MicroRNA (miRNA) Profiling Differentiates Noninvasive Follicular Thyroid Neoplasm with Papillary-like Nuclear Features (NIFTP) from Invasive Encapsulated Follicular Variant of PTC (iEFVPTC)

Sydney D. Finkelstein MD[1], Sara Jackson PhD[1], Nicole Toney MPH[1], Gyanendra Kumar PhD[2], Christina M. Narick MD[1], Lester Thompson MD[3]

[1] Interpace Diagnostics, Pittsburgh, PA; [2] Interpace Diagnostics, New Haven, CT; [3]Southern California Permanente Medical Group, Woodland Hills, CA

INTRODUCTION:

- Follicular patterned thyroid neoplasms with papillary carcinoma nuclear features, with or without invasion, are increasingly recognized as a diverse group of neoplastic processes.
- NIFTP is unlikely to undergo metastatic spread and thus treated by lobectomy.
- However, iEFVPTC, reflecting neoplastic progression with microscopic evidence of capsular invasion, is regarded as malignant.
- Mutational findings between these two closely related states does not discriminate between them.
- Using well characterized cohorts of these two entities, we show in microdissected tissue that microRNA (miRNA) profiling can effectively differentiate these entities.

METHODS:

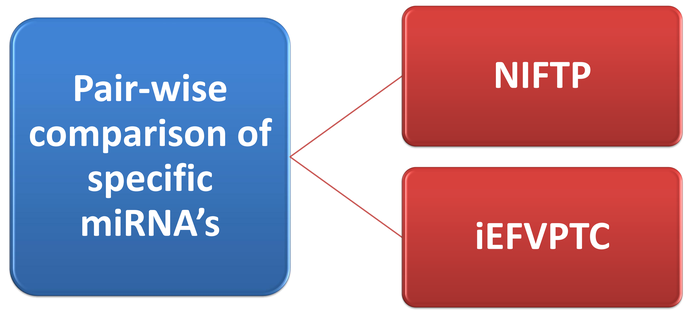
- Unstained FFPE tissue sections of known NIFTP (n=10), encapsulated follicular variant of PTC with invasion (iEFVPTC) (n=10), with positive malignant usual PTC (n=12) and negative benign (n=21) controls, were microdissected.
- Mutational analysis included common oncogene point mutation/fusion detection (ThyGenX).
- miRNA profiling included a 10 marker algorithmic classifier determination in addition to pair-wise analysis of individual miRNA expression differences (ThyraMIR).
- Significant differences in overall classifier and individual pair-wise miRNA expression were searched between NIFTP vs iEFVPTC.

Table 1. Markers in mutation panel and microRNA classifier panel

Next Generation Sequencing Mutation Panel	
BRAF	BRAF_V600E, BRAF_K601E, BRAF_A598V
HRAS	HRAS_G12V, HRAS_G13R, HRAS_Q61K, HRAS_Q61L,HRAS_Q61R
NRAS	NRAS_G12D, NRAS_G13R, NRAS_Q61R, NRAS_Q61K, NRAS_Q61P
KRAS	KRAS_G12D, KRAS_G12V, KRAS_G13D, KRAS_Q61R
PIK3CA	PIK3CA_E542K, PIK3CA_H1047L, PIK3CA_H1047R
RNA Fusion Transcripts	RET-PTC1
	RET-PTC3
	PAX8/PPARγ
10 MicroRNA (MiR) Classifier Panel	
Down-regulated	miR-204-5p, miR-139-5p, miR-29b-1-5p, miR-155-5p, miR-138-1-3p
Up-regulated	miR-375, miR-551-b-3p, miR-146b-5p, miR-31-5p, miR-222-3p

- NIFTP and iEFVPTC did not show strong driver mutational change (*BRAF V600E*)
- Weak Driver mutation status (*N,H,K RAS, PAX8/PPARγ* fusion) did not differentiate NIFTP and iEFVPTC
- NIFTP and iEFVPTC did not show miRNA classifier levels consistent with malignancy

- While the miRNA classifier status was not significantly different between t NIFTP and iEFVPTC, individual pair-wise differences in specific miRNA's among the panel of 10 miRNAs were capable of distinguishing between NIFTP vs iEFVPTC.



CONCLUSIONS:

- **The NIFTP vs iEFVPTC challenge highlights the need for molecular approaches to differentiate between closely related entities that straddle the continuum from benign and malignant states.**
- **The addition of miRNA profiling can provide sets of specific pair-wise compared miRNA markers that meet the molecular diagnostic need to distinguish between NIFTP and iEFVPTC.**