororescence method, using fluorochromes ethidium bromide and acridine orange. Expression of p53 was examined on HeLa cell line, by PCR with p53 wild-type primer. Expression of p21 was examined on HeLa cell line, by immunohistochemistry method. 5-fluorourasil was used as positive control in cytotoxic, apoptotic assay, and expression of p53.

The result showed that calanone and 2,4-dinitrophenilhidrazone calanone has cytotoxic activity on HeLa cell line, with IC_{50} 22.887 dan 61.893 μg/ml respectively, caused cytotoxicity through apoptotic process, increase the expression of the p53 tumor suppressor, but did not elevate the expression of p21.

Key Words : Calanone, 2,4-dinitrophenilhidrazon Calanone, p53 gene, p21 protein, Cervical carcinoma