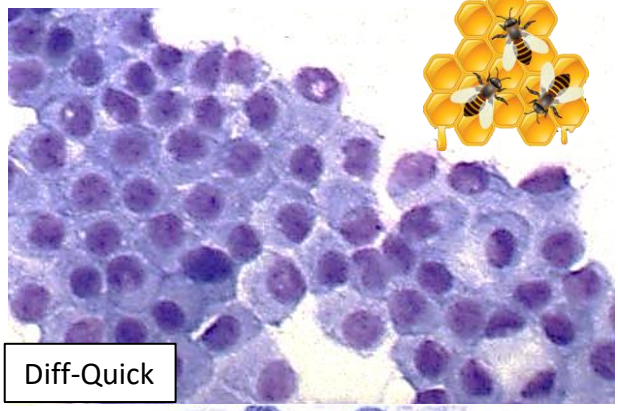
Prepared by Dr. Kurt Schaberg

Thyroid Cytology

Adequacy Criteria

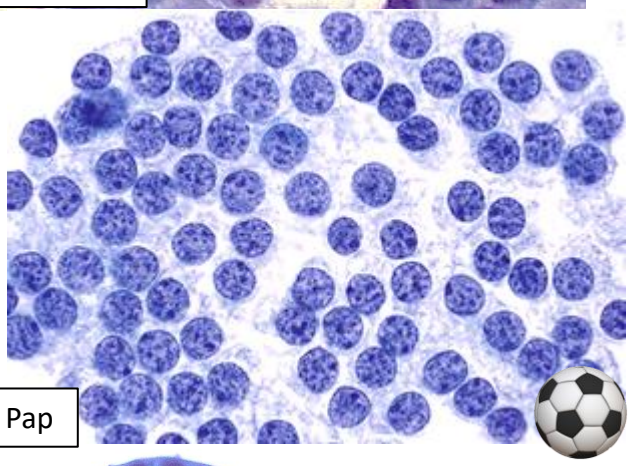


Diff-Quick

Must see at least **6 groups** of well-visualized follicular epithelial cells, each consisting of at **least 10 cells.** *(Ideally on one slide, but not req.)*

Exceptions:
1) Abundant **colloid** with radiographic findings compatible with a colloid nodule
2) Abundant **inflammation** with a solid nodule (lymphocytes, granulomas, or neutrophils)
3) **Atypia**

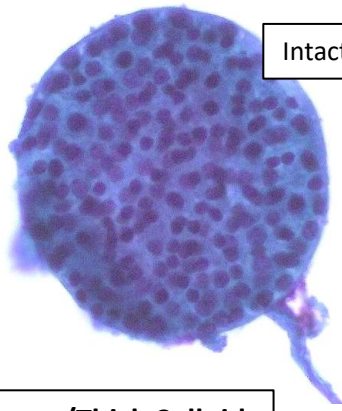
<u>Normal cytology</u>:
Follicular epithelium should be in nice big, flat **monolayered, macrofollicular sheets**, with evenly spaced ("Honeycomb-like") cells. <u>Minimal overlapping</u>. Sometimes see intact big 3-D entire spherical follicles.



Pap

Cells should have round nuclei with **uniformly granular, dark chromatin** (think: "soccer ball").
Nuclei are slightly bigger than an RBC.

If abundant granular cytoplasm ± nucleoli→ Hürthle (Oncocytic) cells (a common change/metaplasia)



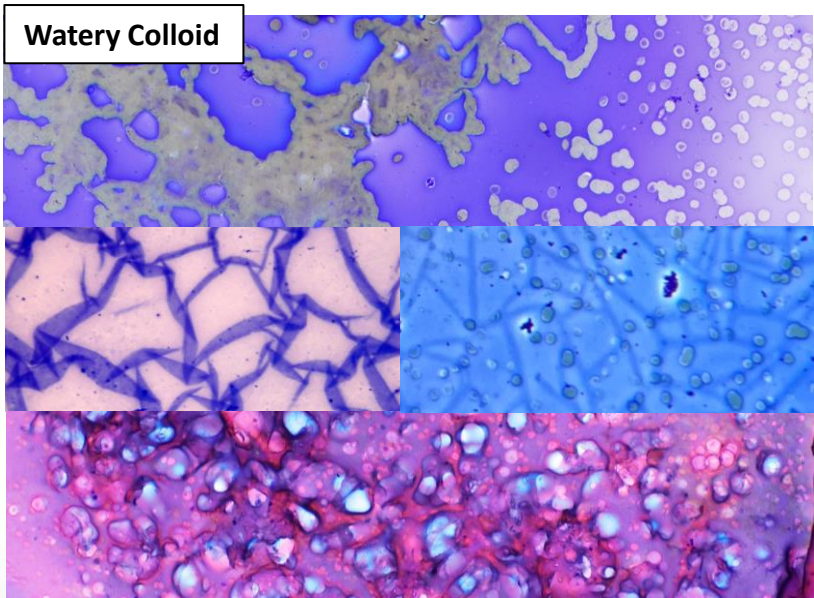
Intact macrofollicle

<u>Colloid</u> can appear "watery" as evenly spread proteinaceous fluid, in "puddles" or "dense/thick" as 3D hyaline globules.

