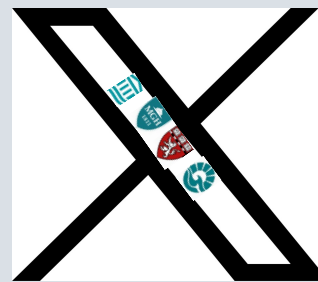


Morphological Predictors of Actionable Thyroid Neoplasia: Rational Approach to Selective Immunohistochemistry & Molecular Testing

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Massachusetts General Hospital

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Harvard Medical School



@PathDocBoston

Disclosure

- No relevant financial disclosures

Agenda

- WHO 2022/2023 – Endocrine Neoplasia
- Follicular thyroid tumors
 - Update in Diagnosis
- Junction of Architecture and Genetics
- Molecular Era of Thyroid Carcinoma
 - Evolving Algorithms
 - Molecular Diagnostics in Thyroid Pathology
 - Molecular Diagnostics in Thyroid Prognosis
 - Molecular Diagnostics in Thyroid Carcinoma Treatment



Background

- Thyroid cancer is frequently “over-diagnosed”
 - Analogous to prostatic adenocarcinoma
- Increased incidence is likely due to increased surgeries & reporting of papillary thyroid carcinomas along with incidental carcinomas
- Diagnosis historically based on nuclear features and not necessarily architecture
 - “Papillary” is an architectural pattern
 - Papillae not needed for the diagnosis (cytology)

Background

- Clinical treatment has been moving in the direction of more conservative, thyroid-sparing therapies
- Conservative therapies have been driven, in part, on outcomes data along with expansion of predictive modalities
- Genetics playing an increased role in both diagnostics, prognostics -> therapeutics

WHO 2022 – Endocrine Tumors



- New edition (5th) came out online
- **Endocrine Tumours**
 - <https://tumourclassification.iarc.who.int/welcome/#>
 - Beta version ahead of print
 - Prepared by 160 authors and editors
Contributors from around the world
 - Includes:
 - 1152 high-quality images
 - 65 whole slide images

WHO 2022 – Endocrine Tumors



- Guidelines are not free of bias
- Guidelines are the highly curated opinions of those writing them, as inherently, most subject matter authors have their own experience from which they have derived their own conclusions
- Pointed out for our viewers just in case you are frustrated by any changes – in some cases, like fashion trends

WHO 2022 – Endocrine Tumors



- Organization
 - Benign Tumors – FA, FA-P, OA, TFND
 - Low-Risk tumors
 - UMPs
 - NIFTP
 - Hyalinizing Trabecular Tumor
 - The Weird Ones
 - Salivary
 - Thymic
 - Thyroblastoma

WHO 2022 – Endocrine Tumors



- Changes (major)
 - Uncertain malignant potential
 - **FT-UMP** (follicular tumor)
 - FTA vs FTC
 - **WDT-UMP** (papillary-like features)
 - NIFTP vs PTC

References:

- Ito Y et al. *Endocr J.* 2022 Jul 28;69(7):757-761. PMID: 35082189.
- Kakudo K. *Gland Surg.* 2018 Aug;7(Suppl 1):S8-S18. PMID: 30175059

WHO 2022 – Endocrine Tumors



- Changes (major)
 - Poorly differentiated thyroid carcinoma
 - **Differentiated high-grade thyroid carcinoma**
 - **High-grade differentiated follicular cell-derived carcinoma**
 - Able to subtype as FTC, OTC, or PTC
 - **Poorly differentiated thyroid carcinoma**
 - Original Turin criteria

References:

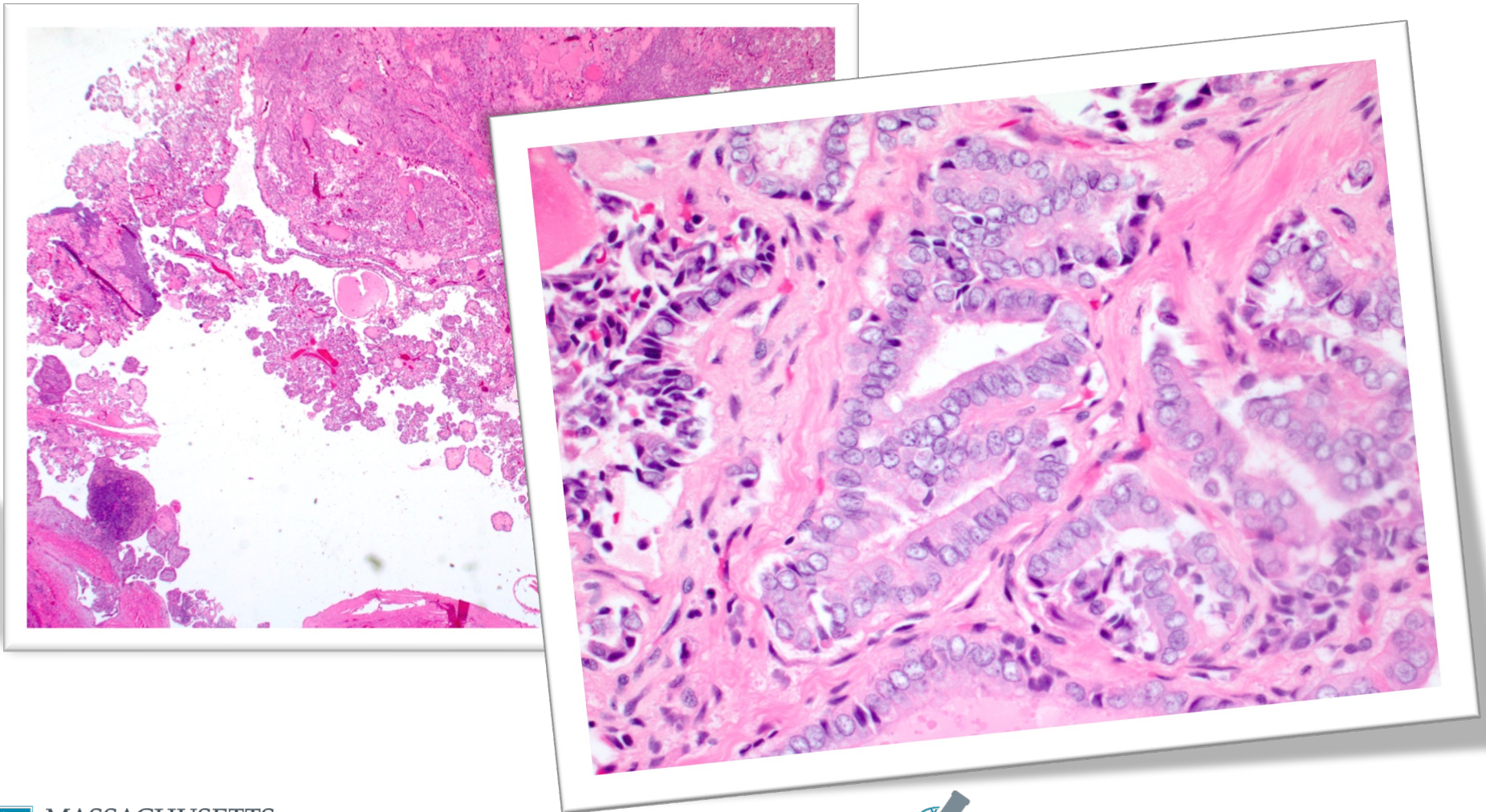
- Thompson LDR. Endocr Pathol. 2023 May 17. Epub ahead of print. PMID: 37195480.
- Cracolici V. Surg Pathol Clin. 2023 Mar;16(1):45-56. PMID: 36739166.
- Wong KS et al. Thyroid. 2021 Jun;31(6):933-940. PMID: 33143568.

WHO 2022 – Endocrine Tumors

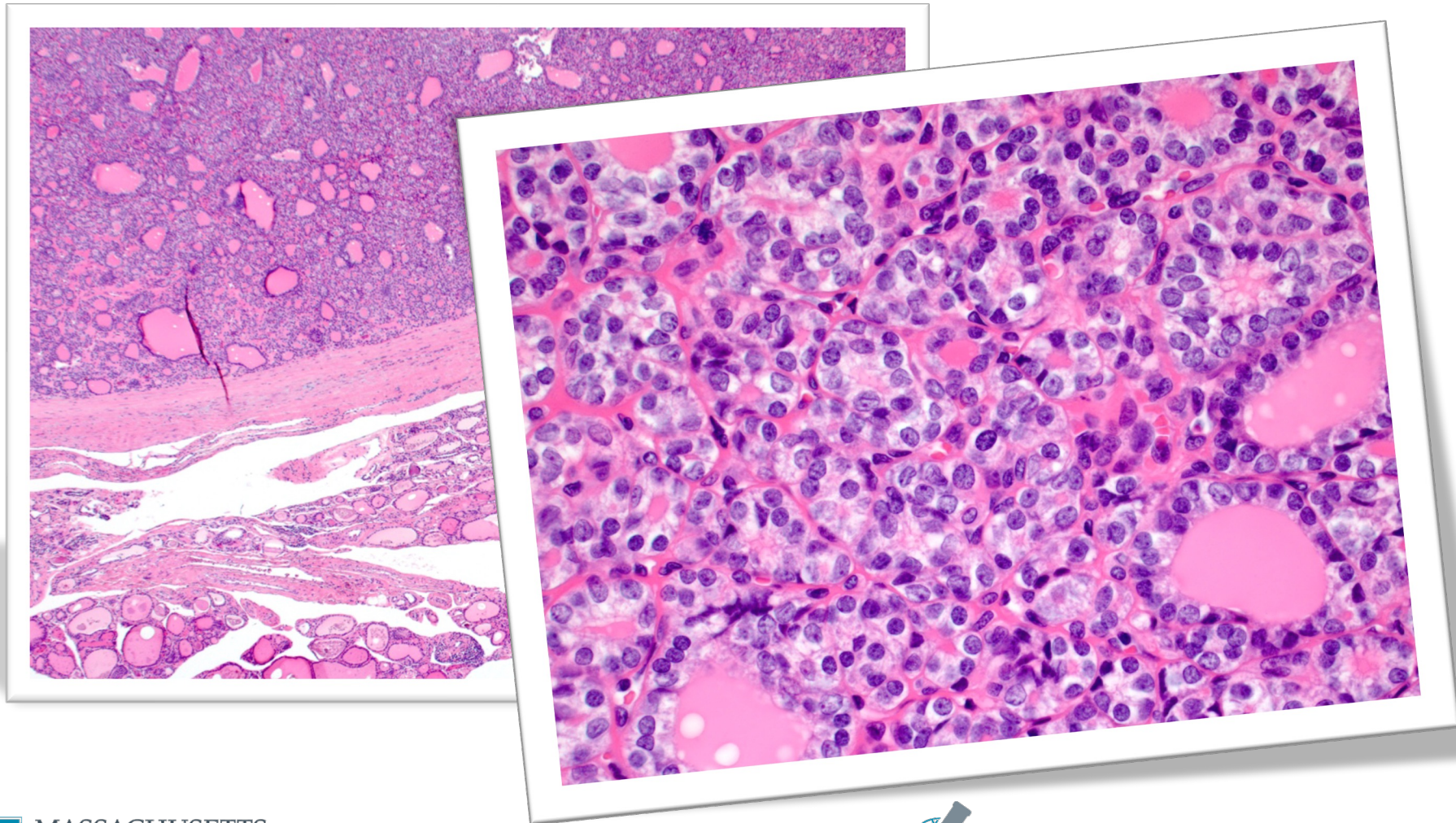


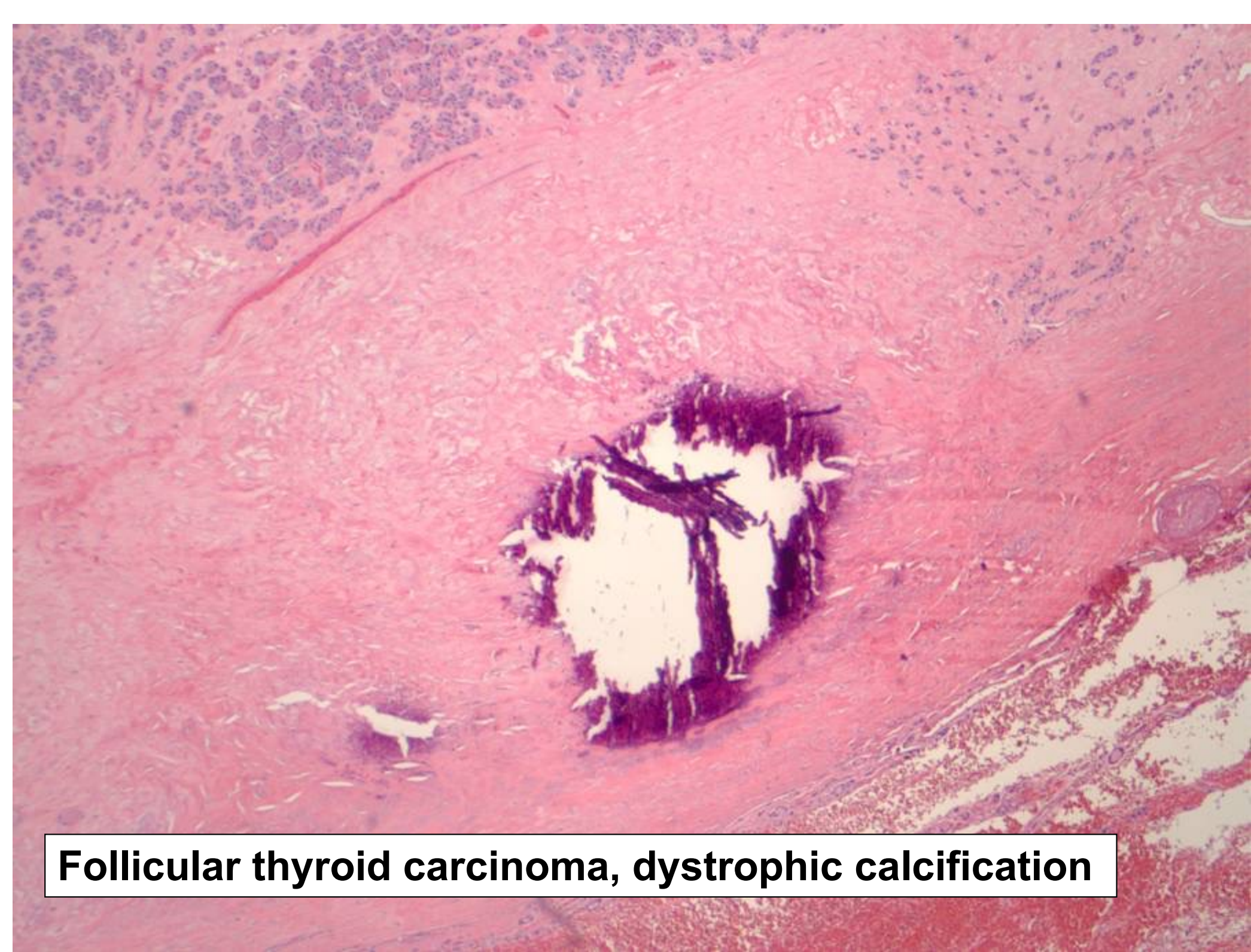
- Changes (minor)
 - **Hürthle cell** is out and **oncocytic** is back
 - **Thyroid Follicular Nodular Disease**
 - Bye-bye multinodular goiter
 - Nodules may be clonal and thus the re-name
 - Variants
 - No comma variant (PTC, classical type)
 - Now as prefix, ie. Classic Papillary Thyroid Carcinoma, Tall Cell Papillary Thyroid Carcinoma

