Murine model to elucidate mechanisms of chemoresistance in lung cancer

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Introduction: Lung cancer is the leading cause of cancer deaths worldwide. Small cell lung cancer (SCLC) is the most aggressive subtype due to its rapid growth and early, widespread metastasis. SCLC is particularly sensitive to chemotherapy and radiation therapy; however drug resistance quickly develops making SCLC overwhelmingly fatal. SCLC is not treated surgically and thus study of SCLC pathogenesis is hampered by the lack of human tissue. Rb loss and p53 mutation are detected in ≥80% of SCLCs. We hypothesized that combined Rb loss and p53 loss or mutation targeted to the murine lung epithelium would result in a manipulatable murine model of human SCLC to elucidate mechanisms leading to chemoresistance.

Objective: Mice with Rb loss and p53 loss or mutation targeted to the lung epithelium were generated and uniformly developed lung tumors as well as mediastinal and/or liver tumors leading to death by ~5.5 months of age (n=16). The goals of this study were to confirm the Rb and p53 allele status in the tumors and tumor derived cell cultures, and to determine whether the cancer phenotype mimicked human SCLC.

Methods: Cre-LoxP technology was used to target Rb and p53 genetic alterations to the lung epithelium. Thus, PCR analyses were conducted on DNA isolated from lung tumors, metastases and tumor cell cultures to confirm Rb and p53 gene recombination, and thus ablation. Mutant p53 expression was assessed by immunohistochemistry. Lung and metastatic tumors were examined histologically and for cell lineage specific marker expression by immunohistochemistry including the SCLC neuroendocrine markers synaptophysin and calcitonin gene related peptide (CGRP), the non-neuroendocrine cell markers Clara cell secretory protein (CCSP) and surfactant protein C (proSPC), and general lung epithelial cell markers Sox2 and thyroid transcription factor 1 (TTF1).

Results: Rb and p53 gene recombination and p53 mutant protein expression were detected in all lung tumors, metastatic tumors and tumor cell cultures. Lung and metastatic tumors were morphologically indistinguishable from human SCLC characterized by nests of malignant cells with scant cytoplasm, finely granular nuclear chromatin, numerous mitotic figures and extensive necrosis. The murine tumors stained positively for neuroendocrine markers and TTF1 similar to human SCLC. As expected, tumors were negative for the parenchymal type II cell marker, proSPC and positive for the conducting airway marker, Sox2. A subpopulation of primary lung tumors, but not metastases, had focal CCSP staining raising the possibility that CCSP expression identifies a distinct SCLC subtype.

Conclusions: A murine model was generated based upon genetic alternations in human SCLC. Mice with combined Rb and p53 loss or mutation targeted to the lung epithelium uniformly develop fatal metastatic lung cancers. The tumors are phenotypically indistinguishable from human SCLC and have similar biologic behavior including rapid growth and early metastasis. This model can be used to investigate the mechanisms of chemoresistance in SCLC.

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