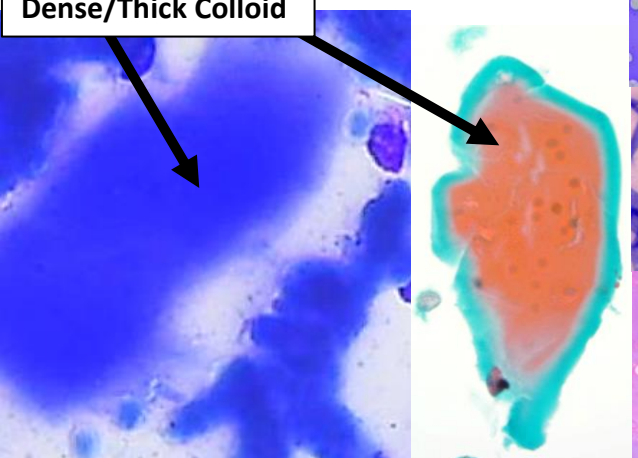
Hints to thin, watery colloid include: Cracks (like glass), Wrinkles (like cellophane), and rouleaux formation.

On ThinPrep: Looks like "tissue paper"

Watery Colloid



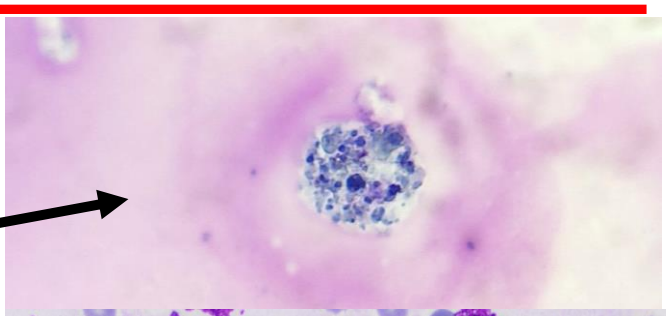
Dense/Thick Colloid

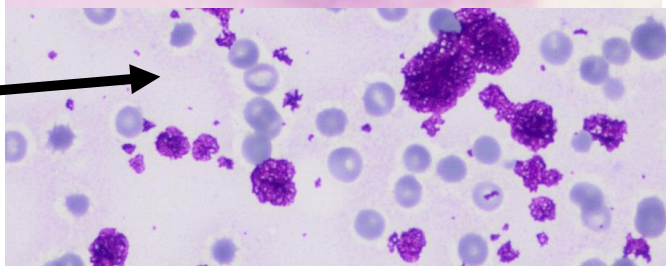


Nondiagnostic

Used for specimens that don't meet adequacy criteria on first page.

<u>*Common scenarios:*</u>
Blood only (paucicellular)
Cyst fluid only
Proteinaceous fluid, debris, and hemosiderin-laden histiocytes
Obscuring ultrasound gel
Bright purple globules (on diff quick)
Obscuring blood
Poor slide preparation/staining





Benign Lesions

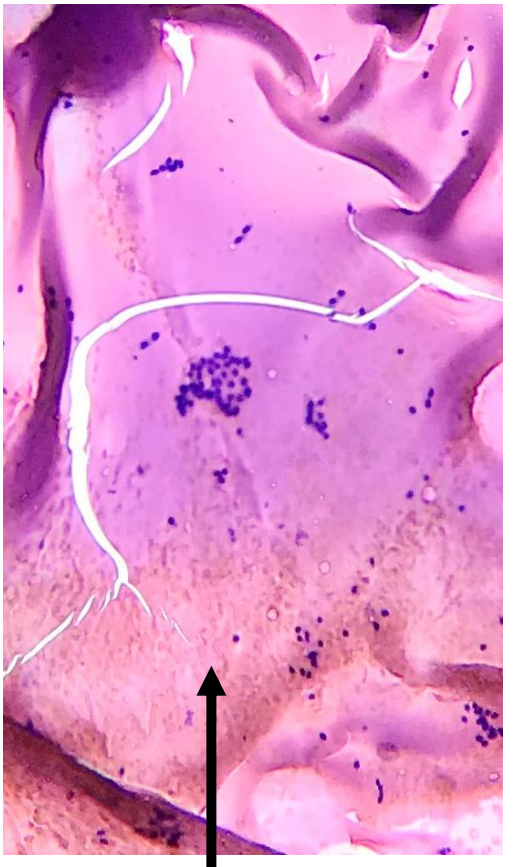
Benign Follicular Nodular Disease *"FND"*

Histologically represent nodular goiter, adenomatoid nodules, hyperplasia, and colloid nodules. Now, refer to as "***Follicular nodular disease***" (FND) as may or may not be neoplastic.

