口腔癌中 LRRC1 參與的抑癌機制探討

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頭頸部鱗狀細胞癌(HNSCC)是一種致命惡性腫瘤，其中又以口腔鱗狀細胞癌(OSCC)是好發率最高的．根據台灣衛生福利部的癌症登記報告中，十大癌症排名之中口腔癌為第七名，但在死亡率中卻為第五名．嚼食檳榔、抽菸與飲酒目前被認為是口腔癌最主要的危險因子，但 HPV 感染、慢性牙周病與飲食習慣也都可能成為口腔癌的危險因子之一．儘管現代醫療進步與發展，口腔癌的五年存活率仍大約為 50%，因此，了解口腔癌的發展與致癌機制有助於改善 OSCC 的治療。

近年來，許多研究證明在基因序列不改變的情況下，基因表達的方式可受某些調控機制而有所不同。這些調控機制包含：包含 DNA 甲基化、組蛋白修飾和 microRNA 的影響。MicroRNA 是一段約 19-22 bp 的非編碼 RNA，功能為直接引發 mRNA 的分解或是間接抑制轉譯的進行，來調節目標基因的表達。許多研究指出，大部分 microRNA 會受到 DNA 甲基化的調控，導致基因在癌細胞中異常表達促使腫瘤的發生。

先前實驗室的研究中，我們從口腔癌的病患檢體中，藉由 DNA methylation array 的分析結果中發現 miR-124-3 呈現高度甲基化，且在細胞株中 miR-124-3 的表現量下降，若在口腔癌細胞株中 overexpress miR-124-3 則能夠恢復其抑癌基因的功能．接著我們想進一步去了解 miR-124-3 在口腔癌中的分子機制與功能．因此透過 TargetScan7.1、miRDB、DianaLabs 三個資料庫當中篩選了一些 miR-124-3 下游基因，在經過文獻比對後，並挑選了 LRRC1 作為接下來的研究目標．根據 qRT-PCR 和 Luciferase assay 發現 miR-124-3 能夠直接去抑制 LRRC1 的表達．因此我建立表達 shLRRC1 的慢病毒載體去感染 OSCC cell lines，並利用 western blotting assay 和 qRT-PCR 去確認 LRRC1 被 knockdown，且觀察口腔癌細胞株生長和遷移的能力是否受影響，並接續進行動物實驗以證實 miR-124-3 透過靶向 LRRC1 以抑制口腔癌。

Tumor-inhibitory function and mechanism of LRRC1 in oral cancer

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Head and neck squamous cell carcinoma (HNSCC) is a fatal malignant tumor, of which oral squamous cell carcinoma (OSCC) is the most likely to develop. According to the cancer registration report of the Ministry of Health and Welfare of Taiwan, oral cancer is the seventh among the top ten cancers, but the fifth in the mortality rate. Betel nut chewing, smoking and alcohol drinking are currently considered to be the main risk factors for oral cancer, and HPV infection, chronic periodontal disease and eating habits may also become one of the risk factors for oral cancer. Despite the progress and development of modern medicine, the five-year survival rate of oral cancer is still about 50%, so understanding the development and pathogenesis of oral cancer may help improve the treatment of OSCC.

In recent years, many studies have proved that without changing the sequence of DNA, the way how DNA sequence is read can be changed. Many regulatory mechanisms can cause differences in gene expression and these include DNA methylation, histone modification and microRNA regulation. MicroRNA is a non-coding RNA of about 19-22 bp, which directly triggers the decomposition of mRNA or indirectly inhibits the progress of translation to regulate the expression of the target gene. Many studies have pointed out that most microRNA is regulated by DNA methylation, resulting in abnormal expression of genes in cancer cells to promote the occurrence of tumors.

In previous laboratory studies, we found from oral cancer patient samples, through the analysis results of a DNA methylation array that miR-124-3 showed a high degree of methylation, and the amount of miR-124-3 in OSCC cell lines decreased. Overexpression of miR-124-3 in these cell lines can restore the function of the gene. We want to further study the molecular mechanism and function of miR-124-3 in oral cancer. Therefore, some downstream genes are predicted through TargetScan7.1, miRDB, and DianaLabs. After a literature comparison, LRRC1 was selected as our research goal. According to qRT-PCR and Luciferase assay, it is found that miR-124-3 can directly suppress the expression of LRRC1. Therefore, I established a lentiviral vector expressing shLRRC1 to infect OSCC cell lines, and used western blotting assay and qRT-PCR to confirm that LRRC1 was knockdown, and observed whether the proliferation and migration of OSCC were affected, and continued to conduct animal experiments. To confirm that miR-124-3 suppresses oral cancer by targeting LRRC1.