In tumours initially diagnosed as pNETs, DNA, RNA and histopathological evidence contributed to re-diagnosis as SPN or miNEN.

**The NETwork! Project (New Zealand)** began with creation of the NETwork! registry, which catalogues all Neuroendocrine tumour patients diagnosed between 1995-2012 in the Auckland region and 2008-2012 across New Zealand. With strong support from NET patient support group (Unicorn Foundation New Zealand), we have ethical approval to retrospectively access clinical formalin-fixed paraffin embedded (FFPE) tissue from NET patient support group (Unicorn Foundation New Zealand), we have ethical registry, which catalogues all Neuroendocrine tumour patients diagnosed between 1995-2012. We aim to conduct genomics on these annotated samples to understand NET biology, as it pertains to clinical management.

**Methods:** 69 sporadic well-differentiated pancreatic neuroendocrine tumours (pNETs) from 60 individuals alongside matched normal tissues underwent targeted DNA sequencing (NimbleGen SeqCap), RNA expression analysis (Affymetrix Microarrays) and immunohistochemistry (IHC) alongside pathological examination, to search for molecular drivers; incidentally providing valuable evidence for re-diagnosis in three patients. Cases selected had a clinical and pathological diagnosis of well-differentiated pNET, expressed at least one of the three neuroendocrine IHC markers (chromogranin A, synaptophysin or CD56) and were surgically resectable at initial diagnosis. This poster describes 3 cases that were excluded from the main analysis because genomics analyses allowed us to recognise that they were not pNETs (for pNET analysis see ENETs 2018 Poster B10).

**In two cases (029 and 048), tumours were re-diagnosed as pancreatic SPNs; originally diagnosed as pNETs by morphological and IHC but noted uncertainty due to some variable SPN-like features.**

Alongside deep pathological examination, evidence for re-diagnosis came from β-catenin mutations, RNA expression patterns, IHC (β-catenin localisation) and transcription factor function.

**Deep targeted DNA-seq** Revealed activating mutations in CTNNB1 encoding β-catenin, pathognomonic for SPNs - present in 90% of all SPN cases.

**Histopathological analysis** Revealed cellular relocalisation of β-catenin (brown) to the nucleus, concordant with SPN status and transcription factor function.

**RNA expression** RNAs up-regulated by β-catenin in wild type (red) across pNETs. Numbers indicate Z-transformed expression.

**Mutational landscape** Revealed mutations in APC, TP53 and FANCA, in contrast to mutations found in pNETs in this study.

**Aneuploidy analysis** Revealed extensive aneuploidy in tumours from patient 057 in contrast to the degree and patterns of aneuploidy seen in pNETs in this study.

**Proliferation analysis** Expression of RNAs encoding cellular proliferation proteins were distinctly elevated in tumours from patient 057 in contrast to pNETs in this study. Concordantly, these tumours had very high MKI67 RNA expression and ki-67 positive tumour cells.

**Solid Pseudopapillary Neoplasm (SPN)**

In two cases (029 and 048), tumours were re-diagnosed as pancreatic SPNs; originally diagnosed as pNETs by morphological and IHC but noted uncertainty due to some variable SPN-like features.

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**Proliferation analysis** Expression of RNAs encoding cellular proliferation proteins were distinctly elevated in tumours from patient 057 in contrast to pNETs in this study. Concordantly, these tumours had very high MKI67 RNA expression and ki-67 positive tumour cells.

**Mixed Neuroendocrine Non-Neuroendocrine Neoplasm (miNEN)**

In one case (057), three tumour samples from the same patient showed possible features of mixed neuroendocrine non-neuroendocrine neoplasm (miNEN).

Alongside deep pathological examination, evidence for re-diagnosis came from mutational landscape, aneuploidy analysis, and proliferation analysis.

**Mutational landscape** Revealed mutations in APC, TP53 and FANCA, in contrast to mutations found in pNETs in this study.

**Aneuploidy analysis** Revealed extensive aneuploidy in tumours from patient 057 in contrast to the degree and patterns of aneuploidy seen in pNETs in this study.

**Proliferation analysis** Expression of RNAs encoding cellular proliferation proteins were distinctly elevated in tumours from patient 057 in contrast to pNETs in this study. Concordantly, these tumours had very high MKI67 RNA expression and ki-67 positive tumour cells.

**Conclusions:** Homing in on precise diagnoses

- SPNs and miNENs share some cytological features with pNETs but are genomically distinct.
- β-catenin IHC should be conducted more frequently when diagnosing pNETs.
- While differentiation of uncommon pancreatic malignancies from pNETs can be challenging, it is important as the diagnosis has different prognostic implications. Genomic analysis provides a further tool for making this critical distinction.
- We believe that combining genomic information with traditional pathological information is likely to generate more precise diagnoses for many tumour types.

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1. Moreno-Bueno et al. (2011) PMID 21701285
2. Abraham et al. (2002) PMID 11344721
3. Herbst et al. (2014) PMID 24462341
4. Amarnath-Ding et al. (2014) PMID 25697910
5. Nagalla et al. (2013) PMID 23618380
6. Harris et al. (2017) PMID 27515676