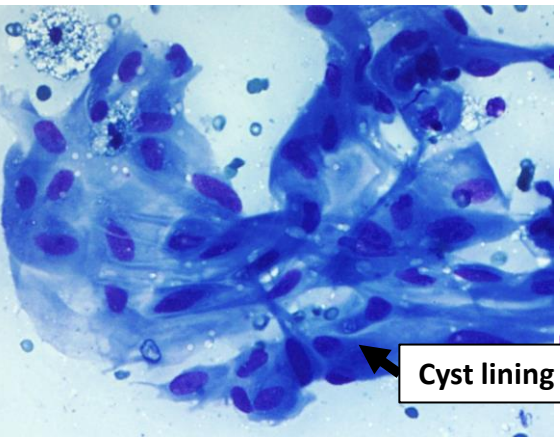
Variable amounts of: **colloid** (the more the better! ;-)**, bland follicular cells, Hürthle cells, and macrophages.**

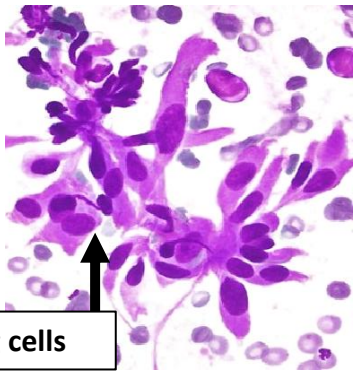
Should be <u>sparse to moderately cellular</u> with a good amount of colloid (often easiest to see on diff-quick).
Occasional microfollicles are permissible.
A little anisonucleosis is ok (attributable to endocrine atypia) as long as there is no nuclear contour irregularities or chromatin clearing

Frequently see <u>*Cystic degeneration*</u>*:* Changes include macrophages, "reparative" stretched cyst lining epithelial cells. These cells are elongated, enlarged, and have finely granular chromatin and squamoid or spindled shape/cytoplasm. Low N:C ratios. Can *mimic* malignancy/AUS, but can forgive if focal and mild.

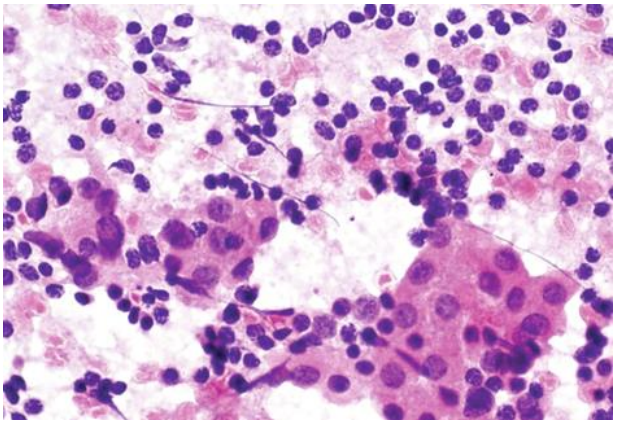


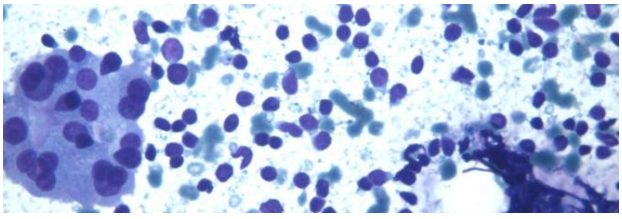
Classic, reassuringly benign findings: A ton of colloid, with scattered bland follicular epithelium ☺





Lymphocytic Thyroiditis



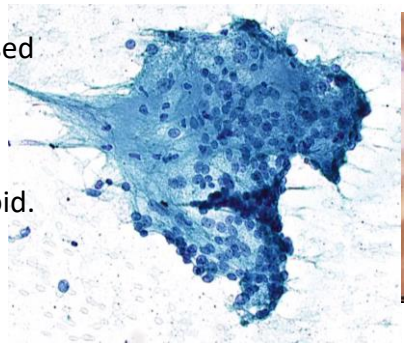


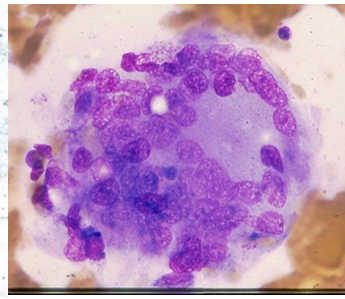
Generalized term that encompasses many conditions, the most common of which is **chronic lymphocytic (Hashimoto's) thyroiditis**. Often middle-aged women with associated circulating autoantibodies.

