Proteotranscriptomic classification and characterization of pancreatic neuroendocrine neoplasms

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Background

Pancreatic neuroendocrine neoplasms (PNENs) are biologically and clinically heterogeneous neoplasms with variable patient outcomes. Little is known about the molecular differences between PNENs and the biological significance of such divergence. Our study aims to uncover the molecular factors that underlie the clinical and biological heterogeneity among PNENs for a better understanding and potential classification of this disease.

Experimental Approach

We used a multi-omics approach to profile and characterize the molecular landscapes of primary PNEN specimens. Formalin-fixed paraffin-embedded primary tumour specimens from 84 patients with PNEN were procured for the study and split into two cohorts for discovery (DISC) and validation (VALI) purposes. Next-generation sequencing profiled the exome and transcriptome, and quantitative mass-spectrometry profiled the global proteome of specimens. Non- negative matrix factorization was used to identify subgroups, followed by differential analysis to identify subgroup-specific features. Potential associations between the identified subgroups and known clinicopathological characteristics were evaluated.

Results

Unsupervised clustering analysis of transcriptome data identified four robust molecular subgroups that were substantiated by proteome analysis ($p=0.0005$) within the discovery cohort and confirmed with the validation cohort. A Proliferative subgroup was enriched with neuroendocrine carcinomas and specimens with >20% Ki67, concomitant with reduced survival probability ($p=0.0024$; logrank test) and higher mRNA expression and protein abundance of cell cycle-related genes. Increased mRNA expression of *ARX* or *PDX1* (adjusted $p<0.05$), similar to previous reports, was found in an Alpha cell-like subgroup and a PDX1-high subgroup, respectively. The Alpha cell-like subgroup specimens showed transcriptomic similarities to pancreatic alpha cells and exhibited enrichment of oxidative phosphorylation, while oncogenic Ras mutations were found in the PDX1-high subgroup. A fourth Stromal/Mesenchymal subgroup

exhibited enrichment of molecular features suggestive of elevated involvement of the tumour microenvironment.

Conclusions
We identified four robust molecular subgroups among PNENs with clinicopathological associations and biological distinctions that may provide potential new directions for patient stratification and treatment strategies to facilitate treatment decisions.