Classic Papillary Thyroid Carcinoma

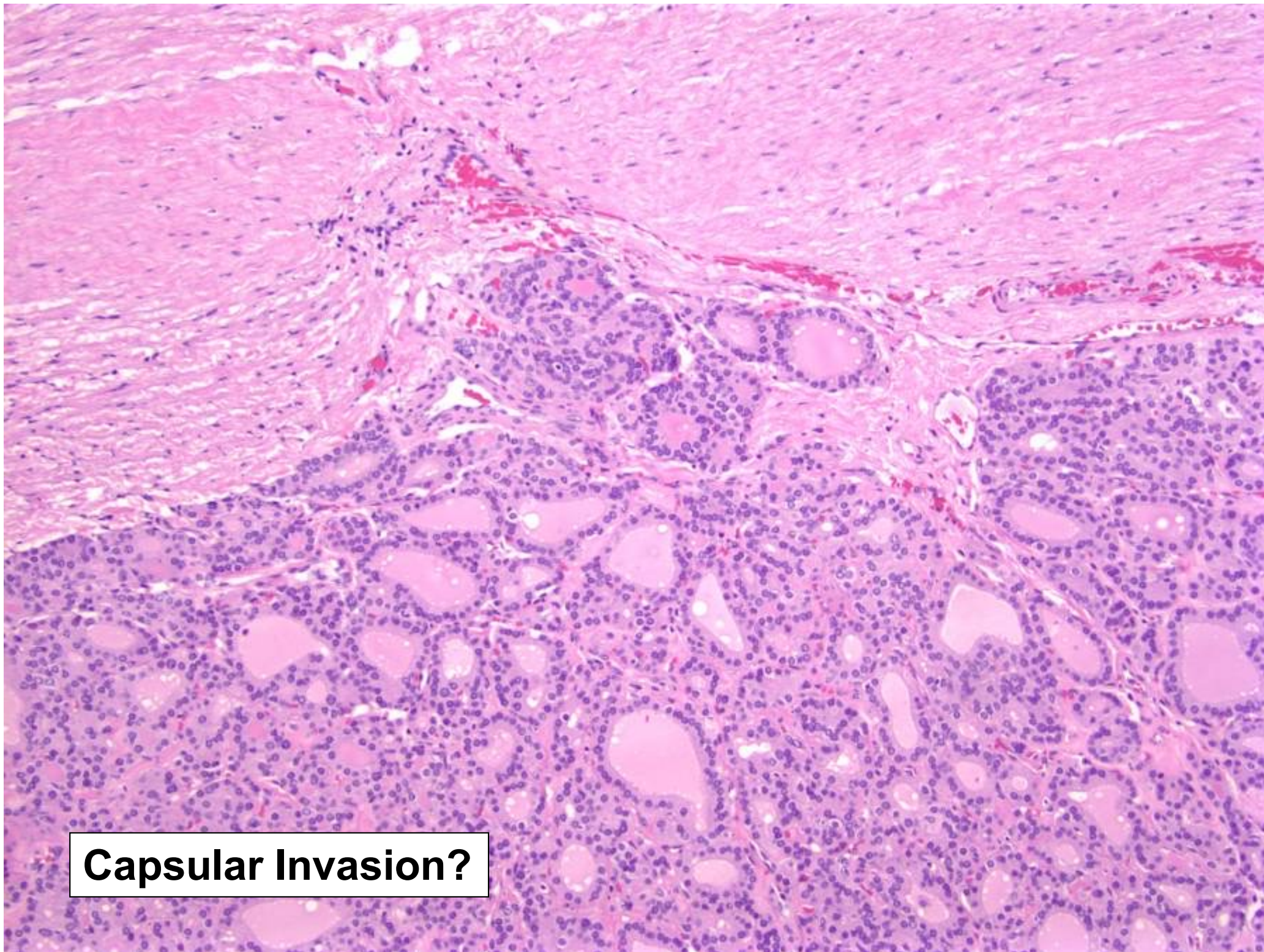


Encapsulated Follicular Variant Papillary Thyroid Carcinoma

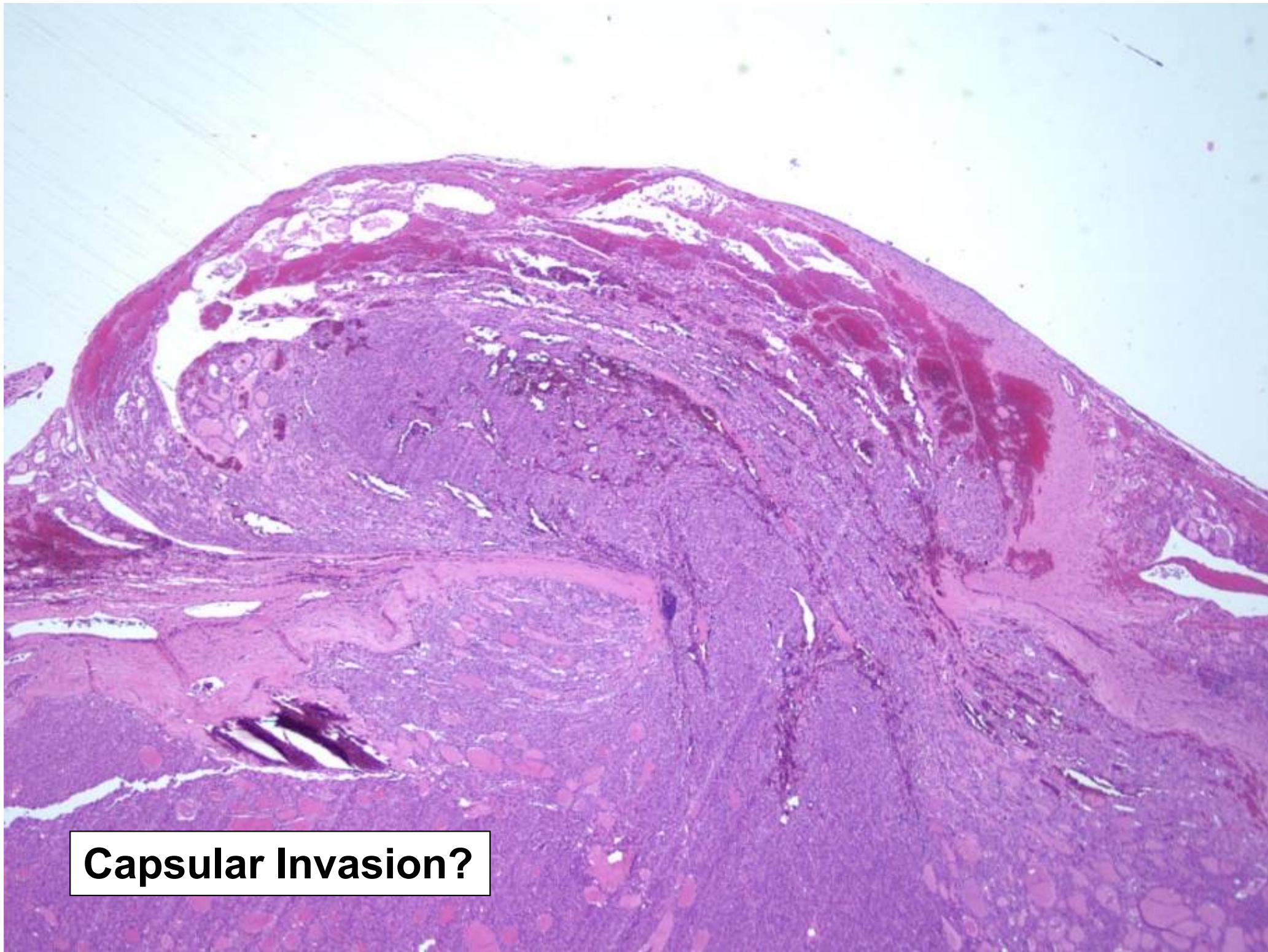




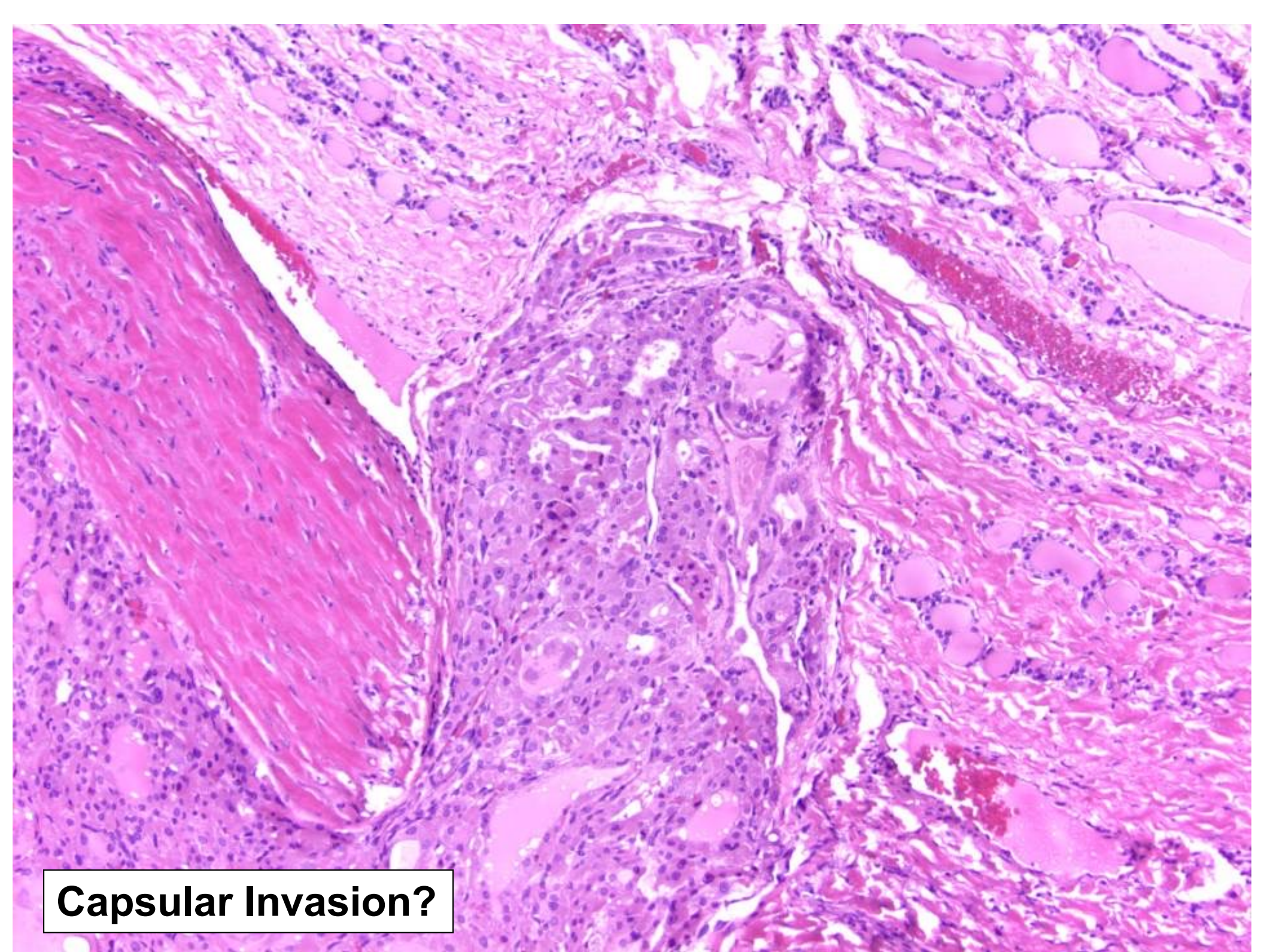
Follicular thyroid carcinoma, dystrophic calcification