Hypercellular smear.
Abundant, polymorphic lymphocytes.
Hürthle/Oncocytic metaplasia common
(Large cells with abundant granular cytoplasm and prominent nucleoli).
Advanced cases may be hypocellular (due to fibrosis).
May have significant anisonucleosis.

No minimum quantity of follicular cells required for adequacy.

Granulomatous Thyroiditis *aka subacute or de Quervain's*

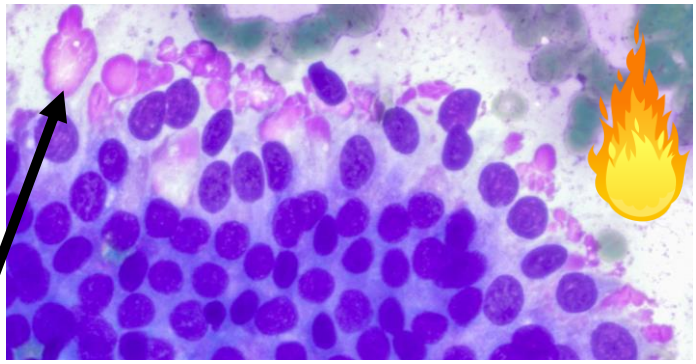




Self-limited inflammatory condition, usually diagnosed clinically.

Clusters of epithelioid histiocytes (i.e., **granulomas**) and **multinucleated giant cells**, often ingesting colloid.
Early can have neutrophils and eosinophils.
Later stages are hypocellular and have lymphocytes.

Graves Disease



Autoimmune thyroid hyperplasia/hyperthyroidism. Although usually diffuse, can have nodules.

Non-specific findings, on spectrum with FND.
Abundant, delicate, foamy cytoplasm.
Enlarged nuclei with prominent nucleoli.
"*Flame cells*" have marginal vacuoles with red/pink frayed edges.

Other Benign Disorders

Pigment: Can see lipofuscin and/or hemosiderin commonly. Can see "black thyroid" pigmentation with tetracycline family antibiotics.

Amyloid goiter: Diffuse amyloid deposition in thyroid gland. Can be 1° or 2°. Amyloid has amorphous look of colloid, but has embedded fibroblasts.

Acute Suppurative Thyroiditis: Bacterial infection→ present with fever and neck pain. Smears show fibrinopurulent debris (abscess).

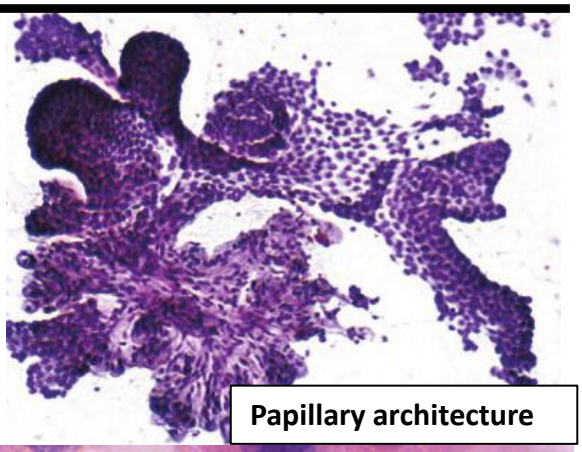
Riedel Thyroiditis: IgG4-related disease of the thyroid with progressive fibrosis. Hypocellular smears.

Tumors

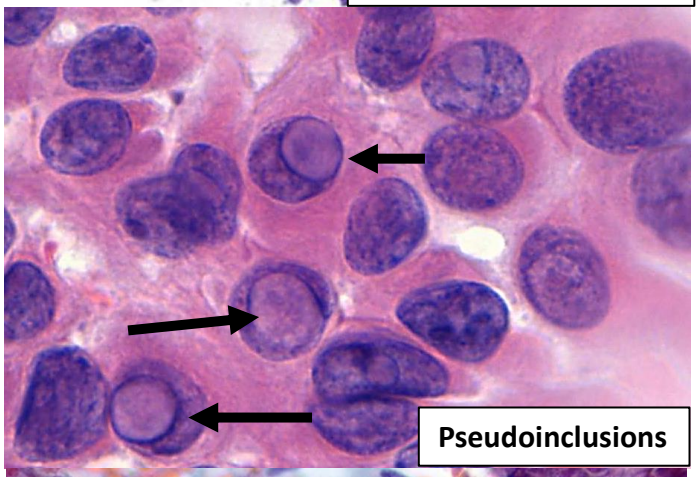
Papillary Thyroid Carcinoma *"PTC"*

Most common malignant thyroid neoplasm
Relatively good prognosis. Spreads via lymphatics to lymph nodes.

