RC-15

Expression profiling with microarray in studying differential gene expression in different primary lung cancer cell lines newly established from local Chinese patients

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Introduction We have previously established new primary non-small cell lung cancer cell lines (the HKULC series) from Hong Kong Chinese patients with non-small cell lung cancer with established growth kinetics as well as morphological and immunohistochemical characterization. The aim of this study was to further characterize these established primary lung adenocarcinoma cell lines with expression profiling. Method Total RNA was extracted from 3 primary lung adenocarcinoma cell lines (HKULC 1,2 and 3) and 4 normal human bronchial epithelial cell lines (NHBE, SAEC and BEAS-2B from Clonetics, and HCCBE1 from UTSW). Reverse transcription, in vitro transcription, biotin labelling, fragmentation and hybridization onto Affymetrix GeneChip HG-U133A were done based on standard protocol from Affymetrix. Hybridized Genechips were scanned and signals captured were first pre-processed with median normalization and scaling. Normalised signals were analysed with Matrix (Microarray Transformation in Excel), a new bioinformatics tool from UTSW for analysis of microarray data. Results: From analysis of fold changes inferred from log (base 2) ratio between individual cell lines and averaged signals from adenocarcinoma cell lines vs averaged normal human bronchial epithelial cell lines, some genes or groups of genes were found to show consistent pattern of differential gene expression. Representative examples are: EGFR, ERBB2, ERBB3(transmembrane receptors coupled with growth or G protein-related signal transduction pathway) were found to be up-regulated in adenocarcinoma cell lines by at least 2 fold compared to the normal human bronchial epithelial cell lines; whereas MYC and MADH4 (transcription cofactors and regulation), CDKN2A, CCND1 and TOB1 (cell cycle and proliferation-related genes), TP53, RB1 and CDH13 (tumour suppressor genes), ABCC1 (drug resistance genes), and TNFRSF6B (anti-apoptotic gene) were found to be down-regulated in lung adenocarcinoma cell lines by at least 2 fold compared to normal human bronchial epithelial cell lines. Hierarchical clustering showed consistent sample and gene clustering of different lung adenocarcinoma cell lines and the normal human bronchial epithelial cell lines. Conclusion Expression profiling of lung adenocarcinoma cell lines identified differentially-expressed genes and could provide substrates and gene targets for in vitro chemotherapeutic trials.

RI – Rheumatology & Immunology

RI-04

Antiphospholipid (aPL) antibody profiles in Chinese patients with systemic lupus erythematosus. MY Mok, *EYT Chan, *K Leung, Yi Lo, WS Wong, CS Lau. Department of Medicine and *Pathology, Queen Mary Hospital, Hong Kong.

Background: aPL antibodies are known to predispose to clinical thrombosis and pregnancy morbidities. Different prevalences have been reported by groups of different cultural background.

Aim: To examine the prevalence of aPL antibodies including lupus anticoagulant (LA), anti-cardiolipin (aCL) and anti-β2GPI (aβ2GPI) antibodies in our Chinese cohort of SLE and to examine their correlation with various clinical manifestations locally.

Methods: Consecutive patients who satisfied the 1982 revised classification criteria for SLE were recruited. Patient records were reviewed for clinical thrombosis and pregnancy morbidity. LA was measured by phospholipid dependent assay, aCL and aβ2GPI antibodies were measured by ELISA. Only patients with results in the high positive range for the latter two antibodies were analysed further for clinical manifestations.

Results: 201 consecutive SLE patients were recruited. Their mean ± SD age at the time of study was 38.3 ± 11.1 and they have been followed up for 10.6 ± 7.0 years. LA was found in 25.4% (51/201) of patients. IgG and IgM aCL antibodies in 31.8% (64/201) and 13.4% (27/201) respectively and IgG and IgM aβ2GPI antibodies in 16/201 (8.0%) and 23/201 (11.4%) respectively. Clinical vascular thrombosis was identified in 27/201 (13.4%) giving a thrombotic risk of 0.01%/patient-year. Recurrent risk in patients with history of thrombosis was 25.9% (7/27). Univariate analysis identified LA, IgG aCL and IgG aβ2GPI antibodies as risk factors for various presentations of clinical thrombosis. Multivariate analysis affirmed the role of LA in the development of clinical thrombosis [RR3.3 CI 1.4-7.7, $p<0.01$] and stroke [RR3.9 CI 1.1-13.3, $p<0.05$] and aβ2GPI antibodies in the development of venous thrombosis [RR7.8 CI 2.0-30.1, $p<0.005$]. aβ2GPI antibodies were the only risk factor found to be strongly relate to recurrence of thrombosis [RR 25.4 CI 2.1-310.8, $p=0.01$] by multivariate analysis. There were only 24 female patients with ≥ 1 pregnancy loss. Only IgG aβ2GPI antibodies were shown to be associated with ≥ 1 unexplained fetal loss at or beyond the 10^{th} gestational week with RR10.2 [CI 1.21-86.59] ($p=0.03$).

Conclusion: The risk of thrombosis in our Chinese SLE cohort was found to be lower than our Caucasian counterparts. In consistent to other studies, aPL antibodies in Chinese cohort were also found to correlate well with clinical thrombosis in particular for LA and aβ2GPI antibodies. The low number of subjects with miscarriages did not give a big enough power to exclude a role of aPL antibodies in pregnancy morbidities in these patients.