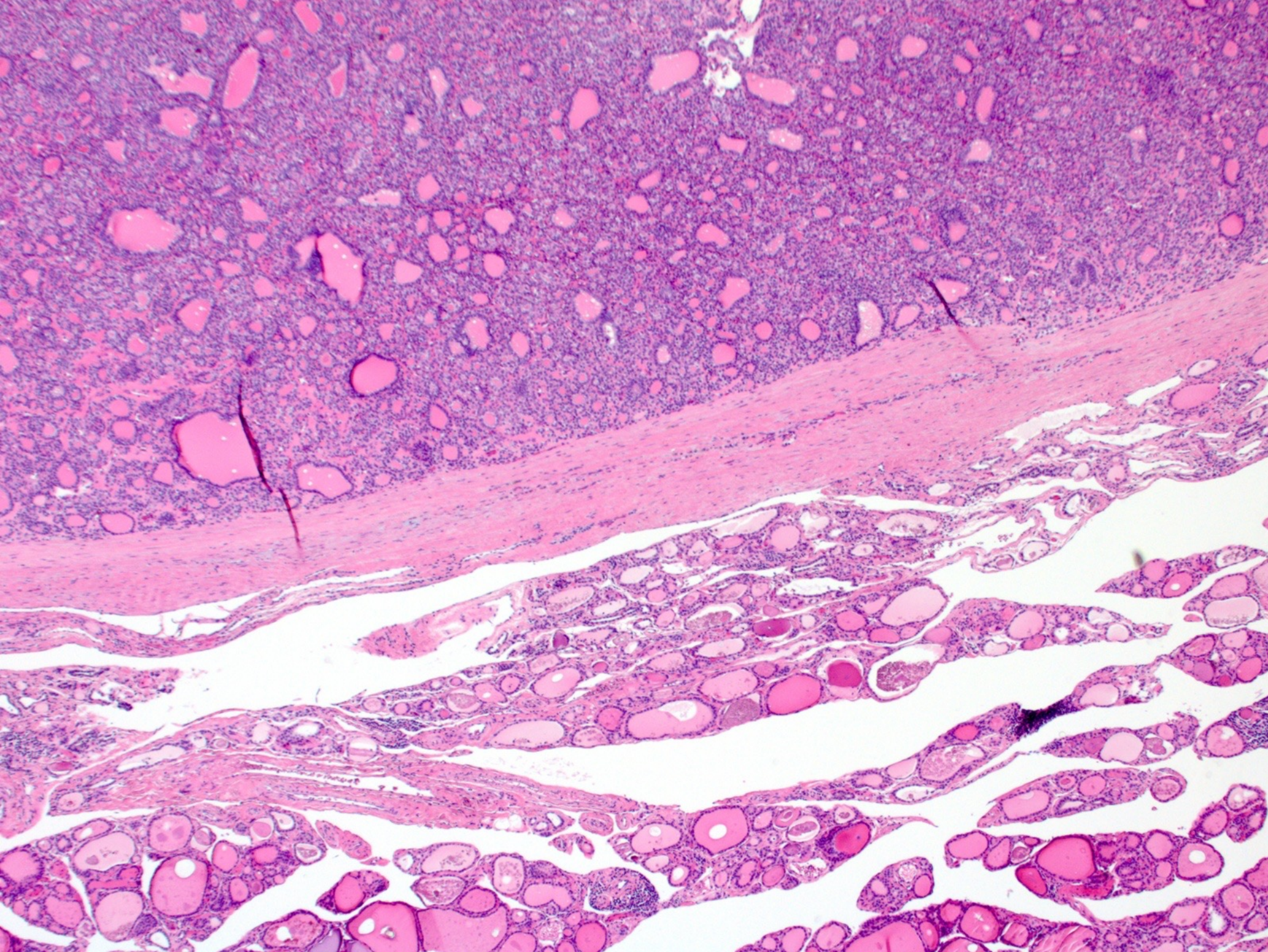
Capsular Invasion?



Capsular Invasion?



Capsular Invasion?



Encapsulated Follicular PTC

Historical View

- Diagnosis based solely upon nuclei
- Capsular invasion - *unnecessary*
- Vascular invasion - *unnecessary*
- Architectural papillae - *unnecessary*
- Cytology Diagnosis:
 - AUS/FLUS (Atypia of Undetermined Significance/
Follicular Lesion of Undetermined Significance)
 - Suspicious for papillary thyroid carcinoma
 - Lack architectural and some nuclear features



Follicular PTC (E/NI)

- Since only nuclear changes needed for diagnosis, broad variability in its rendering
- Leads to over diagnosis with no consensus
 - Not necessarily *mis*-diagnosis
- Frustration among surgeons & endo docs
 - high *intra*-institutional & *extra*-institutional discordance

Over-diagnosis

- Over diagnosis \neq *Misdiagnosis*
- More tumors being discovered due to increased screening
- Subjectivity in diagnosis of FPTC
 - Pathologists (especially low-volume centers) afraid to “under-call”
- More surgeries (historically)
- No recurrence

Predictors of Behavior

- Alexander EK, et al. Preoperative diagnosis of benign thyroid nodules with indeterminate cytology. N Engl J Med. 2012 Aug 23;367(8):705-15.
 - “Masked” genetic test with binary outcome (benign/suspicious)
- Nikiforova MN, et al. Targeted next-generation sequencing panel (ThyroSeq) for detection of mutations in thyroid cancer. J Clin Endocrinol Metab. 2013 Nov;98(11):E1852-60.
 - Limited panel of 12 genes and 284 hot-spots



One Pivotal Publication

One Rational Commentary

- Cancer Genome Atlas Research Network. Integrated genomic characterization of papillary thyroid carcinoma. *Cell*. 2014 Oct 23;159(3):676-90.
 - **Includes follicular (variant) papillary thyroid carcinomas**
- Asa SL, Giordano TJ, LiVolsi VA. Implications of the TCGA genomic characterization of papillary thyroid carcinoma for thyroid pathology: does follicular variant papillary thyroid carcinoma exist? *Thyroid*. 2015 Jan;25(1):1-2.
 - ***BRAF*^{V600E}-like & *RAS*-like PTC**



Cancer

- ***Definition:*** National Cancer Institute of the US National Institutes of Health:
- Cancer is a disease in which some of the body's cells grow uncontrollably and ***spread to other parts of the body.***

Rational Therapeutics

- Revelation of genetic pathways corresponding with biological behavior
- Need for revision of standard approach to thyroid cancer
 - Total thyroidectomy plus radioactive iodine (several tumors which do not even respond)

Thyroid; Vol 26(1); 2016, p31-32 – Recommendation 35B)



ATA Guidelines (2016)

- “For patients with thyroid cancer > 1 cm and < 4 cm *without extrathyroidal extension**, and without clinical evidence of any *lymph node metastases (cN0)*, the initial surgical procedure can be either a bilateral procedure (near-total or total thyroidectomy) or a *unilateral procedure* (lobectomy). Thyroid **lobectomy alone may be sufficient initial treatment for low-risk papillary and follicular carcinomas**; however, the treatment team may choose total thyroidectomy to enable RAI therapy or to enhance follow-up based upon disease features and/or patient preferences.”



ATA Guidelines (2023)

(likely 2024) 🤔

- Guidelines currently in revision
- Additional **ATA Position Statement Regarding Molecular Testing in Thyroid Nodules**
 - Co-chair: Peter Sadow, MassGeneral Pathology
 - Co-chair: Masha Livhits, UCLA Endocrine Surgery
- **Goal:** Thorough review of literature of current modalities, with regard to efficacy, preoperative/postoperative utility, accounting for variably resourced areas, with **rational use recommendations**

Endocrine Pathology Society Working Group - EFVPTC

- 7 countries & 4 continents
- 24 thyroid pathologists
- 2 endocrinologists
- 1 surgeon
- 1 psychiatrist
- 1 thyroid cancer patient
- 1 statistician
- 1 molecular pathologist



Endocrine Pathology Society Working Group - EFVPTC

- 268 cases reviewed
- Independent review of cases by the 24 pathologists with strict developed criteria
- Reviewed over 8 weekly teleconferences
- Consensus in Boston, March 20-21, 2015, in conjunction with annual USCAP meeting
- Initial review blinded to participants with molecular findings – subsequently revealed to be largely *RAS*

Endocrine Pathology Society Working Group - EFVPTC

- Of 109 cases of patients with non-invasive disease meeting criteria of the group, with 10-26 years of follow up, **ALL ALIVE WITH NO EVIDENCE OF DISEASE!!**
- **CONCLUSION:** Nomenclature revision needed
- Revision from: Encapsulated follicular variant of papillary thyroid carcinoma

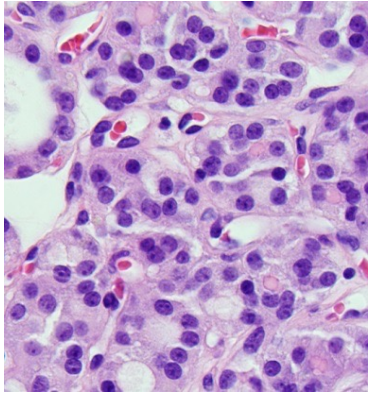


EFVPTC

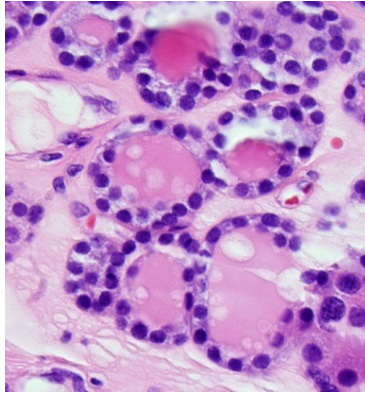
- Main morphological features
 - Follicular growth pattern & PTC nuclear features
- Lack of invasion, distinguishing from invasive EFVPTC
- Clonal origin with a driver mutation, indication the lesion is neoplastic
- Very low risk of adverse outcome

Nuclear Features Score

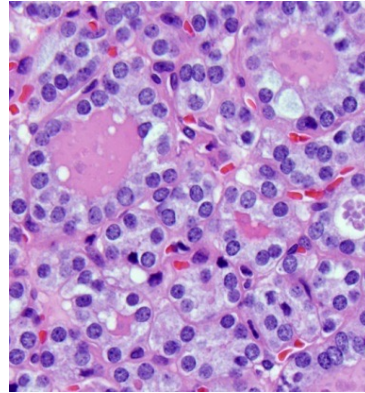
Score 0



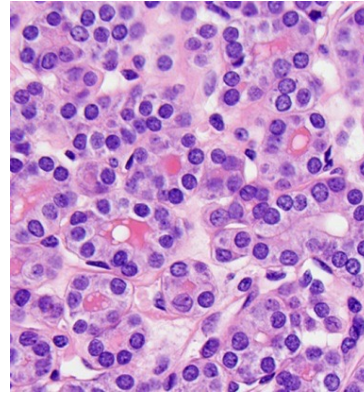
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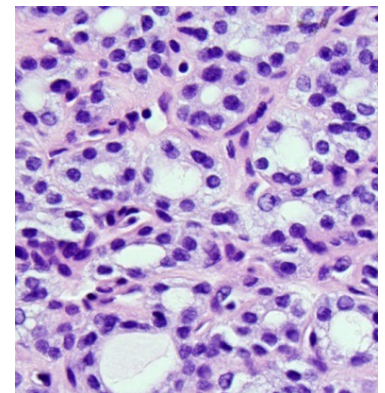
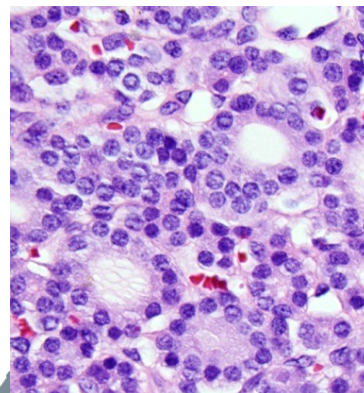
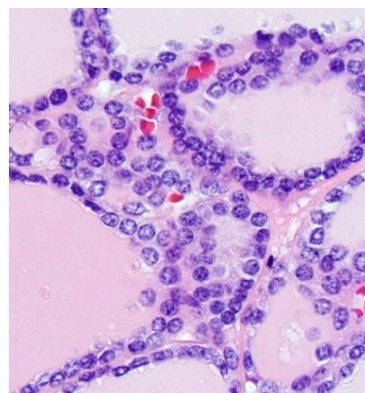
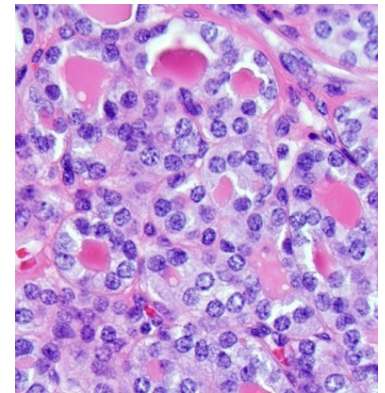
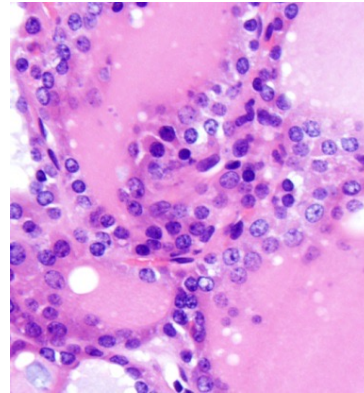
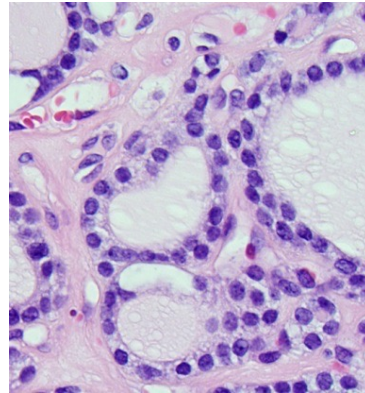
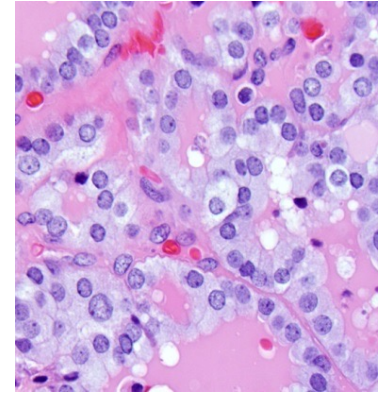
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3