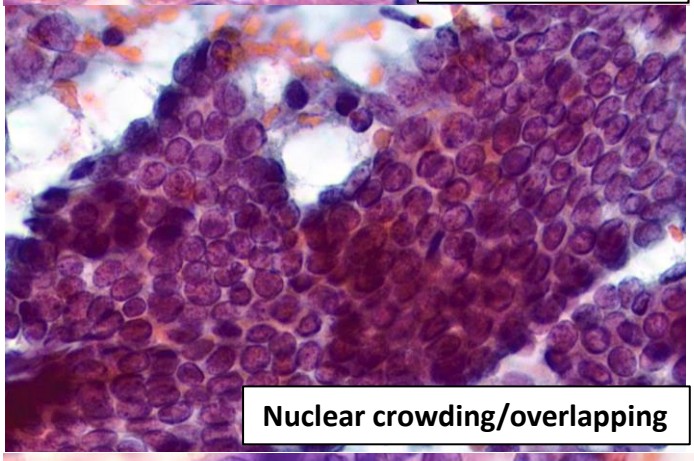
Architecture:
-**Papillary structures** w/ and w/o fibrovascular cores
-May be in monolayered sheets or 3D groups
-Can see cellular swirls ("cartwheel" pattern)


Papillary architecture

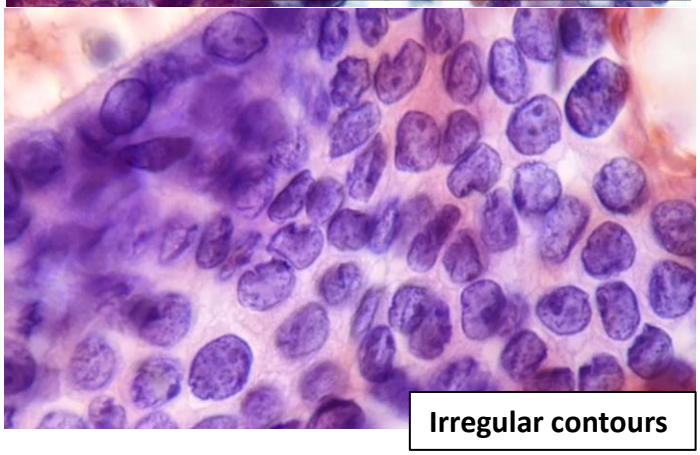
Nuclear features:
-**Enlarged and crowded nuclei,** often molded
  -Nuclear overlapping, elongation
-**Irregularly shaped** to oval nuclei
-Longitudinal nuclear **grooves**
-Intranuclear (cytoplasmic) **pseudoinclusions** (INPI)
-**Powdery**, **pale chromatin**
-Marginal micronucleoli
-Thick nuclear membranes


Pseudoinclusions

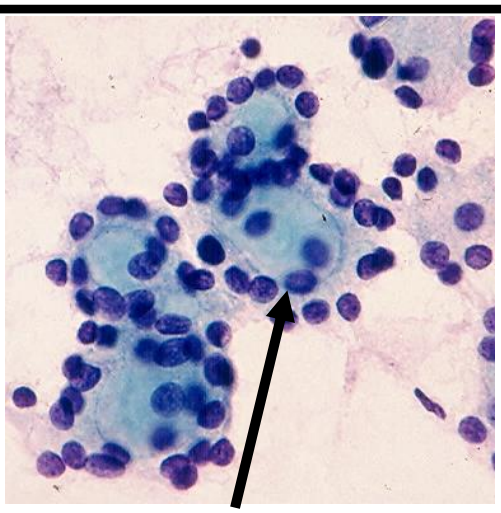
Other features:
-Dense, **squamoid cytoplasm**
-Multinucleated **giant cells**
-Dense, "**Bubble gum" colloid**
-Septate cytoplasmic vacuoles
-Psammoma bodies
-Oncocytic metaplasia
-"Histiocytoid" and "hobnail" cells


Nuclear crowding/overlapping


Bubble gum colloid


Irregular contours

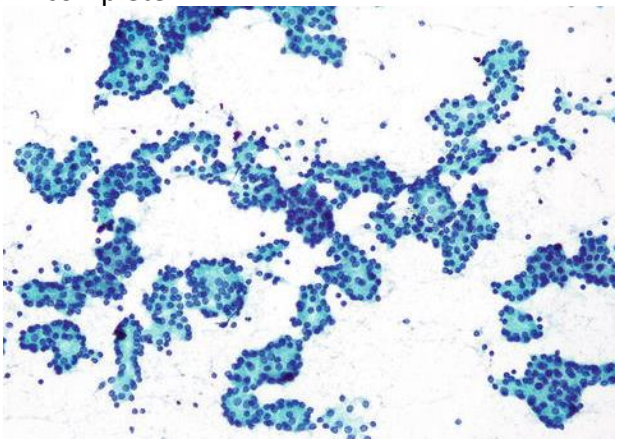
Follicular Neoplasm *"FN"*



Cannot differentiate between Follicular Adenoma and Carcinoma on cytology specimens (need to see capsular or vascular invasion on resection specimen!), so one overarching cytology Dx is given.

