Lung cancer is one of the most common cancer in the world. Cancer metastasis is a key factor in survival of lung cancer patients. Metastasis process requires many complex steps, including cell adhesion, cell growth, death and angiogenesis. To understand the molecular mechanisms of metastasis, we used a lung adenocarcinoma cell line model, low-metastatic CL1-0 and highly metastatic CL1-5 cell lines, to perform the microarray assay. We found that PLEK2 is upregulated in highly invasive CL1-5 lung cancer cell line. Moreover, the role of PLEK2 and lung cancer is not clear. We established stable knockdown PLEK2 gene system in CL1-5 lung adenocarcinoma cells. The shRNA is a sequence of RNA that makes a tight hairpin turn that can be used to silence target gene expression via RNA interference (RNAi). Finally, we suggested that knockdown PLEK2 gene expression can inhibit lung cancer cells migration, invasion, proliferation, and anchorage-dependent growth ability. We found that the shPLEK2 cells when compared to parental cells have differ morphologies. These result can help us understanding the role of PLEK2 gene in lung cancer cells. Its potential role in NSCLC progression provides an attractive target for anticancer therapy.

Keywords: lung cancer, metastasis, Pleckstrin 2, PI3K, Epithelial to mesenchymal transition
4.6 建立抑制PLEK2之系統

4.6.1 菌體培養

4.6.2 菌體保存

4.6.3 質體DNA萃取

4.6.4 轉染至細胞內 (transfection)

4.6.5 建立抑制PLEK2之細胞株

4.7 西方墨點法

4.7.1 蛋白質樣品的製備

4.7.2 蛋白質定量

4.7.3 前置作業 (製作SDS膠體)

4.7.4 SDS膠體的製備

4.7.5 SDS膠體電泳

4.7.6 轉漬法

4.7.7 杂合反應 (Hybridization)

4.8 細胞功能分析實驗

4.8.1 細胞生長速率分析 (MTT Assay)

4.8.2 細胞群落能力分析 (Colony Formation Assay)

4.8.3 細胞癌化能力分析 (Soft Agar Colony Formation Assay)

4.8.4 細胞移動能力分析 (Wound Healing Assay)

4.8.5 細胞遷移能力分析 (Transwell Migration Assay)

4.8.6 細胞入侵能力分析 (Transwell Invasion Assay)

4.8.7 細胞形態之觀察

5. 結果

5.1 不同肺癌細胞株中PLEK2基因mRNA的表現量

5.2 建立肺腺癌CL1-5細胞株中，抑制PLEK2的表現系統

5.3 抑制PLEK2細胞株生長速率降低

5.4 抑制PLEK2細胞群落形成的能力降低

5.5 抑制PLEK2細胞癌化程度降低

5.6 抑制PLEK2細胞株會抑制細胞爬行能力

5.7 抑制PLEK2細胞株會抑制其遷移能力

5.8 抑制PLEK2之細胞株會抑制入侵能力

5.9 抑制PLEK2對細胞形態的影響

6. 討論

7. 結論

参考文獻

附錄