4



Nuclear Features Score

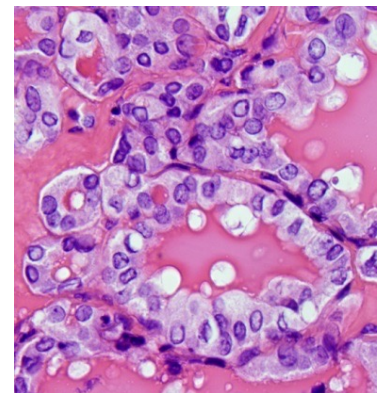
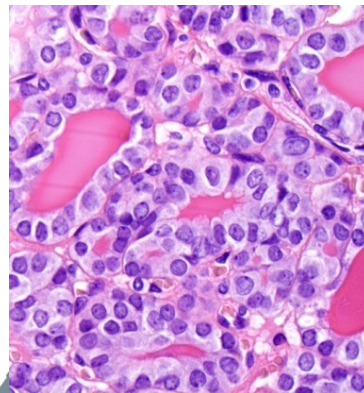
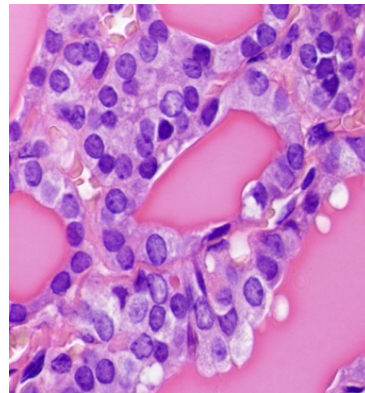
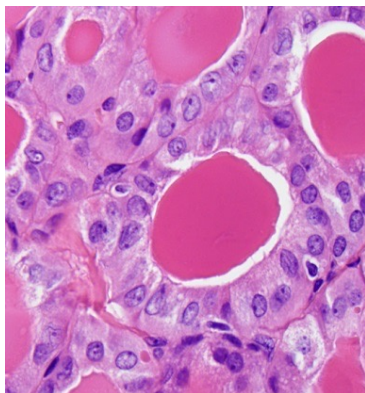
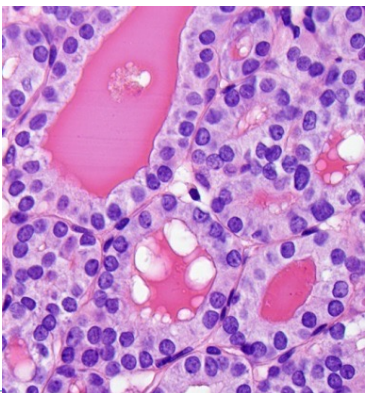
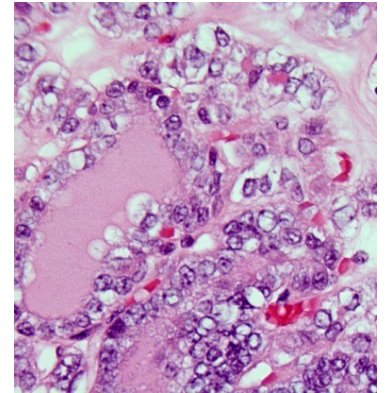
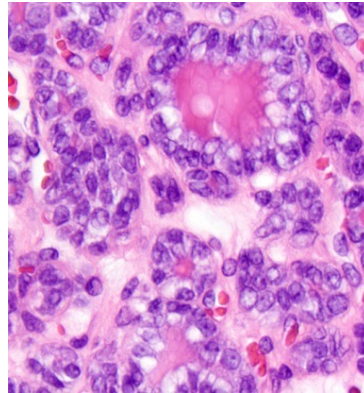
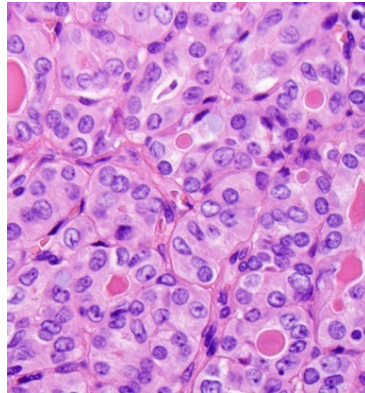
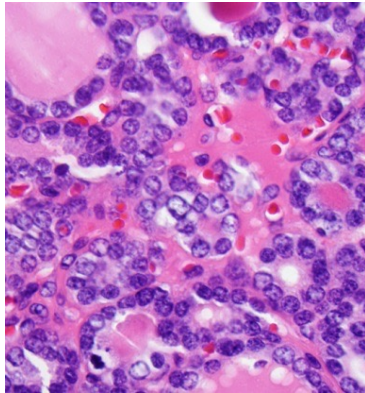
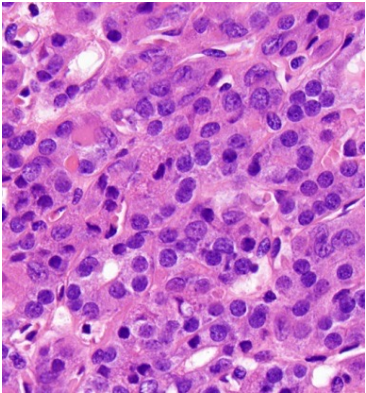
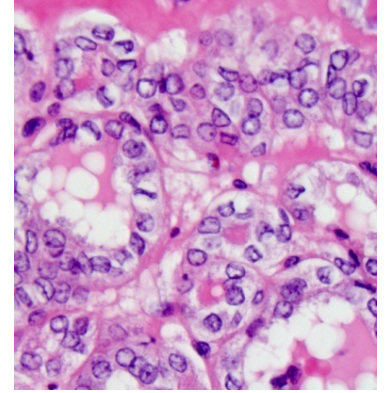
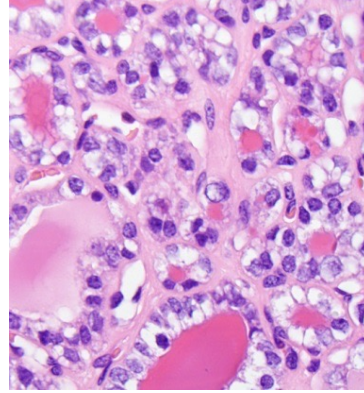
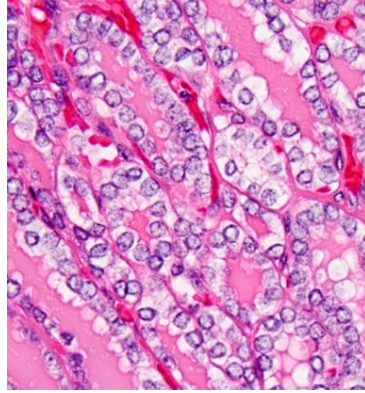
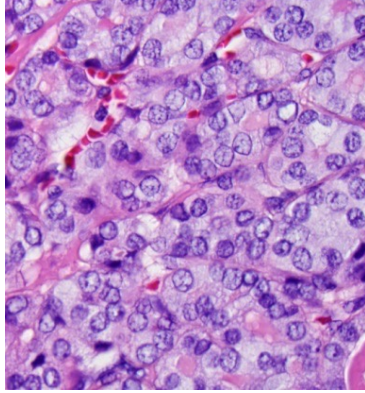
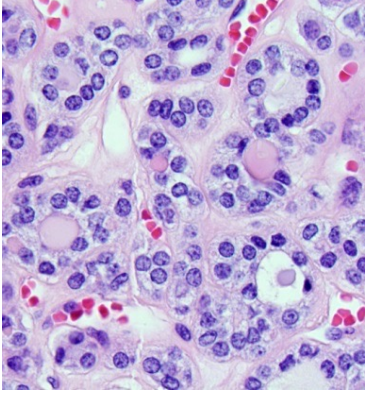
Score 5

6

7

8

9



Nuclear Score 0-3

Nuclear features:

1. Size and Shape

- Enlargement
- Elongation
- Overlapping

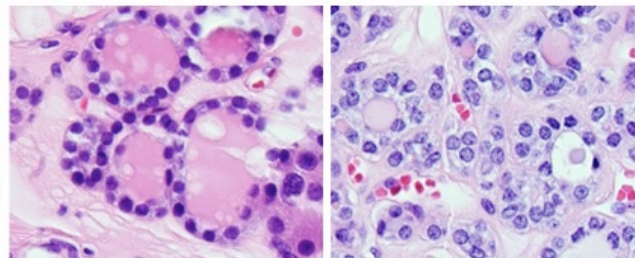
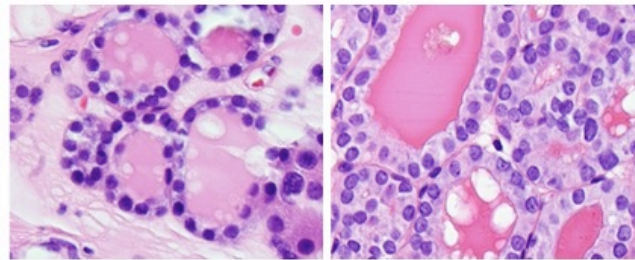
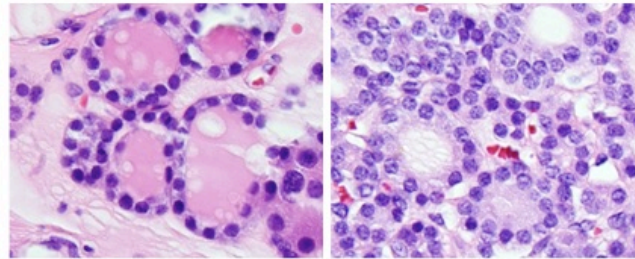
2. Membrane Irregularities

- Irregular contours
- Grooves
- Pseudoinclusions

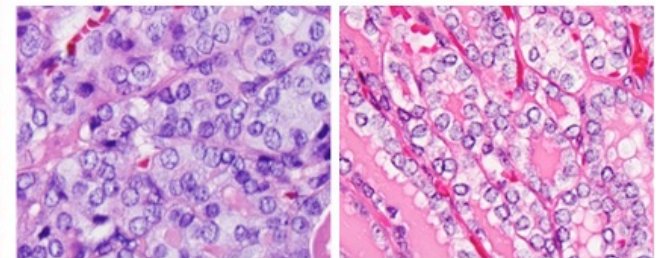
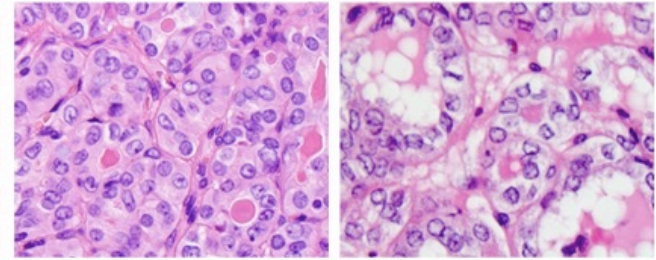
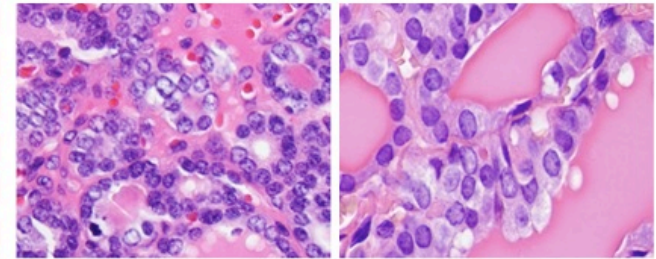
3. Chromatin Characteristics

- Chromatin clearing
- Margination of chromatin to membrane
- Glassy nuclei

Absent/insufficiently expressed (0)



Present/Sufficient (1)



Slight changes not sufficient to call "yes"

Nomenclature

- Nomenclature discussion was extensive
- Non-invasive follicular thyroid neoplasm with papillary-like nuclear features (NIFTP)
- Non-invasive EFVPTC no longer classified as malignant (WDT-UMP)

Nikiforov et al JAMA Oncology, Aug 2016

New Wastebasket

- Nuclear scoring system was set up for consistency (not used)
- Range of ***nuclear*** scores would deem the lesion benign (0) or NIFTP (not zero)
- Interestingly, the diagnosis does seem to be getting aggressively abused

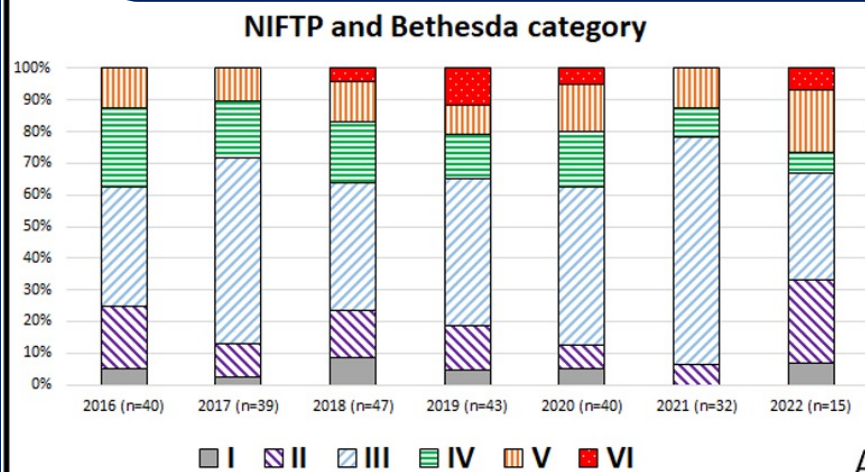
Follow-up Studies Beginning

- Taneja C et al. Clinicopathologic Characteristics and Postsurgical Follow-Up of Noninvasive Follicular Thyroid Neoplasm with Papillary-Like Nuclear Features in the Postnomenclature Revision Era. *Thyroid*. 2022 Nov;32(11):1346-1352. PMID: 35876443.
 - 79 patients, up to 75% *RAS* (100% *RAS*-like), 2.4 years follow-up (up to almost 6 years)
 - No recurrence (no additional treatment, most lobectomy)