Moderately or **Markedly cellular**
Significant **alteration in follicular architecture** (i.e. *non*-macrofollicular)
→ Repetitive **microfollicular pattern**, single cells, and/or cell **crowding/overlapping in trabeculae.**
→ This should be the *dominant* pattern to Dx FN (occasional microfollicles are acceptable in FND)
→ **Minimal colloid**
Usually minimal cytologic atypia.

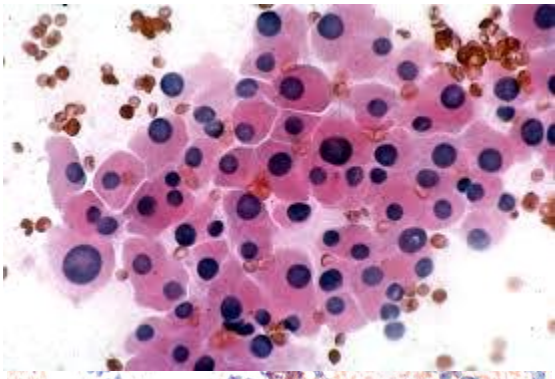
Microfollicle: less than 15 cells arranged in a circle that is at least 2/3 complete

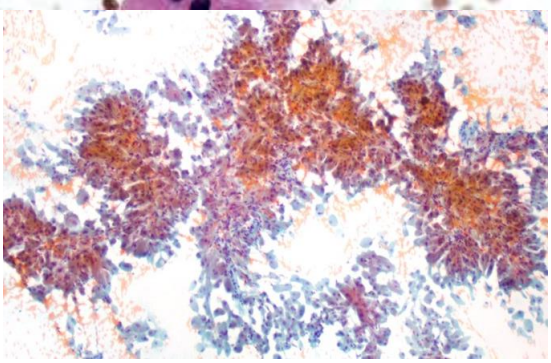


If less cellular→ consider AUS
If cytologically malignant→ consider "Suspicious for malignancy" or just "Malignant"

Note: The Bethesda system no longer endorses saying "Suspicious for FN" even though up to 1/3 of cases will be non-neoplastic, hyperplastic nodules… just put in a disclaimer for that possibility in your report I guess ¯_(ツ)_/¯

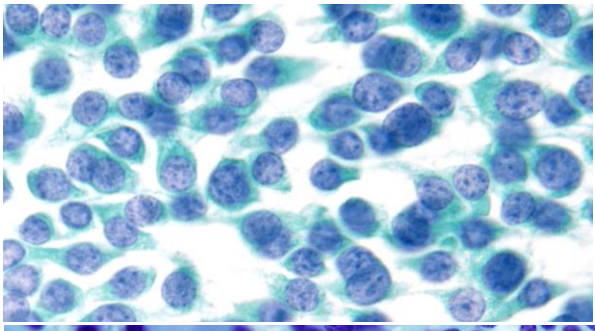
Oncocytic (Hürthle cell) Follicular Neoplasm *"FN-OFN"*

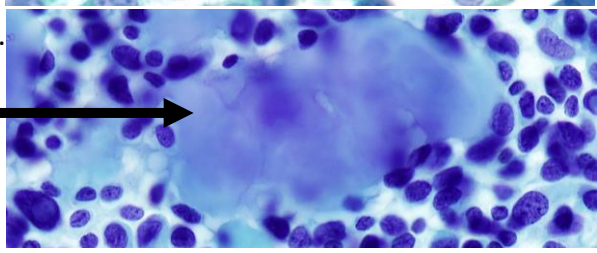




Exclusively oncocytes (or almost exclusively)
→Abundant granular cytoplasm, enlarged round nucleus, prominent nucleolus
Moderately or **Markedly cellular**
Significant **alteration in follicular architecture** (i.e. *non*-macrofollicular)
→ **Single cells, sheets and/or trabeculae.**
Minimal colloid
Transgressing blood vessels
Absence of inflammation
Atypia/Anisonucleosis
→ Large oncocytes with ≥2x variability nuclear size
→ Small oncocytes with high N:C ratios
→ Binucleation is common
→ Careful though: While oncocytic atypia is pretty much always seen in malignancy, atypical oncocytes can be seen in FND and lymphocytic thyroiditis. However, if there is NOT atypia, consider downgrading to AUS or benign (Not specific, somewhat sensitive)

