DOCTORAL THESIS

Characterization of furanodienone as a potential agent to treat breast cancer

Li, Yingwei

Date of Award:
2011

Characterization of Furanodienone as a Potential Agent to Treat Breast Cancer

LI Yingwei

A thesis submitted in partial fulfillment of the requirements

for the degree of

Doctor of Philosophy

Principal Supervisor: Prof. FONG Wang Fun, David.

Hong Kong Baptist University

June 2011

ABSTRACT

Breast cancer is the most common cancer and the second leading cause of women death worldwide. Several receptors including ERα, EGFR and HER2 play an important role in the development and progression of breast cancer. To date, endocrine therapy blocking estrogen receptor alpha (ERα) activity and HER2 targeted therapies have been shown to be promising approaches in the treatment of breast cancer. However, not all patients respond to these agents and identification of novel agents that can inhibit ERα, EGFR and HER2 signaling remains important. The aims of this study were to investigate furanodienone, an active chemical component isolated from Rhizoma Curcumae whether had anti-cancer activities and inhibited ERα, EGFR/HER2 signaling in human breast cancer cells.

In HER2-overexpressing human breast cancer cells, we observed that furanodienone caused cell cycle arrest in BT474 cells and induced apoptosis in SKBR3 cells. Furanodienone interfered with EGFR/HER2 signaling in treated cells as shown by decreases in phosphorylated EGFR, HER2, Akt, Gsk3β and an increase in $p27^{kip1}$ protein. Furanodienone also inhibited EGF-induced phosphorylation of EGFR, HER2, Akt and Gsk3β resulted in inhibiting EGF-induced cell cycle progression in BT474 cells and EGF-induced cell growth in both BT474 and SKBR3 cells. In BT474 and SKBR3 cells, EGFR-specific siRNA knockdown did not affect the cell growth inhibitory effect of furanodienone. On the contrary, specific siRNA knockdown of HER2 increased cellular resistance to furanodienone toxicity. In HER-2 deficient MDA-MB-231 cells the transfection and expression of HER2 increased the sensitivity of cells to furanodienone toxicity.

In ERα-positive MCF-7 and T47D cells, furanodienone inhibited cell proliferation in a dose (10–160 μM) dependent manner. ERα-negative MDA-MB-231 cells were less sensitive to furanodienone than MCF-7 and T47D cells. Furanodienone could effectively block 17β-estradiol (E2)-stimulated MCF-7 cell proliferation, cell cycle progression and induce apoptosis evidenced by the flow cytometric detection of sub-G1 DNA content and the appearance of apoptotic nuclei after DAPI staining. Furanodienone specifically down regulated ERα protein and mRNA expression levels without altering ERβ expression. Furanodienone treatment inhibited E2-stimulation of estrogen response element (ERE)-driven reporter plasmid activity and ablated E2-targeted gene (e.g. c-Myc, Bcl-2 and cyclin D1) expression which resulted in the inhibition of cell cycle progression and cell proliferation, and in the induction of apoptosis. Knockdown of ERα in MCF-7 cells by ERα-specific siRNA decreased the cell growth inhibitory effect of furanodienone.

In conclusion, furanodienone were able to inhibit EGFR/HER2 signaling pathway in HER2-overexpressing BT474 and SKBR3 cells. More importantly the effect of furanodienone was specifically dependent on HER2, but not EGFR, expression. Furanodienone also inhibited ERα signaling in ERα-positive MCF-7 cells. These results

suggest that furanodienone and its related compounds may provide novel approaches for the prevention and treatment of breast cancers.

TABLE OF CONTENTS