Inhibition of Hsp60 expression by doxorubicin and replicative senescence instauration in mucoepidermoid carcinoma cells

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Objective

The resistance of cancerous cells to chemotherapy is the main limitation for cancer treatment. Since cancer cells proliferate indefinitely, a requisite for their immortalization is to bypass replicative senescence (RS). RS is a natural barrier used by cells against tumorigenesis and some key components of senescence process may be regulated by changes in chaperones levels. Doxorubicin (DOX) is an anthracycline antibiotic used in cancer chemotherapy with inhibiting effect on some molecular chaperones. This work is to determine the effect of DOX on the expression levels of the chaperonin Hsp60 in a lung carcinoma cell line (NCI-H292) to reveal a new anticancer mechanism.

Materials and Methods

NCI-H292 cell were treated with DOX (20nM, 40nM, 80nM, 160nM) for 5 days. To determine the effect of DOX on RS, selected markers of senescence and cell cycle analysis were conducted. Moreover, Hsp60 expression level was evaluated using qPCR and western blot. Hsp60 hyperacetylation and Hsp60 ubiquitination was investigated using immunoprecipitation.

RESULTS: A dose-dependent reduction of cell viability was observed. Cells exhibited typical senescent phenotype, positivity for SA β-gal assay and showed cytoskeleton remodelling due to an increase of vimentin level at 40nM, 80nM, 160nM. Moreover, cell cycle analysis showed that the cells were in the G0/G1 phase at 80nM, 160nM. The treatment increased the expression mRNA, reduced the protein level and induced hyperacetylation and ubiquitination of Hsp60.

Conclusion

DOX treatment resulted in cellular senescence, which seems to coincide with decreased level and hyperacetylation of Hsp60. This reduction could be due to its ubiquitination. Hsp60 is involved in tumor-invasiveness. The deregulation of Hsp60 activity, could affect protein folding and cause proteotoxic stress ultimately resulting in the switch of pro-survival signaling to cancer cell senescence. Finally, DOX could interfere with the pro-tumoral role of Hsp60 which could be used as target for chemotherapeutic treatment.