Metabolomic Profiling for Detection of Lung Neuroendocrine Tumors

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Disclosure Statement: R.A.B is the president and CEO of BioMark Diagnostics, Inc. and JF.H. is Chief Scientific Officer and General Manager of BioMark Diagnostic Solutions Inc. P.J. has received funding for research from Biomark Diagnostics inc.

Introduction

Pulmonary neuroendocrine tumors (NETs) represent 20% of all lung cancers and include **carcinoids** (typical (TC) and atypical (AC)) and **neuroendocrine carcinomas** (**small cells** (**SCLC**) and **large cells** (**LCNEC**)). They are commonly diagnosed late in the course of the disease mostly due to the late onset of symptoms and/or the intrinsic aggressive evolution of SCLC and LCNEC (1).

A characteristic of cancer is a **major reprogramming of cellular energy metabolism** to support cell growth and proliferation (2). Recently, **metabolites** have been considered reliable **biomarkers** (3), allowing the development of a minimally invasive routine blood test that can be used for screening as well as the monitoring of the disease evolution for patients.

Methods

Our study included a total of 120 plasma samples with biopsy-confirmed of NETs patients (breakdown of the neuroendocrine neoplasms: 50 carcinoids, 40 SCLC, 30 LCNEC), 227 of controls and 466 NSCLC patients (Figure 1) . Here, a targeted, quantitative mass spectrometry (MS) - based metabolomics approach was used to analyze the plasma samples using a combination of direct injection (DI) MS and reverse-phase high performance liquid chromatography (HPLC) tandem mass spectrometry (MS/MS). The sample set was split into a training and a testing set (Figure 2). Metabolites concentration, clinical data, and smoking history were used to determine the optimal panel of biomarkers and optimal logistic regression models.

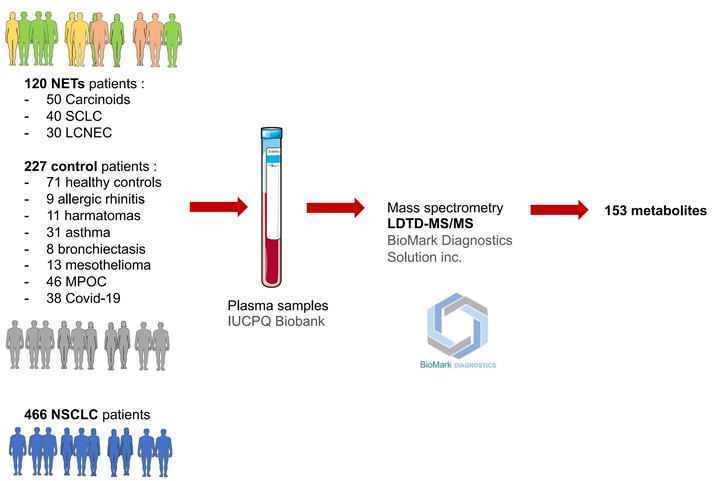

Figure 1: Methodological procedure leading to the measurement of plasma concentrations of 153 metabolites.

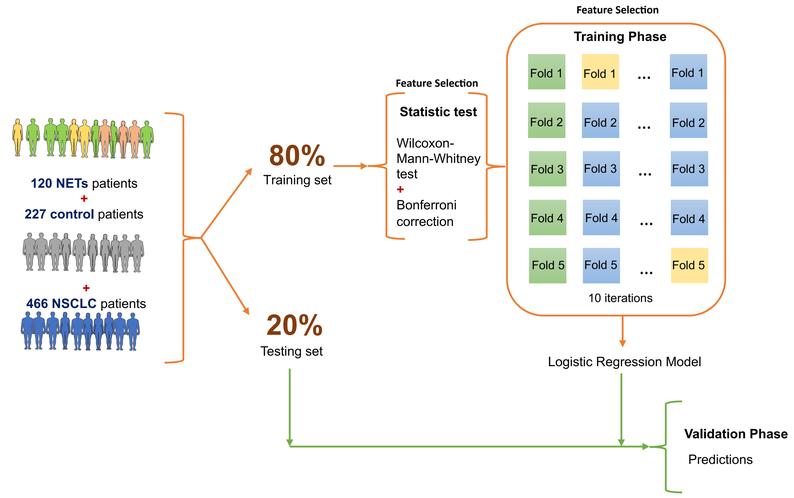

Figure 2: Study design. Separation of the cohort into training and testing sets. The training set is used for statistical tests, followed by cross validation to obtain a logistic regression model. The model is used as a prediction model on the testing set.

