Updates to The Bethesda System for Reporting Thyroid Cytopathology

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Disclosures

• No conflict of interest or disclosures
The Bethesda System for Reporting Thyroid Cytopathology

- Uniform terminology for reporting thyroid cytopathology
- Involved physicians caring for and managing pts with thyroid nodules
- Associated risk of malignancy (ROM) with diagnostic categories
- Atlas published in 2010
Triple Test for CARE

Radiographic Findings

Clinical Findings

Cytologic Findings

Excellent Patient Care
Thyroid FNA

US-guided FNA biopsy

ROSE—smears (Pap &/or Diff Quik stained)

Cytospin or Cell block

Molecular Tests

Afirma

ThyGenX

ThyroMIR

ThyroSeq

In House

Diagnosis
Cytology Preparations

• Smears
  • Pap-stained (optimal for nuclear features)
  • DQ-stained (fast for IA)

• Liquid base (Pap-stained)
  • ThinPrep
  • Cytospins

• Cell block (H&E)
### Bethesda System for Reporting Thyroid Cytopathology: Risk & Clinical Management

<table>
<thead>
<tr>
<th>Diagnostic Categories</th>
<th>Updated ROM (prior)</th>
<th>Management</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nondx/Unsatisfactory</td>
<td>5-10%</td>
<td>Repeat FNA with US</td>
</tr>
<tr>
<td>Benign</td>
<td>0-3%</td>
<td>Clinical &amp; US F/U</td>
</tr>
<tr>
<td>Atypia/follicular lesion of undetermined significance</td>
<td>~10-30% [5-15%]</td>
<td>Repeat FNA, molecular testing, or lobectomy</td>
</tr>
<tr>
<td>Follicular Neoplasm/ Hürthle Cell Neoplasm</td>
<td>25-40% [15-30%]</td>
<td>Lobectomy or molecular testing</td>
</tr>
<tr>
<td>Suspicious for Malignancy</td>
<td>50-75% [60-75%]</td>
<td>Lobectomy or near-total thyroidectomy</td>
</tr>
<tr>
<td>Malignant</td>
<td>97-99%</td>
<td>Lobectomy or near-total thyroidectomy</td>
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Cibas ES, Ali SZ. TBSRTC 2017: 2nd ed
# Bethesda System for Reporting Thyroid Cytopathology: Optional Notes

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<td></td>
</tr>
<tr>
<td>Follicular Neoplasm/ Hürthle Cell Neoplasm</td>
<td>25-40%</td>
<td>F/U includes follicular adenoma, follicular CA, FVPTC, &amp; NIFTP</td>
</tr>
<tr>
<td>Suspicious for Malignancy</td>
<td>50-75%</td>
<td>Suspicious for FVPTC &amp; NIFTP</td>
</tr>
<tr>
<td>Malignant</td>
<td>97-99%</td>
<td>~3-4% as PTC may prove to be NIFTP</td>
</tr>
</tbody>
</table>

Cibas ES, Ali SZ. TBSRTC 2017: 2nd ed
Indeterminate Follicular Lesions

• 10-25% of thyroid FNAs:
  • AUS/FLUS
  • Follicular Neoplasm
  • Suspicious for Malignancy
• “High risk” pts still undergo unnecessary surgeries for benign dz
• Can we improve outcomes
AUS/FLUS

• Atypia
  1. Nuclear atypia (?? PTC)
  2. Architectural atypia (microfollicles)
  3. Nuclear & Architectural atypia
  4. Hürthle cell atypia
  5. NOS-atypia cannot be classified

• Often includes compromised or less than optimal FNAs (scant cellularity, distortion artifact, obscuring blood)

• Variable rates in literature: Range 1-25% of thyroid FNAs
Colloid nodule vs AUS
Colloid nodule with cystic degeneration vs AUS
AUS

Single atypical group of cells with enlarged oval nuclei with fine chromatin, nuclear grooves, nuclear inclusions in an otherwise benign-appearing sample

