10P - Detection of early-stage lung cancer using 5-hydroxymethylcytosine signatures in circulating cell-free DNA

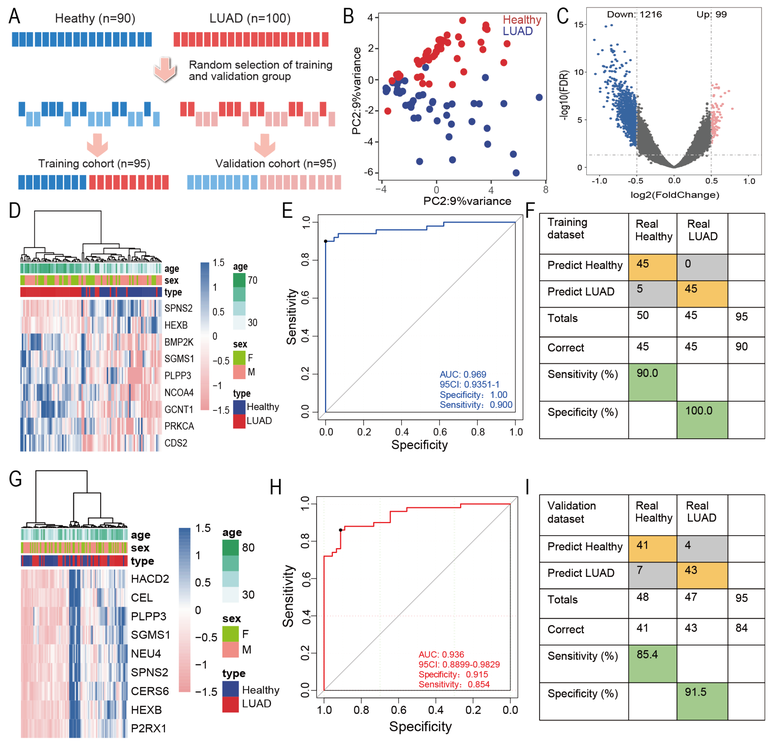
Xiaozheng Kang[1,2*], Ruiyao Ma[3*], Xiaoxiao Li[3*], Yingzhu Chen[1*], Hangyu Chen[3], Zhen Liang[1], Haitao Zhou[1], Guobing Xu[1], Chaoran Dong[4#], Jian Lin[3#]

Background:

Early lung cancer detection aims to identify the malignancy at the stage where surgical cure is possible, outcomes are superior, and treatment is less morbid. 5-Hydroxymethylcytosine (5hmC) signatures in circulating cell-free DNA (cfDNA) as diagnostic signatures have been examined in different types of cancer. However, little is known in the field of early lung cancer detection.

Methods:

We utilized well established 5hmC-Seal method (1ml plasma per patient) to map the 5hmC profiles in cfDNA from a cohort of 100 newly diagnosed early-stage lung adenocarcinoma (LUAD) patients and 90 healthy individuals. We first identified the Differentially 5hMc-enriched Regions (DhMRs) by comparing LUAD patients to healthy individuals. Then, using a machine learning algorithm, we separated samples into training (n=95) and validation (n=95) cohorts and developed a 5hmC-based diagnostic model from the training cohort to diagnose LUAD patients in the validation cohort.



The cfDNA 5hmC signatures for early LUAD diagnosis. (A) Workflow for building the diagnostic model. (B) PCA plot of 5hmC feature signatures from LUAD patients and healthy individuals in the training cohort. (C) Volcano plot (LUAD patients vs. healthy). Significantly altered hMRs (abs (log2 Foldchange) ≥0.5; FDR <0.01) are highlighted in red (up) or blue (down) using the LUAD patients as the reference. Grey dots represent the hMRs that are not differences. (D) Unsupervised hierarchical clustering of hydroxymethylation signatures between LUAD patients and healthy cfDNA in the training cohort. Each row represents an individual patient, and each column is a 5hmC marker. (E) The receiver operating characteristic (ROC) curve of the diagnostic model with 5hmC feature signatures in the training cohort for LUAD patients. (F) Confusion matrix that shows the model performance in the training cohort. (G) Unsupervised hierarchical clustering of hydroxymethylation signatures between LUAD patients and healthy cfDNA in the validation cohort. (H) The receiver operating characteristic (ROC) curve of the diagnostic model with 5hmC feature signatures in the validation cohort for LUAD patients. (I) Confusion matrix that shows the model performance in the validation cohort.

Results:

We established a diagnostic model based on nine 5hmC feature signatures, and the obtained diagnostic model discriminated patients with LUAD from healthy individuals with high accuracy (area under curve = 0.936, 95%CI: 0.890 – 0.983), high sensitivity (86.0%) and specificity (91.1%) in the validation cohort.

Conclusion:

Our findings demonstrated the usefulness of cfDNA 5hmC signatures for diagnosing LUAD, with the potential to be used for the early detection of patients with LUAD.

1. Key Laboratory of Carcinogenesis and Translational Research (Ministry of Education), Peking University Cancer Hospital & Institute, Beijing, China.
2. National Cancer Center/National Clinical Research Center for Cancer/Cancer Hospital, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing, China.
3. Department of Pharmacy, Peking University Third Hospital, Beijing, China.
4. Institute of Materia Medica, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing, China.
*Those authors contributed equally to this work.

Corresponding author:
Chaoran Dong, dongcr@imm.ac.cn, Institute of Materia Medica, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing, China.
Jian Lin, linjian@pku.edu.cn, Department of Pharmacy, Peking University Third Hospital, Beijing, China.