17-Demethoxygeldanamycin inhibits self-renewal of breast cancer stem cells through EZH2/c-Myc/Bmi1 pathway

W.W. Chang
School of Biomedical Sciences, Chung Shan Medical University, Taichung, Taiwan

Aim/Background: Breast cancer stem cells (BCSCs) could be enriched by cell sorting according to cell surface markers (CD24-CD44+) or intracellular aldehyde dehydrogenase (ALDH) activity as well as by tumorsphere cultivation. Because of the involvement of BCSCs in tumor initiation, drug resistance or metastasis, to target BCSCs is considered as the key for successful breast cancer therapy. Heat shock protein 90 (Hsp90) is a kind of stress proteins and is known to be overexpressed in a variety of cancers. We previously discovered that 17-demethoxygeldanamycin (17-DMAG), one of the Hsp90 inhibitors, could decrease ALDH+ population of human breast cancer cells. Here we would like to study the molecular mechanisms of the inhibitory effect of 17-DMAG in suppression of self-renewal capability of BCSCs.

Methods: The self-renewal of human breast cancer cells including MDA-MB-231 and AS-B244 was determined by mammosphere formation assay. The interaction of Hsp90 between Bmi1 or c-Myc was analyzed by immunoprecipitation (IP). Chromatin IP was used for study the binding of c-Myc to Bmi1 promoter. Luciferase based reporter assay was used for determination of the activity of c-Myc. The expression of Bmi1, c-Myc and EZH2 was determined by western blot. Overexpression of EZH2 by lentivirus transduction was used to study the role of EZH2 in the inhibitory effect of c-Myc and Bmi1 expression.

Results: We found that 17-DMAG could inhibit self-renewal of BCSCs in a dose dependent manner. 17-DMAG down-regulated the expression of Bmi1 in both mRNA and protein level. IP analysis revealed that Bmi1 did not interact with Hsp90 suggesting that Bmi1 is not a client of Hsp90. 17-DMAG inhibited the expression of c-Myc, suppressed the binding of c-Myc in Bmi1 promoter as well as the transcriptional activity of c-Myc. We also discovered that 17-DMAG diminished the expression of EZH2. Overexpression of EZH2 could overcome the inhibitory effect of 17-DMAG in suppression of Bmi1 and c-Myc expression.

Conclusions: Our data demonstrated that 17-DMAG could suppress the self-renewal of BCSCs through EZH2/c-Myc/Bmi1 pathway. Hsp90 inhibitors such as 17-DMAG may serve as effective agents in breast cancer treatment.

Disclosure: All authors have declared no conflicts of interest.