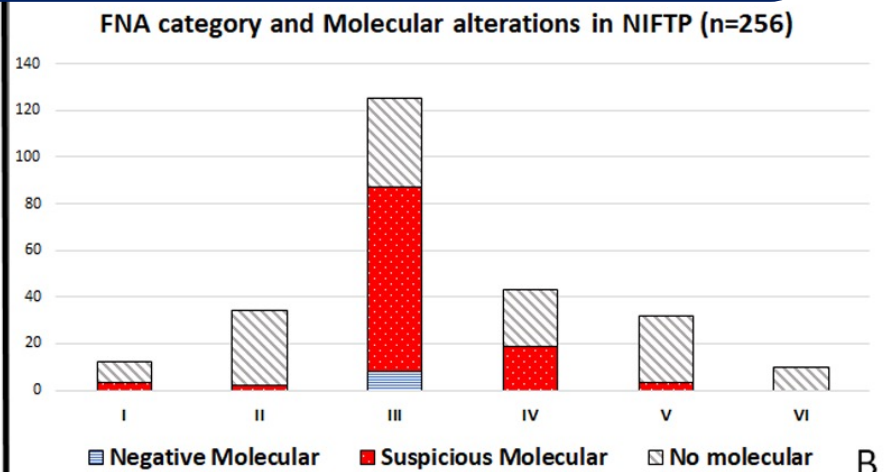


Follow-up Studies Beginning

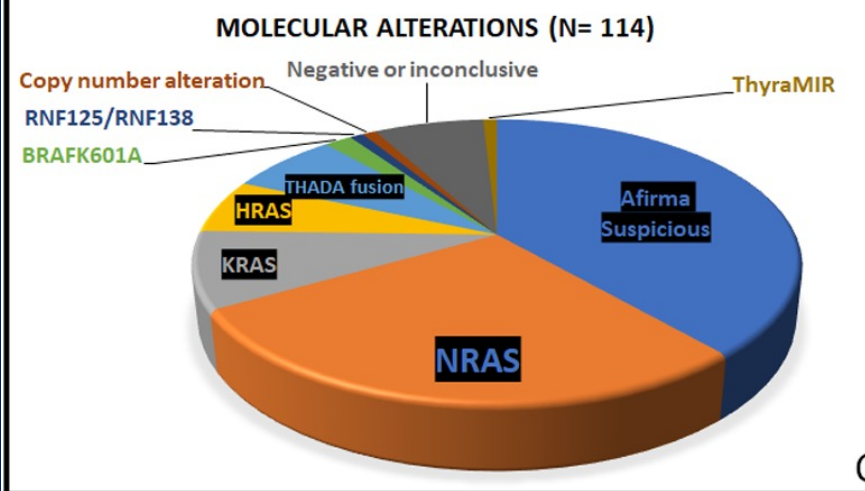
Alzumaili BA et al. A Comprehensive Study on the Diagnosis and Management of Noninvasive Follicular Thyroid Neoplasm with Papillary-Like Nuclear Features. *Thyroid*. 2023 May;33(5):566-577. PMID: 36960710.



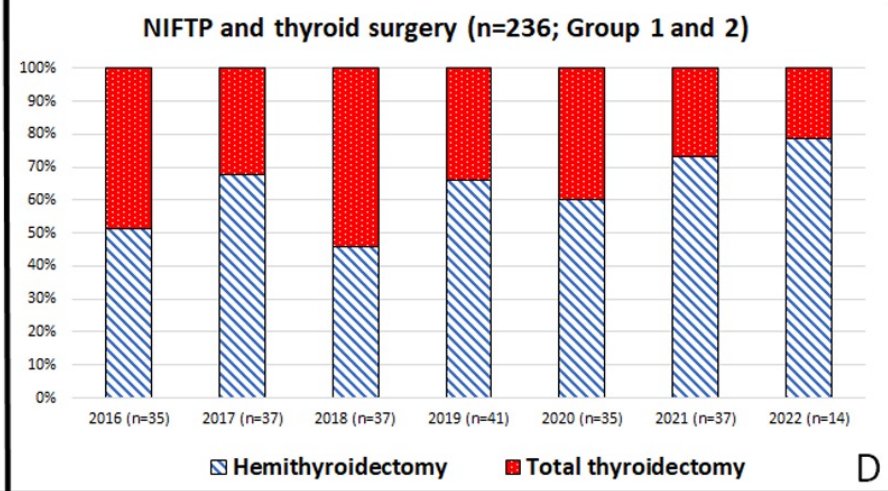
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B



C

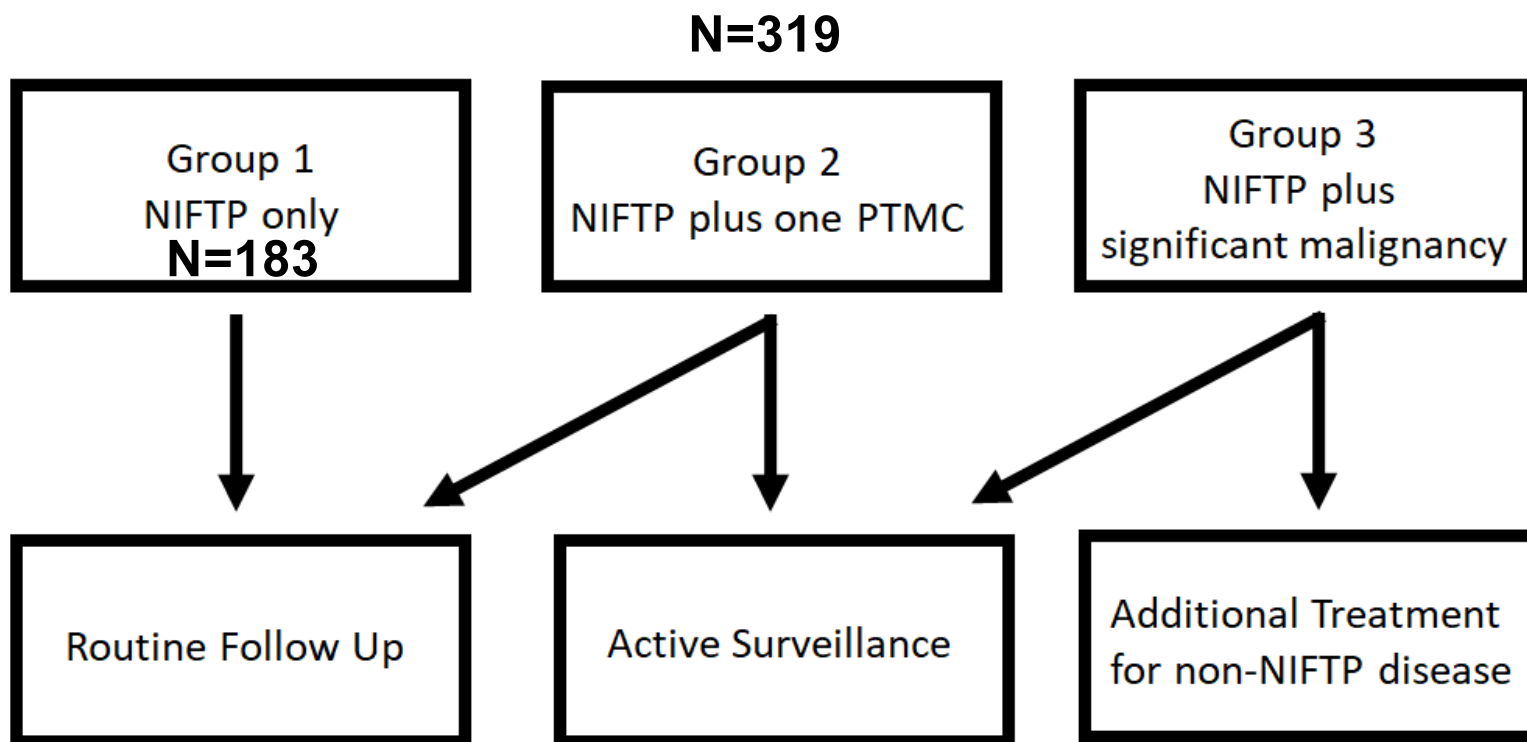


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Follow-up Studies Beginning

Alzumaili BA et al. A Comprehensive Study on the Diagnosis and Management of Noninvasive Follicular Thyroid Neoplasm with Papillary-Like Nuclear Features. *Thyroid*. 2023 May;33(5):566-577. PMID: 36960710.

Management Algorithm Following Diagnosis of NIFTP



Clinical Translation

- NIFTP requires no additional surgery
- May drive more lobectomies
 - ATA guidelines for low-risk ca (2016)
 - **Revised ATA guidelines due late Fall 2023**
 - Updated Bethesda (2017) w no changes
 - Expected left-shift in risk of malignancy
 - **Revised Bethesda (expected Jul/Aug 2023)**
- Diagnosis of NIFTP driven by absence of capsular/vascular invasion (same as FTA)

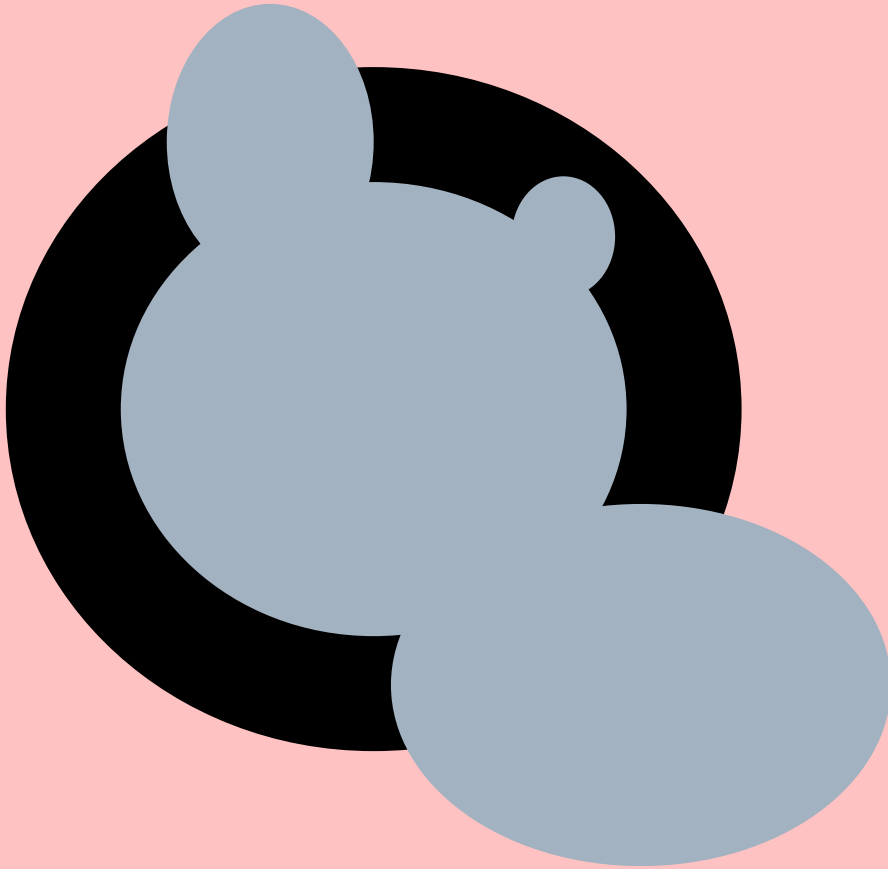
Does FPTC still exist?

- Answer: YES!!!
- FPTC is an *architectural* entity
- Two types FPTC:
 1. Infiltrative FPTC/ECPTC
 - Tumor not well-circumscribed with marked infiltration of the surrounding thyroid tissue
 - Generally associated with *BRAF* p.V600E
 2. EFPTC with capsular/vascular invasion
 - Consider as follicular thyroid carcinoma (*RAS-like*)

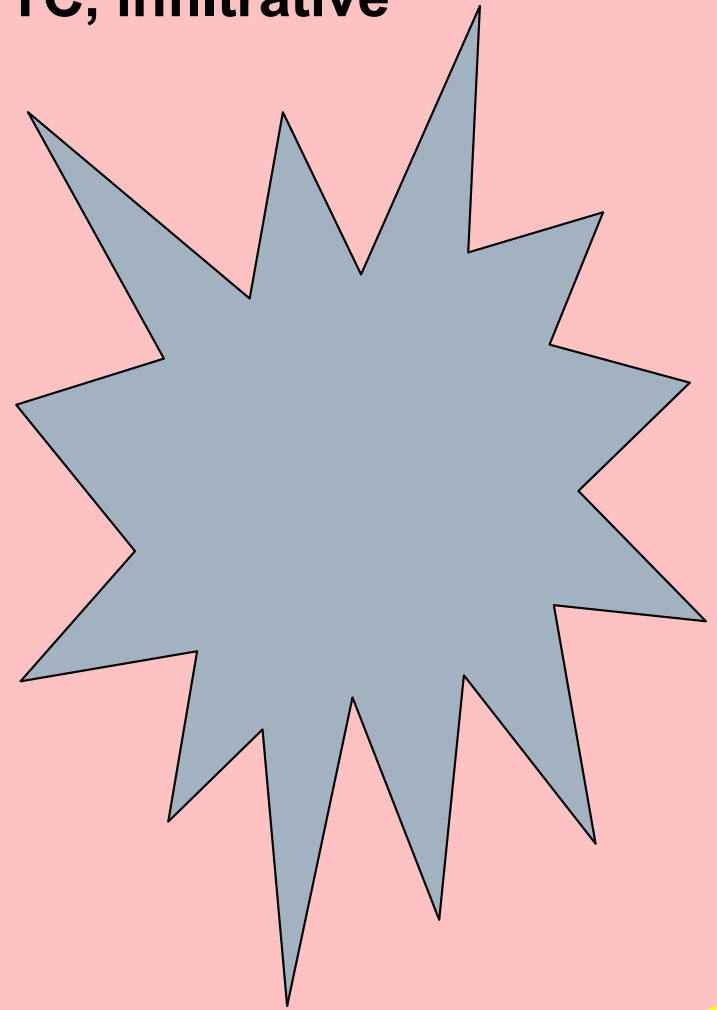


Invasion

FVPTC with capsular invasion



FVPTC, infiltrative



NIFTP is to EFPTC (FTC)
as
FA is to FTC

Non-invasive papillary thyroid neoplasm with papillary-like nuclear features

- well-circumscribed or encapsulated, non-invasive

How do we assess this?

review the capsule

Follicular adenoma (adenomatous nodule)

- well-circumscribed or encapsulated, non-invasive

How do we assess this?

review the capsule

Caveat

- Exclusion criteria for NIFTP is not only **lack of capsular invasion** (requiring submission of capsule) but also **<1% true papillary structures (or any)**
- One case with BRAFV600E
 - Review showed no capsular invasion
 - Entire capsule submitted ***but not entire tumor!***

Zhao L et al., Cytological, molecular, and clinical features of noninvasive follicular thyroid neoplasm with papillary-like nuclear features versus invasive forms of follicular variant of papillary thyroid carcinoma. *Cancer*. 2017 May;125(5):323-331.

