

Peptidomics Unveils Distinct Acetylation Patterns of Histone and Annexin A1 in Differentiated Thyroid Cancer

Supplementary material

Tables

Table S1. Discrimination of the thyroid nodules of the entire studied population. Diagnosis by fine-needle aspiration biopsy (FNAB) cytology [The 2023 Bethesda System for Reporting Thyroid Cytopathology (Ali, Baloch et al. 2023)] and histology diagnosis [The 2022 WHO Classification of Thyroid Neoplasms (Lloyd, Osamura et al. 2022)] of all samples, including subtypes in parenthesis. Plus, discrimination of the thyroid nodules used for HR-MAS ¹H NMR metabolomics. Abbreviations: AUS – atypia of undetermined significance; BT – benign tumours; FTC – follicular thyroid carcinoma; PTC – papillary thyroid carcinoma.

Individual	Sample	Cytology diagnosis	Histology diagnosis	HR-MAS ¹ H NMR
TR008	A	AUS	BT (Follicular adenoma)	
TR012	A	Benign	BT (Follicular nodular disease)	✓
	B	Benign	BT (Follicular nodular disease)	
TR013	A	Follicular tumour	PTC (Follicular)	✓
TR014	A	Benign	BT (Follicular nodular disease)	
	B		BT (Follicular nodular disease)	
	C		BT (Follicular nodular disease)	
TR015	A		BT (Follicular nodular disease)	
	B		BT (Follicular nodular disease)	
TR018	A	Benign	BT (Follicular nodular disease)	✓
TR019	A	Benign	BT (Follicular nodular disease)	
	B		BT (Follicular nodular disease)	
TR020	A	Follicular tumour (oncocytic)	FTC (Oncocytic)	✓
TR021	A	Benign	BT (Follicular adenoma)	✓
	B	AUS	BT (Follicular nodular disease)	
TR023	A	Benign	BT (Follicular nodular disease)	
TR025	A		BT (Follicular nodular disease)	
	B		BT (Follicular nodular disease)	
	C		BT (Follicular nodular disease)	
TR026	A	Follicular tumour	BT (Follicular nodular disease)	✓
	B		BT (Follicular nodular disease)	
TR028	A	Follicular tumour	PTC (Follicular and solid)	✓
TR029	A	Benign	BT (Follicular nodular disease)	
TR030	A		BT (Follicular nodular disease)	
TR031	A	Benign	BT (Follicular nodular disease)	✓
	B		BT (Follicular nodular disease)	
TR032	A	Follicular tumour	PTC (Follicular)	✓
TR034	A	Papillary carcinoma	PTC (Classical and follicular)	✓
	B		PTC (Classical and follicular)	
TR035	A	Benign	BT (Follicular nodular disease)	
TR037	A		BT (Follicular nodular disease)	✓
	B		BT (Follicular nodular disease)	
	C		BT (Follicular nodular disease)	
	D		BT (Follicular nodular disease)	
TR038	A	Non-diagnostic	BT (Follicular adenoma)	✓
	B		BT (Follicular nodular disease)	
TR039	A	Benign	BT (Follicular nodular disease)	✓
TR040	A	Benign	BT (Follicular nodular disease)	✓
	B		BT (Follicular nodular disease)	
TR041	A	Benign	BT (Follicular nodular disease)	
TR043	A	Benign	BT (Follicular nodular disease)	
	B		BT (Follicular nodular disease)	
	C		BT (Follicular nodular disease)	
	D		BT (Follicular nodular disease)	
TR045	A	Non-diagnostic	BT (Follicular nodular disease)	
	B		BT (Follicular nodular disease)	
TR046	A	Benign	BT (Follicular nodular disease)	
	B	Benign	BT (Follicular nodular disease)	
TR047	A	Benign	BT (Follicular nodular disease)	✓
	B		BT (Follicular nodular disease)	
TR048	A	Benign	BT (Follicular nodular disease)	✓
TR049	A	Benign	BT (Follicular nodular disease)	

