

MORTALIN-P53 INTERACTION IN CANCER CELLS IS STRESS DEPENDENT AND CONSTITUTES A NOVEL TARGET FOR LIVER CANCER THERAPY

W.-J. Lu¹, N.P. Lee¹, S.C. Kaul², R.T. Poon¹, R. Wadhwa², J.M. Luk³.

¹*The University of Hong Kong, Hong Kong, Hong Kong S.A.R., China;* ²*National Institute of Advanced Industrial Science & Technology (AIST), Tsukuba, Japan;* ³*National University of Singapore, Singapore,*

BACKGROUND AND AIMS: The mortality rate of HCC is high due to tumor recurrence and lack of effective treatment. By proteomics analysis of matched tumor and non-tumor tissues, mortalin was identified as a marker for hepatocellular carcinoma (HCC) metastasis and recurrence, suggesting its tight link in HCC development and recurrence. The aim of this study is to examine the role of mortalin in hepatocarcinogenesis. **METHODS:** The mortalin expression was examined in 100 HCC patients by IHC staining and western blotting. The clinical parameters were analyzed by SPSS. The mortalin specific shRNA was constructed for the loss function investigation. The apoptotic cell was detected by In Situ TUNEL staining. The interaction of Mortalin and p53 in liver cancer cell-line was examined by CoIP assay. The location of mortalin and p53 were observed by immunofluorescence staining. **RESULTS:** Mortalin was overexpressed in the 61% of clinical samples. The clinical-pathology analysis showing its tight link with HCC recurrence ($p = 0.005$) and the microsatellite formation ($p = 0.028$), a crucial feature of intra-hepatic metastasis. The further functional investigation has shown that unlike most of the cancer cells, HepG2 hepatoma lacked mortalin-p53 interaction and is resistant to apoptosis induced in response to knockdown of mortalin expression. Furthermore, the mortalin-shRNA-sensitive HCC cells show a higher degree of p53 phosphorylation. Since phosphorylation in p53 is a sign for genotoxic stress, we treated HepG2 cells with Cisplatin at sub-lethal concentrations and demonstrated that under these conditions p53 is (i) phosphorylated at the critical site, (ii) can be co-precipitated with mortalin, and (iii) apoptosis is induced by mortalin-shRNA. Again, the mortalin shRNA induced apoptosis can be rescued by p53-shRNA or p53 inhibitor. Mortalin knock-down also sensitizes the cancer cells to H₂O₂ and Doxorubicin induced stress. When the interaction of mortalin with p53 was challenged with a chemical or peptide inhibitor, apoptosis is induced in the mortalin-shRNA sensitive cells and in the non-sensitive cells but only in the presence of stress inducing reagents. **CONCLUSIONS:** Mortalin-p53 interaction is part of a cell stress response, which impairs p53 apoptotic ability in HCC cells. Targeting mortalin-p53 interaction is a promising strategy for HCC therapy.