Implications for NIFTP

- Diagnosis requires **submission of entire lesion, including capsule**
- Assessment for *BRAF* p.V600E may be helpful adjuvant in difficult cases
 - Efficacious immunohistochemical markers

The “capsule”

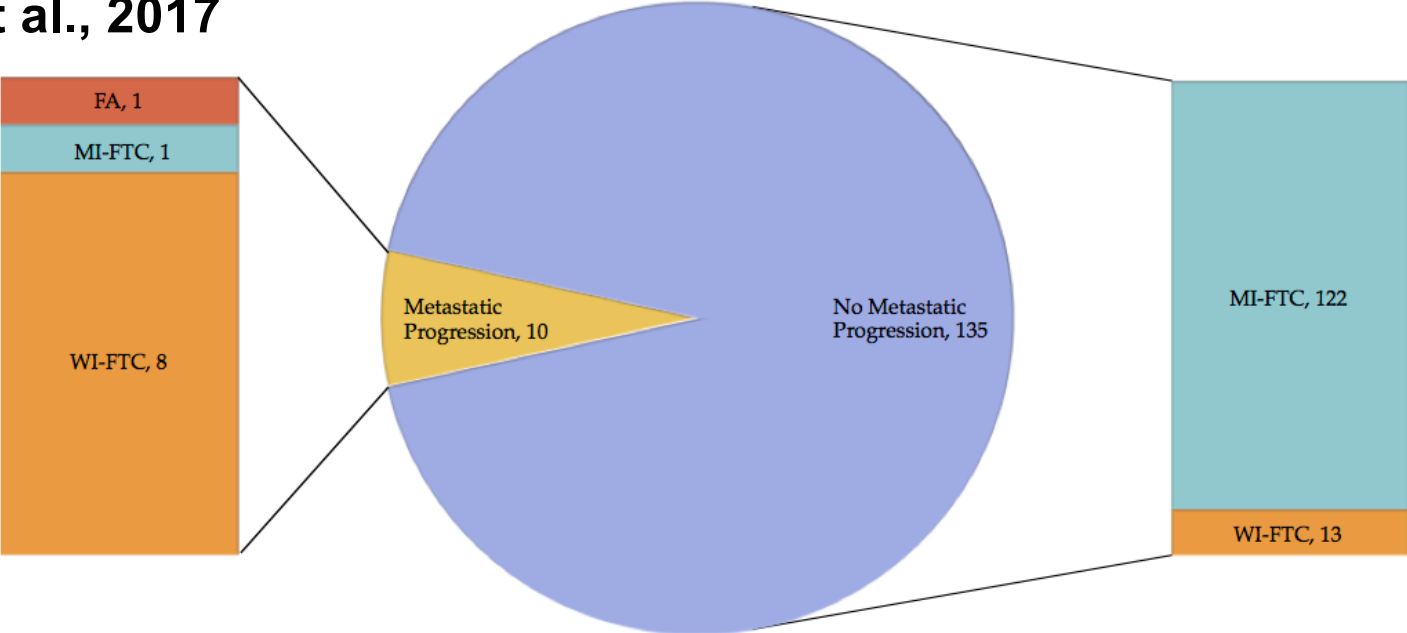
- NIFTP does not require a capsule
- Requires tumor to be well-circumscribed
- How does one assess the “capsule” if there is no capsule?
- For NIFTP, the “capsule” becomes the tumor:non-tumor interface
- The interface should be “adequately sampled”

Tumor capsule submission

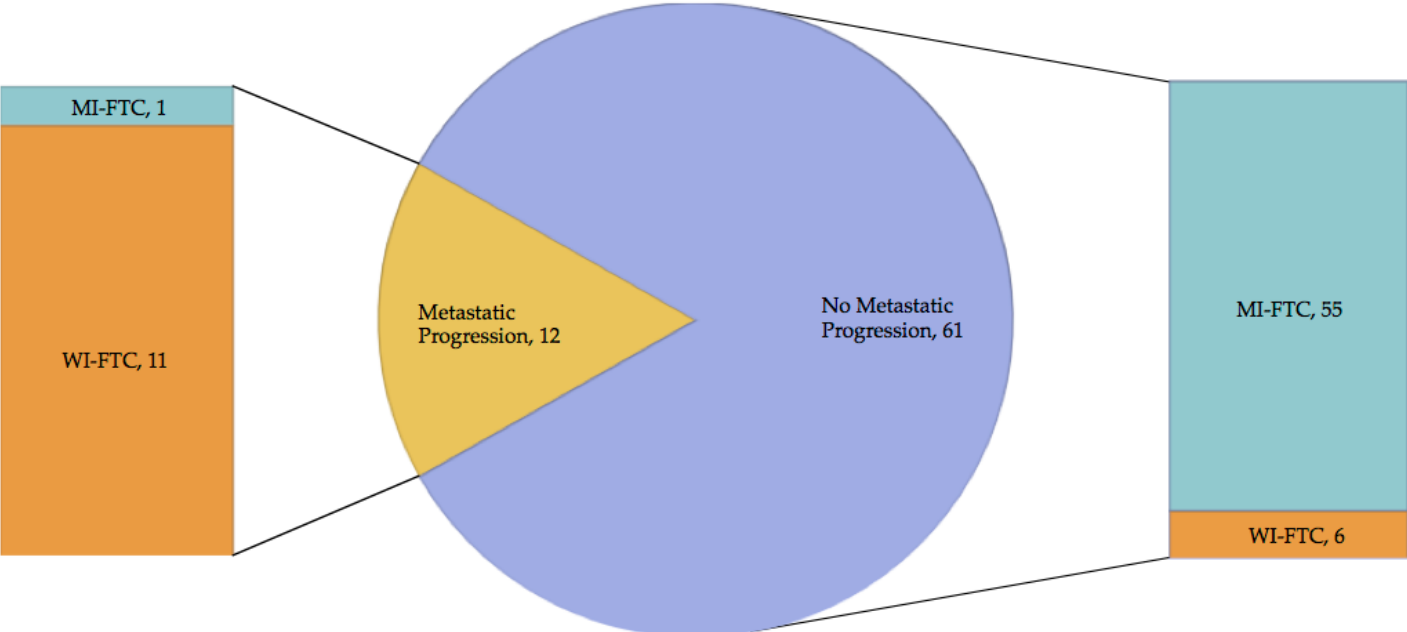
- Trend is for entire capsule submission, without respect to tumor size, to exclude invasion, this is up for debate
- Endocrine pathologists and routine practice in US prefer entire submission (as for FTA)
- European colleagues do not
- FTC study indicates may not matter
 - Glomski K, Nosé V, Faquin WC, Sadow PM. Metastatic Follicular Thyroid Carcinoma and the Primary Thyroid Gross Examination: Institutional Review of Cases from 1990 to 2015. *Endocr Pathol.* 2017 Jun;28(2):177-185.



Glomski et al., 2017



FTC cases with complete capsule submission



FTC cases with incomplete capsule submission

Adequate Sampling of NIFTP

- As exclusion for NIFTP also is $<1\%$ architectural papillae (if present: classical PTC with predominantly follicular architecture), simple tumor:parenchyma (capsule) sampling may not be adequate; the entire tumor may need to be reviewed to exclude papillae



Updated Criteria Review

- Nikiforov YE, Baloch ZW, Hodak SP, Giordano TJ, Lloyd RV, Seethala RR, Wenig BM. Change in Diagnostic Criteria for Noninvasive Follicular Thyroid Neoplasm With Papillary-like Nuclear Features. *JAMA Oncol.* 2018 Aug 1;4(8):1125-1126.
 - Based upon 80 publications in the literature since 2016
 - Allows for NO well-formed papillae
 - Molecular rule-outs*

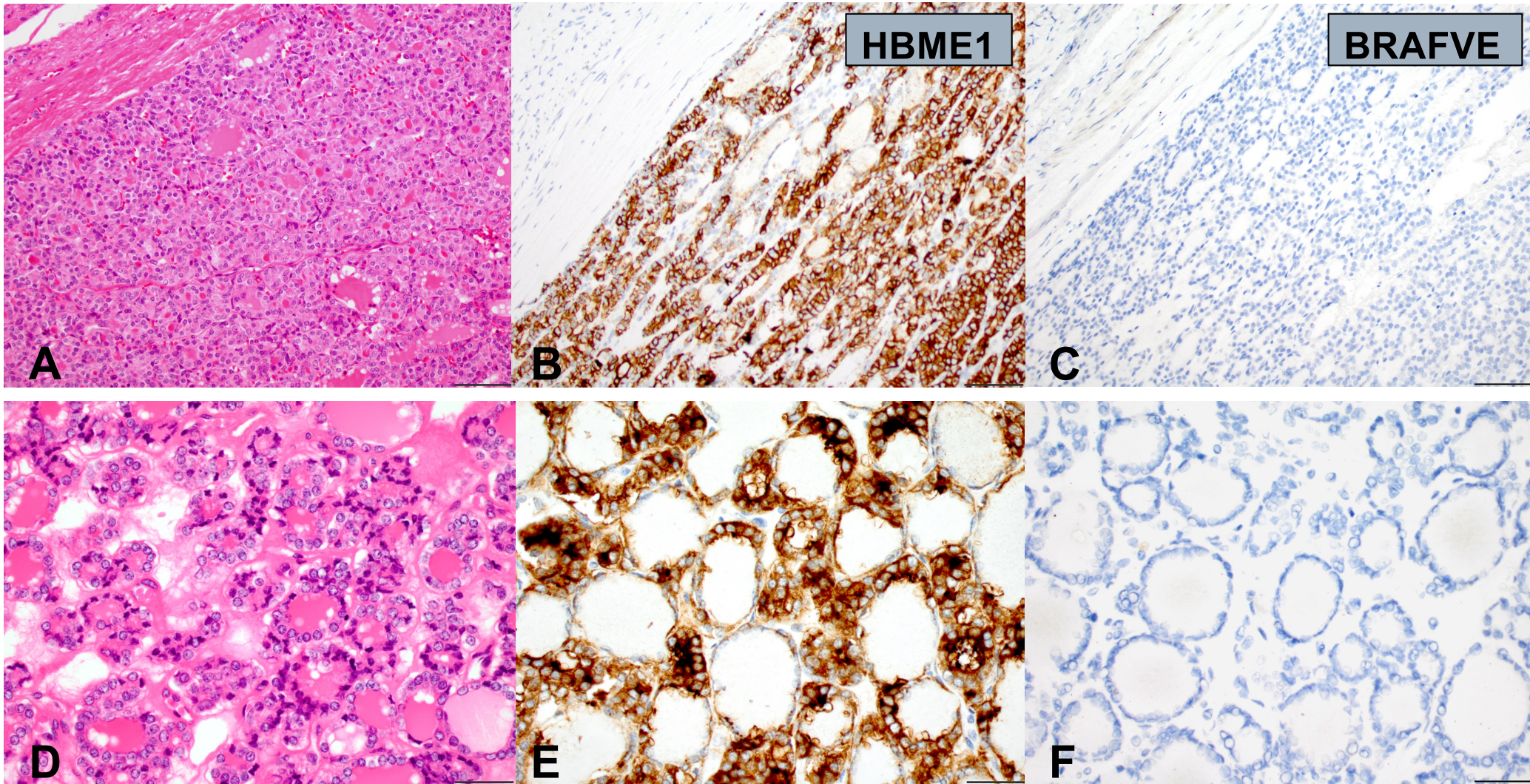
Adequate Sampling of NIFTP

- Glomski study showed that for the vast majority of FTC, representative capsule may be sufficient
 - Suggests prospective studying
- NIFTP, if ***entire tumor*** not submitted, how many ECPTC will be missed?
- Is *BRAF* immunostain sufficient adjuvant test to preclude entire submission?

Subcentimeter NIFTP & BRAFVE

- Several monoclonal antibodies concordant with *BRAF* p.V600E
- In subcentimeter microcarcinomas, up to 65% may have *BRAF* p.V600E
- If well-assessed, BRAF negative (HBME1 positive)

Status of NIFTP



Johnson DN and Sadow PM, Hum Pathol, 2018 Dec; 82:32-38.