	TR050	A	Papillary carcinoma	PTC (Classical and follicular)	✓
	TR051	A	Papillary carcinoma	PTC (Classical)	✓
	TR052	A	Benign	BT (Follicular nodular disease)	
		B	Benign	BT (Follicular nodular disease)	
	TR053	A	Papillary carcinoma	PTC (Classical)	✓
	TR054	A	Papillary carcinoma	PTC (Diffuse sclerosing)	✓
	TR055	A	Papillary carcinoma	PTC (Classical)	✓
	TR056	A	Benign	BT (Follicular nodular disease)	✓
		B	Benign	BT (Follicular nodular disease)	
	TR057	A	Non-diagnostic	BT (Follicular nodular disease)	✓
	TR058	A	Non-diagnostic	BT (Follicular nodular disease)	
		B		BT (Follicular nodular disease)	
	TR059	A	Benign	BT (Follicular nodular disease)	
	TR060	A	Benign	BT (Follicular nodular disease)	
		B	Benign	BT (Follicular nodular disease)	
		C	Benign	BT (Follicular nodular disease)	
	TR061	A	Benign	BT (Follicular nodular disease)	
		B	Benign	BT (Follicular nodular disease)	
	TR063	A	Oncocytic adenoma	BT (Oncocytic adenoma)	
Individuals	43		Non-diagnostic	4	Inadequate or different from cytology
			AUS	2	
		8 (15.7%)			

In the sample column different letters represent different nodules studied for the same individual.

Table S2. Top 20 statistically different proteins and peptides with fold-change (FC), p-value from the Mann-Whitney test and adjusted p-value based on FDR (Adj p-value).

Protein	FC	p-value	Adj p-value
P04083 ANXA1	2.44	2.74E-07	1.48E-04
Q9UKK3 PARP4	2.25	5.80E-07	1.48E-04
P08727 K1C19	4.24	5.80E-07	1.48E-04
P50995 ANX11	1.58	5.80E-07	1.48E-04
Q9UPN3 MACF1	1.28	7.41E-07	1.48E-04
Q9NZM1 MYOF	1.44	7.42E-07	1.48E-04
P06753 TPM3	1.7	8.05E-07	1.48E-04
Q15149 PLEC	1.7	8.05E-07	1.48E-04
Q99536 VAT1	1.09	8.05E-07	1.48E-04
P02763 A1AG1	-3.02	8.73E-07	1.48E-04
O60831 PRAF2	1.41	9.08E-07	1.48E-04
Q6UX53 MET7B	4.21	9.46E-07	1.48E-04
Q9BUF5 TBB6	1.35	9.47E-07	1.48E-04
Q14195 DPYL3	2.17	1.11E-06	1.48E-04
Q9BTV4 TMM43	1.95	1.21E-06	1.48E-04
Q9BSJ8 ESYT1	1.36	1.21E-06	1.48E-04
P46821 MAP1B	2.09	1.53E-06	1.48E-04
Q9P2R3 ANFY1	0.92	1.66E-06	1.48E-04
Q13509 TBB3	2.98	1.66E-06	1.48E-04
P02545 LMNA	1.28	1.66E-06	1.48E-04