Medullary Carcinoma





Rare. Can be sporadic or inherited (part of MEN 2A&B)
Derived from Parafollicular C cells→ stain with **Calcitonin**!
***RET* mutations** (somatic or germline)

Moderate to **Marked Cellularity**. Often discohesive.
Plasmacytoid, polygonal, to spindled cells.
Mild to moderate pleomorphism.
"Salt and Pepper" chromatin. Can have intranuclear **inclusions**.
Granular cytoplasm with small granules.
Occasional **amyloid** fragments

IHC: **(+) Calcitonin,** Synaptophysin/Chromogranin, TTF1, ±PAX8
(-) Thyroglobulin

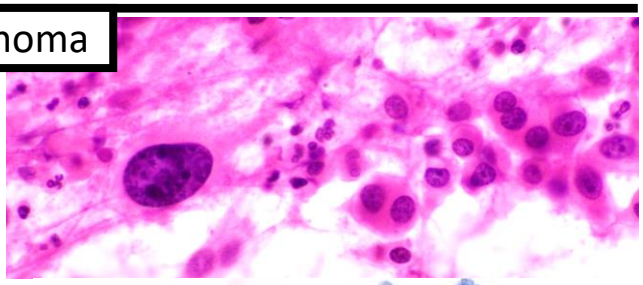
Poorly-Differentiated Thyroid Carcinoma

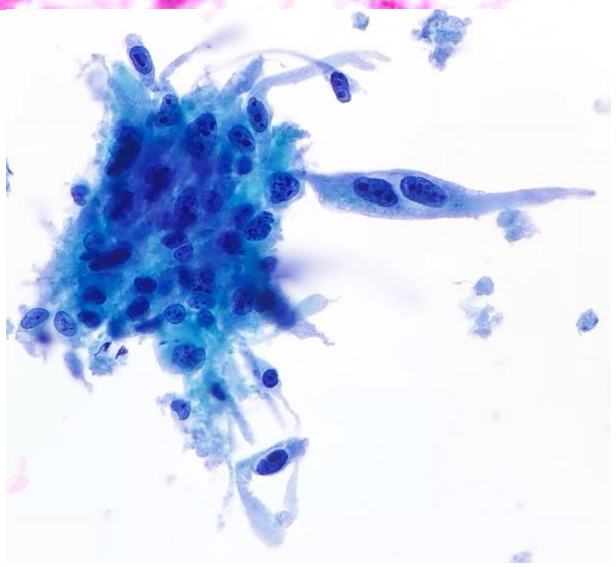
Thyroid carcinomas with **necrosis** and **high mitotic activity** that have an intermediate prognosis between well-differentiated thyroid carcinomas (e.g., PTC) and Anaplastic carcinoma.

Uniform population of cells. High N:C ratios. Cellular smears. Scant/absent colloid.
Apoptoses, **mitoses**, and/or **necrosis**.
Should consider/exclude a metastasis.

Often hard to specifically diagnose on FNA. Often diagnosed as "malignant" or "follicular neoplasm," etc… Compared to Anaplastic carcinoma, which also has mitoses and necrosis, PDTC does *not* have extreme pleomorphism or Sarcomatoid features.

Undifferentiated (Anaplastic) Thyroid Carcinoma



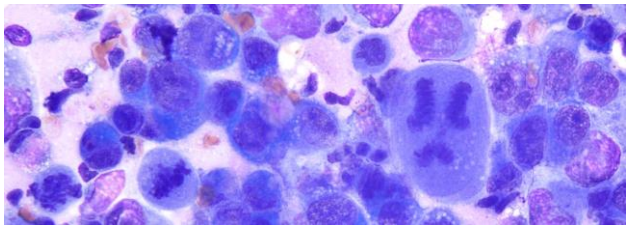


Extremely **aggressive**. Poor prognosis (<1yr survival).
Classically **older women with rapidly growing**, hard neck mass → trouble breathing

Variable cellularity. Often discohesive.
Epithelioid to Spindled cells.
Enlarged, **pleomorphic** nuclei. Chromatin clumping.
Often associated **necrosis and inflammation** (often PMNs).
Can see osteoclast-like giant cells.
Abundant **mitoses**, often abnormal.

Squamous cell carcinoma is considered a subtype (but have to exclude a metastasis!)