Fine Lines of Distinction

- Does size matter? $? \geq 1$
- **Microcarcinomas as NIFTP?**
 - Xu B et al. Endocrine. 2018 Jan;59(1):143-150.
- Since most microcarcinomas are incidental, NIFTP likely should be reserved for intentionally excised lesions

Status of NIFTP

- NIFTP has debuted in Aug 2016
- Several manuscripts in print since its debut (at least 387 as of June 4, 2023)
- Most updated diagnostic criteria in the most recent WHO – Endocrine 2022
 - Baloch ZW, et al. *Endocr Pathol.* 2022 Mar;33(1):27-63.
- Incidence of PTC should decrease
- WT-UMP

Implications of NIFTP

- Reduced diagnosis of papillary thyroid carcinoma
- **Reduced over-treatment** of indolent thyroid tumors – NIFTP has raised awareness
 - Ghossein R et al. Large (>4 cm) Intrathyroidal Encapsulated Well-Differentiated Follicular Cell-Derived Carcinoma Without Vascular Invasion May Have Negligible Risk of Recurrence Even When Treated with Lobectomy Alone. *Thyroid*. 2023 May;33(5):586-592. PMID: 36884299.
- Reduced need for consultation due to **increased reproducibility** of morphological features
- Bethesda FNA criteria modified 2017 (2023)

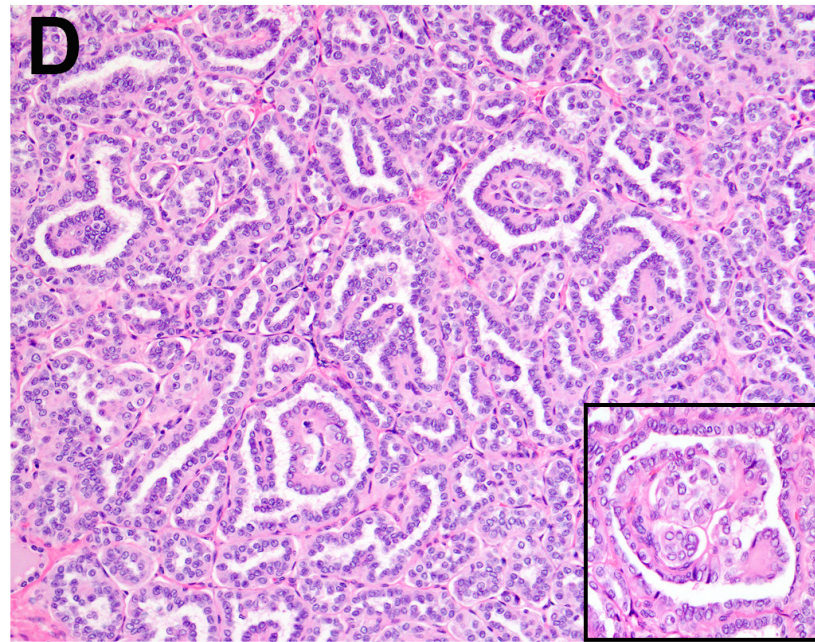
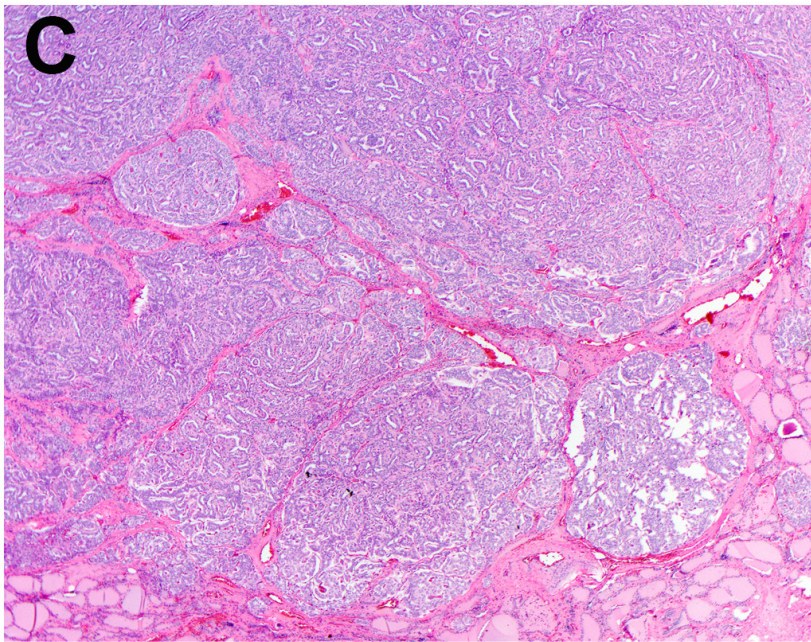
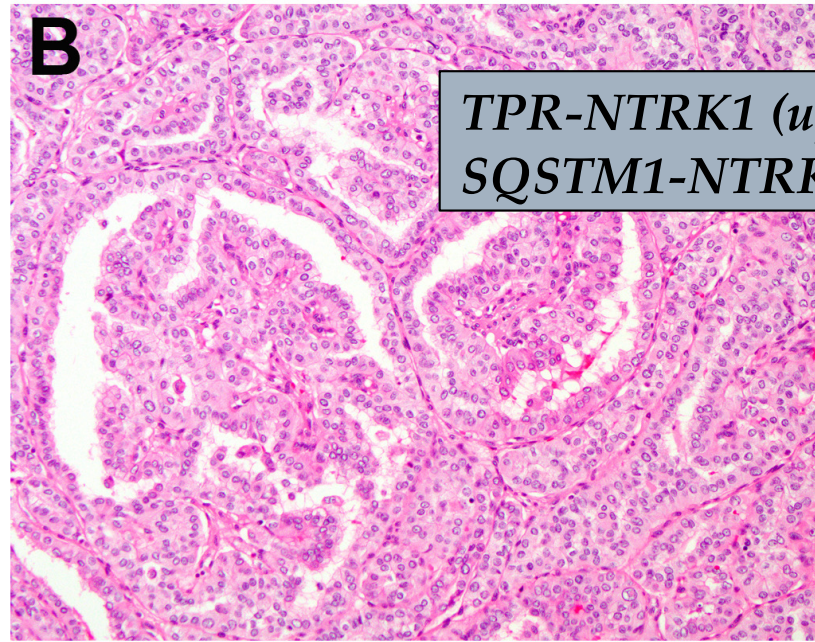
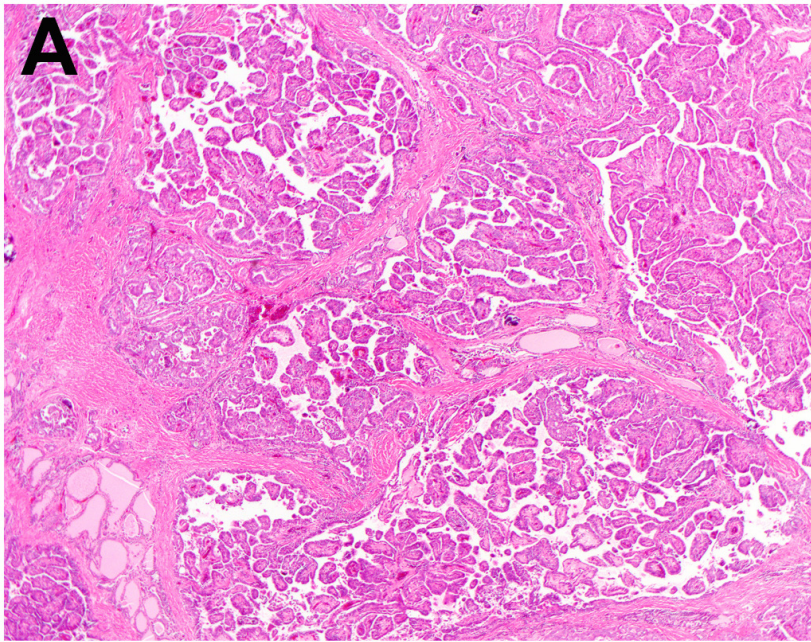
Molecular Diagnostics

- Atypia of Undetermined Significance (BIII)
- Follicular neoplasm (BIV)

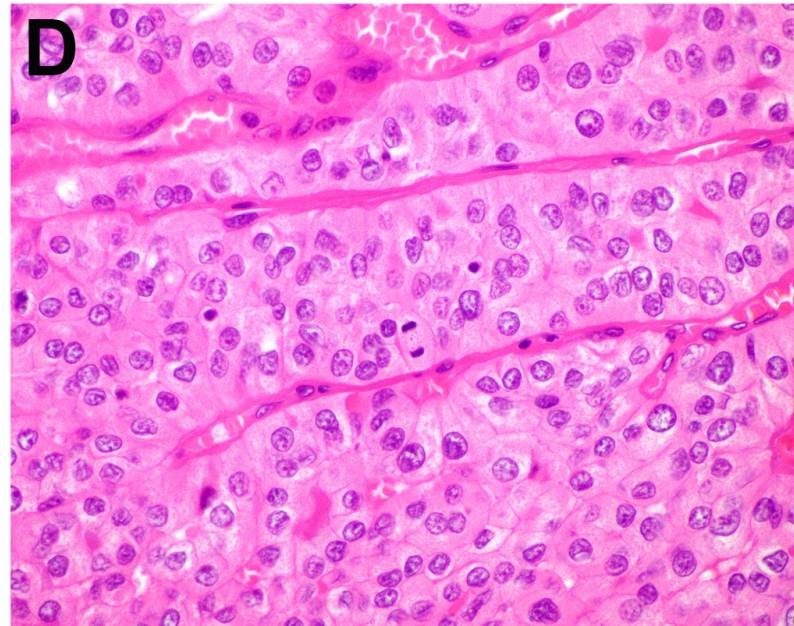
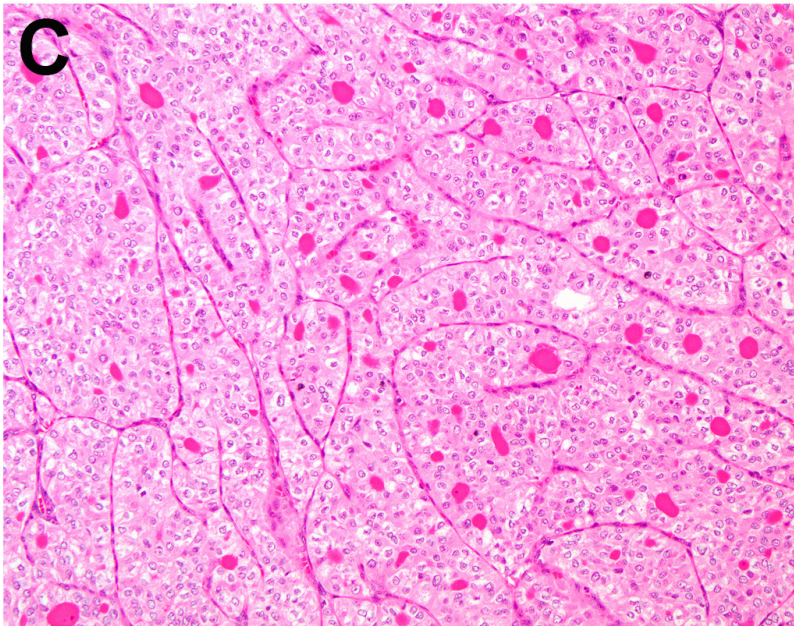
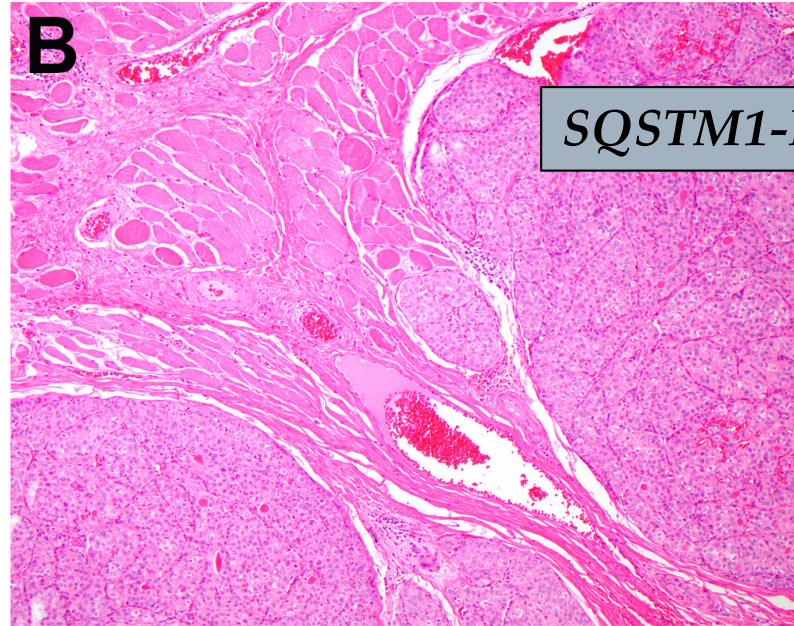
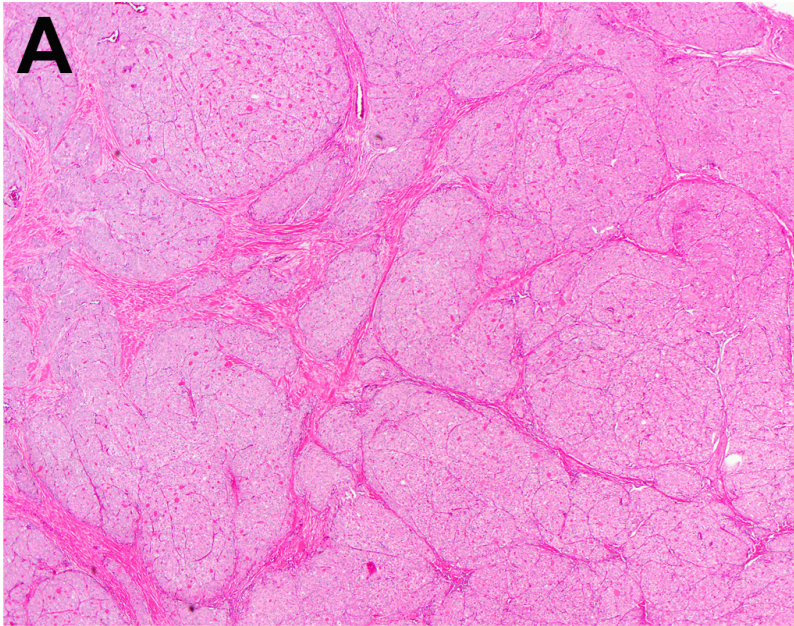
- Molecular Testing
 - *RAS*-like/*BRAF* p.V600E, others
 - Decision tree