Peptides	FC	p-value	Adj p-value
P16402 H13 [1Ac]-SETAPLAPTIPAPAEKTPVKK	4.88	1.31E-06	1.16E-04
P10412 H14 SETAPAAPAAPAPAEKTPVK	3.22	2.27E-06	1.16E-04
P08670 VIME FGGPGTASRPSS	2.59	3.10E-06	1.16E-04
P10412 H14 [1Ac]-SETAPAAPAAPAPAEKTPVKK	3.72	3.10E-06	1.16E-04
P16403 H12 [1Ac]-SETAPAAPAAAPAEKAPVKK	3.16	6.63E-06	1.98E-04
Q9Y3U8 RL36 ALRYPMAY	2.70	8.29E-06	2.05E-04
O00479 HMG4 DASTLQSQKAEGTGDAK	2.07	9.62E-06	2.05E-04
Q5VTE0 EF1A3 [1Ac]-GKEKTHINIVVIGHVDSGKST	3.73	1.98E-05	3.69E-04
P16403 H12 [1Ac]-SETAPAAPAAAPAEK	3.79	2.82E-05	4.32E-04
P10412 H14 [1Ac]-SETAPAAPAAPAPAEKTPVKKK	2.73	3.03E-05	4.32E-04
Q9BX97 PLVAP GLAMEHGGSYA	-2.47	3.25E-05	4.32E-04
P04083 ANXA1 [1Ac]-AMVSEFLKQ	3.90	3.48E-05	4.32E-04
P16402 H13 [1Ac]-SETAPLAPTIPAPAEKTPVKKK	3.76	3.99E-05	4.55E-04
P10809 CH60 AKDVKFGADARALMLQGVDLLADAVAV	2.61	4.28E-05	4.55E-04
Q5VTE0 EF1A3 IGGIGTVPVGRVETGVLPKPG	3.19	5.25E-05	5.21E-04
P02671 FIBA SQLQKVPPEWKALTDMPQM	-2.60	6.87E-05	5.76E-04
P60709 ACTB FAGDDAPRAVFPISIVGRPR	4.91	7.35E-05	5.76E-04
P47914 RL29 AAAPASVPAQAPKR	3.15	7.35E-05	5.76E-04
P16402 H13 [1Ac]-SETAPLAPTIPAPAEK	2.71	7.35E-05	5.76E-04
P08729 K2C7 [1Ac]-SIHFSSPVFTSRSA	-2.59	0.000116	8.67E-04

Figures

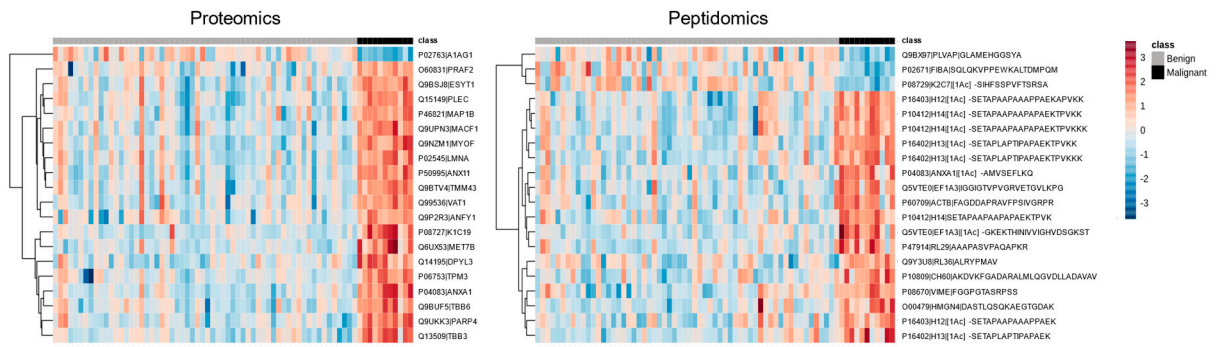


Figure S1. Heatmap with hierarchical clustering of top 20 statistically different proteins (left panel) and peptides (right panel). Euclidean distance measure and ward clustering method. Proteins are represented by their Uniprot accession number and entry name. Peptides are identified by their protein the same way, plus the one code amino acid sequence.

References

- Ali, S. Z., Z. W. Baloch, B. Cochand-Priollet, F. C. Schmitt, P. Vielh and P. A. VanderLaan (2023). "The 2023 Bethesda System for Reporting Thyroid Cytopathology." *Thyroid*.
- Lloyd, R., R. Osamura and J. Rosai (2022). *WHO Classification of Tumours Editorial Board. Endocrine and Neuroendocrine tumours*, Lyon: IARC.