IHC: (+) Pancytokeratin, PAX8; (-) TTF1, Thyroglobulin



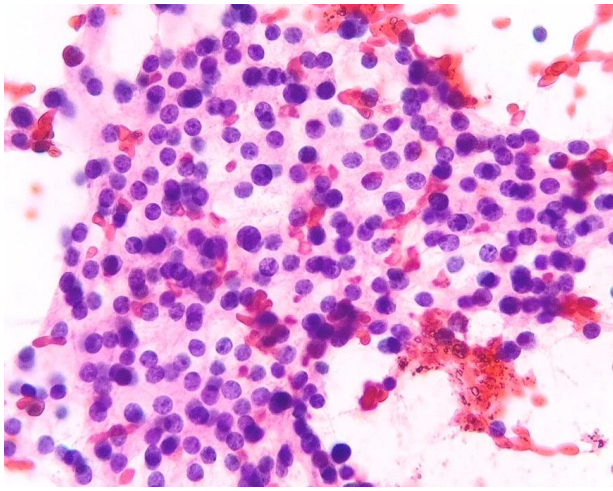
Metastases/Other Tumors

Most common sources of **metastases**: Lung, Breast, Melanoma, Colon, Kidney.

Lymphoma: Most common are Diffuse Large B-Cell Lymphoma (DLBCL) and Extranodal Marginal Zone B-Cell Lymphoma of Mucosa Associated Lymphoid Tissue (MALT). Usually in the setting of Chronic lymphocytic thyroiditis.

Rare primary tumors (pretty much anything! ;-): Mucoepidermoid carcinoma, Thymoma, SETTLE, etc…

Parathyroid Adenoma



Often clinically elevated serum Calcium and PTH.
Often deep/posterior, but may be intrathyroidal.

No **colloid.** Trabecular/packeted fragments.
May have triangular "wedge" shapes.
Hypercellular with **Small monotonous cells**.
Round nuclei. Salt and pepper chromatin.
Dispersed individual cells in the background.

IHC: (+) GATA3, PTH, Chromogranin (less so, synaptophysin)
(-) TTF1, Thyroglobulin

Cyst fluid and/or a needle rinse can be sent for PTH levels

Regarding "NIFTP"

Non-invasive follicular thyroid neoplasm with papillary-like nuclear features (NIFTP) cannot be reliably separated from the Follicular variant of PTC (FVPTC) by cytology. NIFTP usually has microfollicular or crowded architecture with mild nuclear changes.

To avoid overtreating NIFTP, which is indolent, follicular-patterned aspirates with nuclear changes that raise the possibility of FVPTC or NIFTP (e.g., mild nuclear changes) are best classified as "Follicular neoplasm."

Findings that take NIFTP out of consideration: 1) Papillary architecture, 2) Psammoma bodies, 3) frequent pseudoinclusions, 4) Sheet-like architecture

Atypia of Undetermined Significance ("AUS")

Indeterminate category for cases with one or more findings that raise concern for malignancy, but are insufficient for a more definitive diagnosis. Only low risk of malignancy.

Usually either ***mild*** **or** ***focal*** **nuclear changes** or **architectural changes**.
Only fair interobserver reproducibility.

Usually treated with follow up with **repeat FNA** and/or **molecular testing.**

Possible scenarios:
Sparsely cellular aspirate with lots of microfollicles or Hürthle cells (A specimen needs to be markedly cellular to Dx a FN)
Mild PTC nuclear changes (e.g., some enlargement, grooves, and/or powdery chromatin)
Psamommatous calcifications without other PTC findings
Atypical lymphocytes concerning for lymphoma

Suspicious for Carcinoma

Indeterminate category used when the features raise a strong suspicion for malignancy, but the findings are not conclusive for a definitive diagnosis. High risk of malignancy.

Mainly useful to maintain the high PPV of a "malignant" diagnosis.

Frequently treated with surgery (lobectomy vs total thyroidectomy).
Sometimes may employ molecular or repeat FNA, particularly when planning extent of surgery.

Molecular

For AUS or FN aspirates, molecular testing can be useful to risk stratify lesions to determine the risk of malignancy and next steps in treatment.

Papillary Thyroid Carcinoma: *MAPK Pathway: BRAF (most classic PTC's), V600E (most common) RAS (associated with follicular variant & NIFTP), ALK, NTRK fusions*

Medullary Carcinoma: *RET (think MEN2A&B)*

Follicular Neoplasms (FA & FN): *RAS point mutations most common, PPARG rearrangements (often with PAX8), PTEN*

Poorly Differentiated and Anaplastic: (*Additional*, late events, more aggressive tumors)
TP53, CTNNB1, TERT promoter mutations, ATK1, PIK3CA

The Bethesda System

Try to put all FNAs into one of these categories to help the clinicians determine the next appropriate steps in management.

Diagnostic Category		Risk of Malignancy	Management
I	Unsatisfactory/ Nondiagnostic	~15%	Repeat US-guided FNA
II	Benign	~5%	Clinical/US follow-up
III	AUS	~25%	Repeat FNA and/or Molecular testing
IV	Follicular Neoplasm	~30%	Lobectomy
V	Suspicious for Malignancy	~75%	Thyroidectomy
VI	Malignant	~97%	Thyroidectomy