Dawn of Actionable Mutations

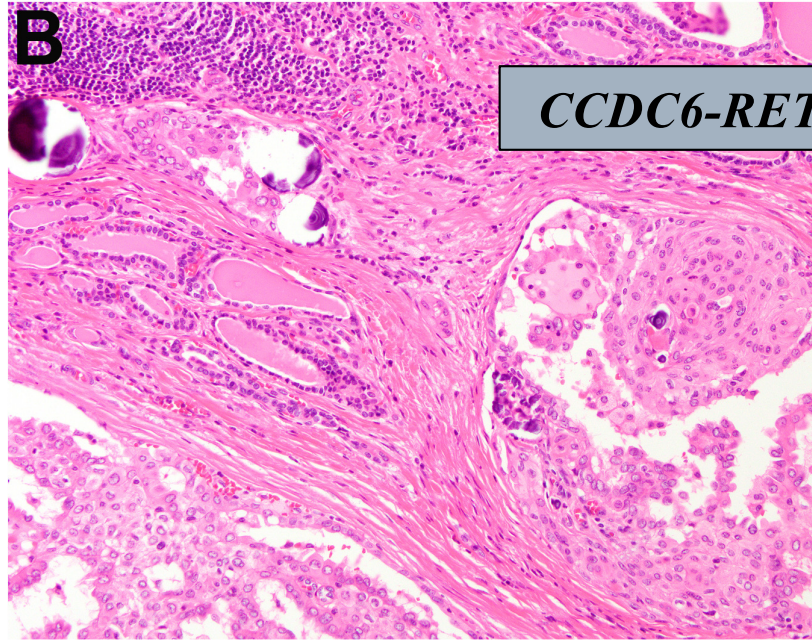
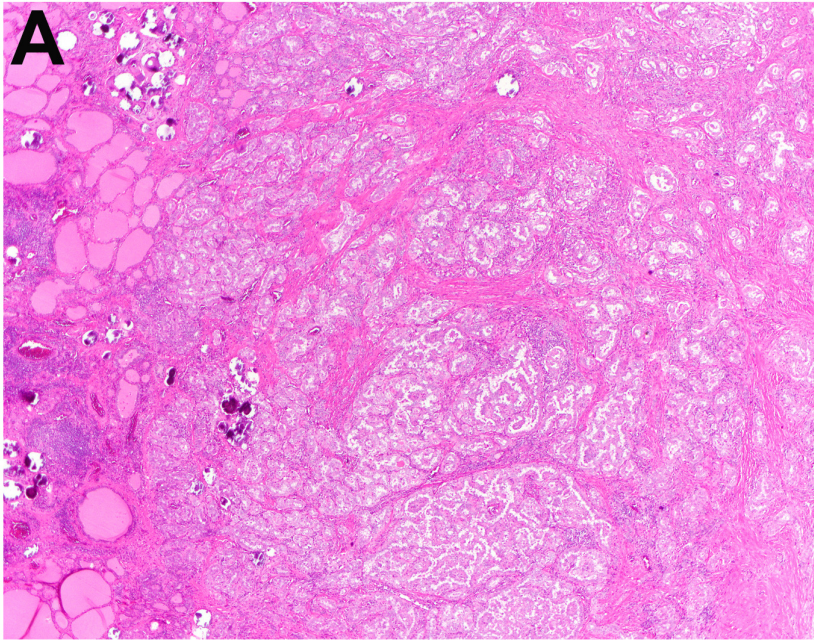
- Clinicopathologic and molecular characterization of NTRK-rearranged thyroid carcinoma (NRTC)
 - Chu YH, et al. *Mod Pathol*. 2020 Nov;33(11):2186-2197.
- Clinicopathologic features of kinase fusion-related thyroid carcinomas: an integrative analysis with molecular characterization
 - Chu YH, et al. *Mod Pathol*. 2020 Dec;33(12):2458-2472.



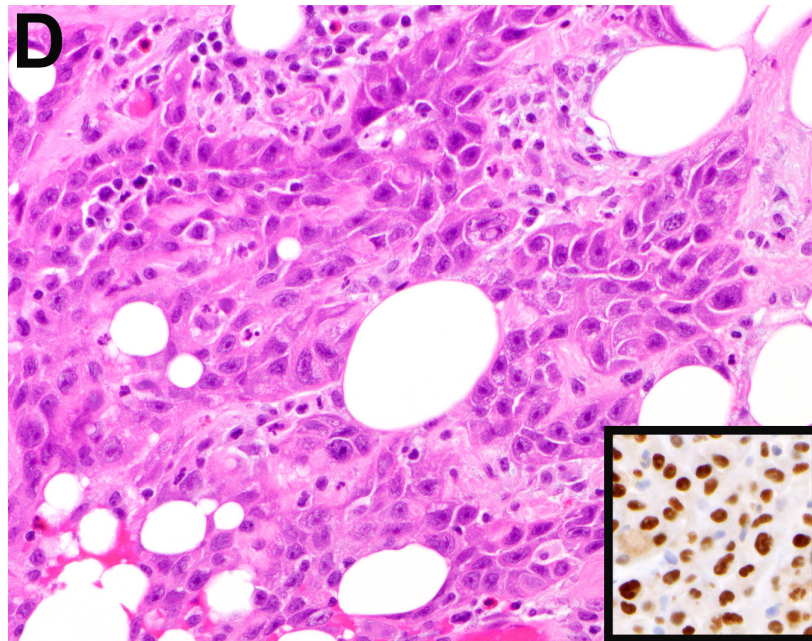
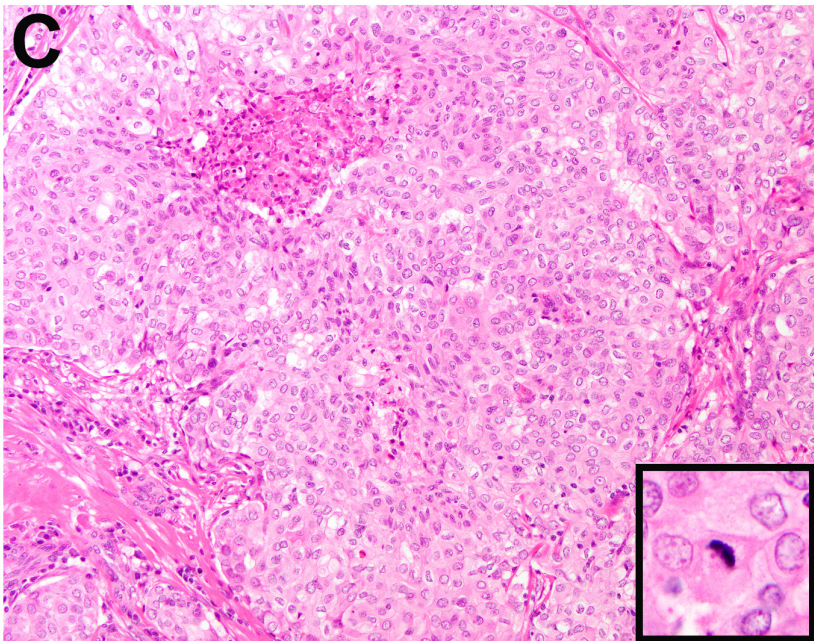
Chu YH, et al. *Mod Pathol*. 2020 Nov;33(11):2186-2197.



Chu YH, et al. Mod Pathol. 2020 Nov;33(11):2186-2197.

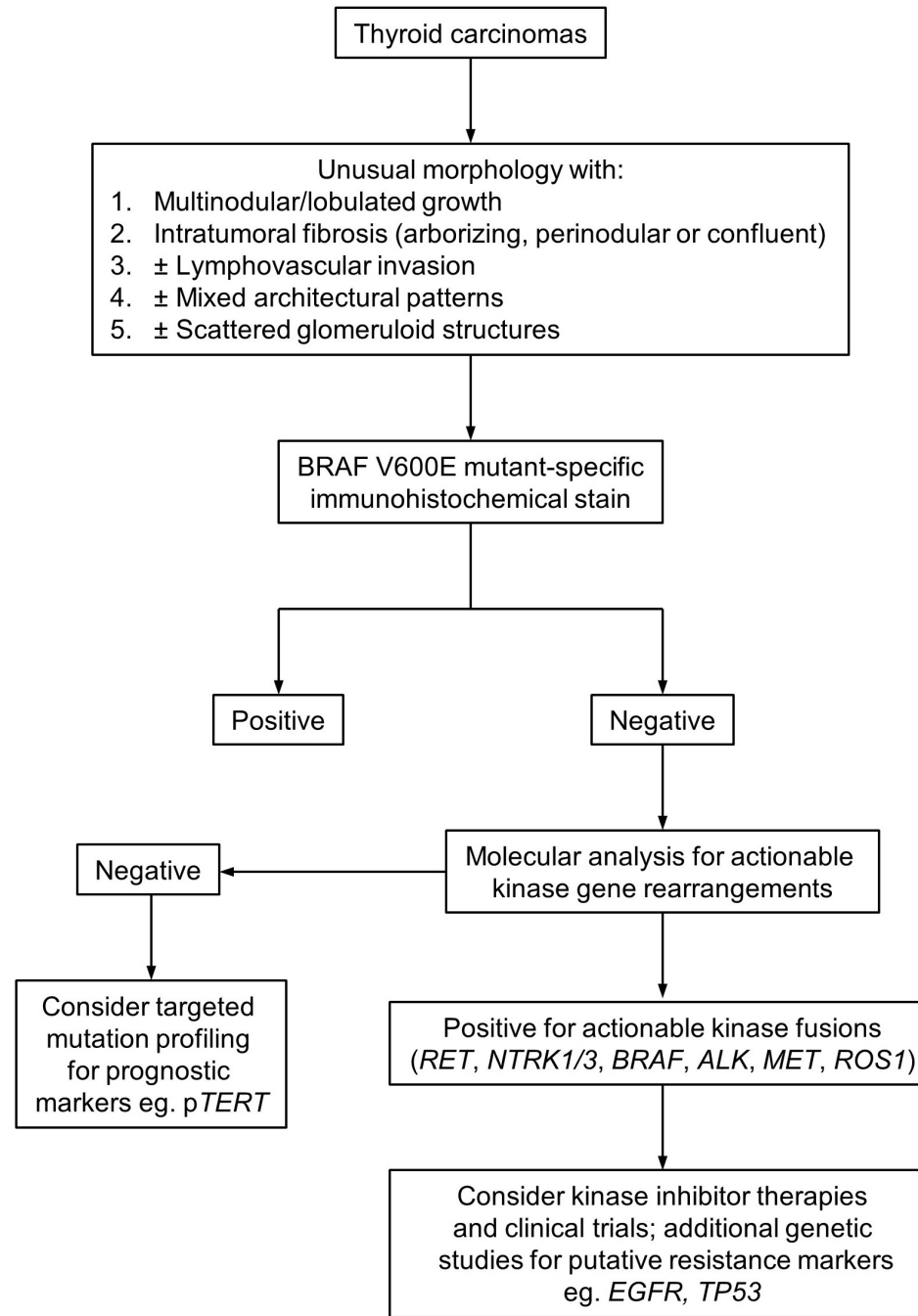


CCDC6-RET fusion PTC



Chu YH et al, *Mod Pathol*. 2020 Dec;33(12):2458-2472.

Algorithm



RET

- Kinase fusion-related carcinomas
- Most commonly associated with **medullary thyroid carcinoma**
- Despite point mutations (most commonly M918T) being most common, behavior is activated *RET* kinase domain

Small Molecule Inhibitors of Kinase

- Wirth LJ, et al. Efficacy of Selpercatinib in *RET*-Altered Thyroid Cancers. *N Engl J Med*. 2020 Aug 27;383(9):825-835.

Grading System for MTC

- Xu B, et al. International Medullary Thyroid Carcinoma Grading System: A Validated Grading System for Medullary Thyroid Carcinoma. *J Clin Oncol*. 2022 Jan 1;40(1):96-104.
- Williams JF, et al. Grading of Medullary Thyroid Carcinoma: an Interobserver Reproducibility Study. *Endocr Pathol*. 2022 Sep;33(3):371-377.



Grading System vs Genetics

- Reports on traditional methods of pathological evaluation, such as grading system or staging seem antiquated
- Too much faith in genetics?
- Integrated approach?
- Follow-up studies post-resistance
 - Seki Y, et al. Liquid biopsy for the detection of resistance mutations to ROS1 and RET inhibitors in non-small lung cancers: A case series study. *Respir Investig.* 2022 Sep 9:S2212-5345(22)00114-9.
- Extragenomic modifications (ie. methylation)

Summary

- The past 10 years have changed the definitions and implications for thyroid carcinomas
- Approach to diagnostics in follicular thyroid tumors is still controversial
- Multidisciplinary integrated diagnostics are essential
- Integrated reporting will yield most effective communication

Summary

- Discussions of expected standards of care for assessment and reporting are essential
- Updated expectations regarding the role of molecular genetics in diagnosis and rational therapeutics are essential, including timing of testing
- Issues of equity of care must be considered when developing diagnostic protocols given disparities in resources available

Conclusion

- As we have moved into the molecular era and closer toward integrated diagnostics (including the whole genome), so, too, must rational diagnostics evolve, integrating prognostic information and recommended clinical next steps, moving pathology consultation practice to a higher level