Results

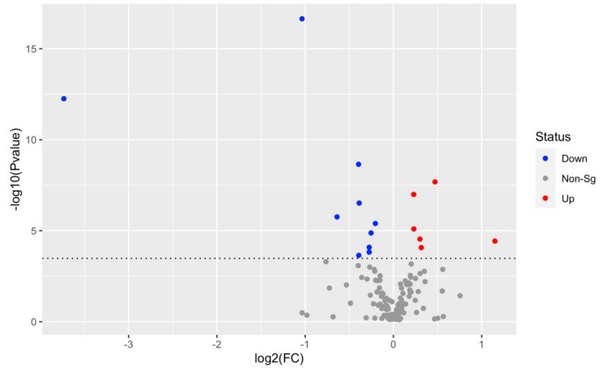

Figure 3: Difference in metabolite concentrations between lung neuroendocrine tumors and control patients. A Wilcoxon Mann Whitney test followed by a Bonferroni correction showed 10 and 6 metabolites with significantly decrease and increase concentration, respectively, in pulmonary neuroendocrine tumors.

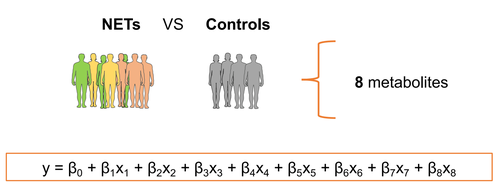

Figure 4: Logistic regression model constructed with 8 metabolites to discriminate lung neuroendocrine tumors from controls.

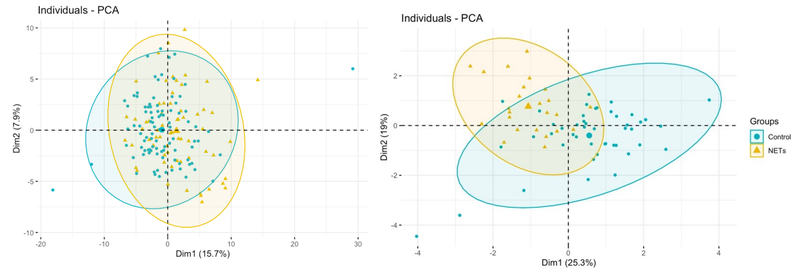

Figure 5: Contribution of metabolite panel reduction in distinguishing neuroendocrine tumors vs control group. Principal component analysis (PCA) where the first two dimensions explain 23.6% of the variance then 44.3% with the use of 153 metabolites and the panel of 8 metabolic biomarkers, respectively.

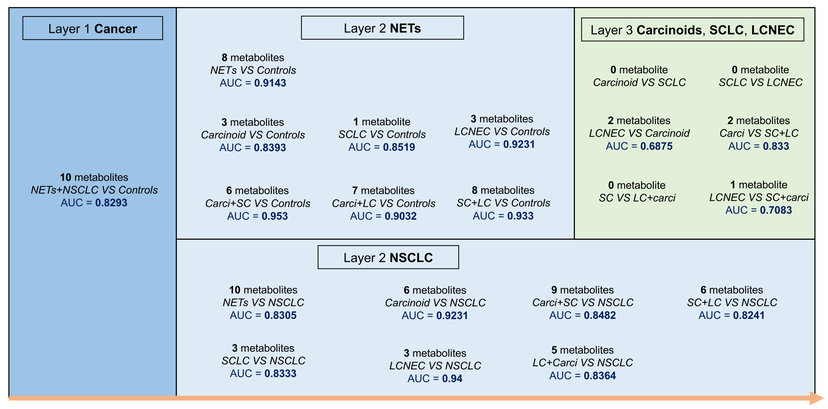

Figure 6: Summary of established metabolites panels. A Bonferroni-corrected Wilcoxon Mann Whitney test was followed by a logistic regression model construction. A cross validation was performed. The accuracy reflects the performance of the binary class prediction model on the testing set.

Conclusion

- We were able to **detect neuroendocrine tumors** from **plasma samples** with good performance (**AUC = 0.91** for NETs vs controls).
- NETs do show a **reprogramming of their metabolism** reflected by the presence of a panel of **distinct metabolic biomarkers**. It appears that **NET subtypes** can be distinguished from each other and from **NSCLC**.
- We believe that **multi-metabolites panels** could allow the implementation of a **routine screening test** for **NETs** and aid in **monitoring clinical evolution** of **neuroendocrine carcinomas**. Moreover, due to the non-specificity of the symptoms of NET patients at early stage, we believe that this test may support **early diagnosis**.

References: (1) Hung, Surgical Pathology, 2019; (2) Hanahan and Weinberg, Cell, 2011; (3) Huang et al., Nature communications, 2020

Acknowledgements: We would like to thank the **IUCPQ biobank** for providing both data & samples.