Lobectomy: Adenomatoid nodule
AUS

Papillary-like fragments but doesn’t have nuclear features of PTC

Adenomatoid nodule
Hypocellular aspirate with microfollicles and scant colloid
Comment: The differential diagnosis for Hürthle cell lesion includes hyperplastic Hürthle cell nodule within chronic lymphocytic thyroiditis or an adenomatoid/multinodular goiter vs. Hürthle cell neoplasm (i.e., Hürthle cell adenoma vs. Hürthle cell carcinoma). Surgical excision would be needed for a more definitive subclassification, if clinically indicated.
Follicular/Hürthle Cell Neoplasms

- FNA can’t distinguish follicular adenoma from carcinoma (need tissue to evaluate for capsule & vascular invasion)
- Majority are benign lesions
- Rare cases of NIFTP
- ROM: 25-40%
  - Majority are follicular ca
  - Minority FV-PTC
- Management: lobectomy, molecular testing
Comment: The differential diagnosis for follicular neoplasm is follicular adenoma vs. follicular carcinoma. Surgical excision is required for a more definitive subclassification, if clinically indicated.
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Hürthle Cell Neoplasm

Comment: The differential diagnosis for Hürthle cell neoplasm is Hürthle cell adenoma vs Hürthle cell carcinoma. Surgical excision is required for a more definitive subclassification, if clinically indicated.
Follicular Neoplasm ??? PTC, FV

Comment: The differential diagnosis for follicular neoplasm is follicular adenoma vs. follicular carcinoma vs. follicular variant of papillary thyroid carcinoma. Surgical excision is required for a more definitive subclassification, if clinically indicated.
Hyalinizing Trabecular Tumor

Non Invasive Follicular Neoplasm with Papillary-like Nuclear Features (NIFTP)

- **Major Features on Histology**
  - Encapsulation of clear demarcation (no capsular or lymphovascular invasion)
  - Follicular growth pattern
  - Nuclear features of PTC: nuclear clearing, elongation, crowding, irregular contours, grooves, nuclear pseudoinclusions

- **Exclusion Criteria on Histology**
  - True papillae
  - Psammoma bodies
  - Infiltrative borders
  - Tumor necrosis
  - High mitotic activity
  - Other variants of PTC

Diagnostic criteria should not change for lesion that shows nuclear features of PTC but without papillae and/or psammoma bodies; “HASTY at this point”

Appropriate to comment in the report that NIFTP cannot be excluded

## Distribution of Cytologic Dx Preceding Infil / Inv FVPTC & NIFTP

<table>
<thead>
<tr>
<th>Cytologic Category</th>
<th>Infiltrative FVPTC or Invasive eFVPTC</th>
<th>Noninvasive eFVPTC or NIFTP</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Site A n=27</td>
<td>Site B n=24</td>
</tr>
<tr>
<td>Benign</td>
<td>0%</td>
<td>0%</td>
</tr>
<tr>
<td>AUS/FLUS</td>
<td>15%</td>
<td>0%</td>
</tr>
<tr>
<td>FN/SFN</td>
<td>11%</td>
<td>62.5%</td>
</tr>
<tr>
<td>Suspicious for PTC</td>
<td>44%</td>
<td>37.5%</td>
</tr>
<tr>
<td>Malignant</td>
<td>30%</td>
<td>0%</td>
</tr>
</tbody>
</table>

## Distribution of NIFTP & FVPTC Cases Among Three Institutions

<table>
<thead>
<tr>
<th>Thyroid Surgical Resections 2013-15</th>
<th>Institution A</th>
<th>Institution B</th>
<th>Institution C</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Total Resections</strong></td>
<td>2120</td>
<td>1714</td>
<td>1256</td>
</tr>
<tr>
<td>All PTC</td>
<td>1368 (65%)</td>
<td>386 (22%)</td>
<td>529 (42%)</td>
</tr>
<tr>
<td>• FVPTC (among PTC)</td>
<td>469 (34%)</td>
<td>177 (46%)</td>
<td>124 (23%)</td>
</tr>
<tr>
<td>• NIFTP (among FVPTC)</td>
<td>66 (14%)</td>
<td>69 (39%)</td>
<td>15 (12%)</td>
</tr>
<tr>
<td>Follicular/Hurthle cell CA</td>
<td>25 (1%)</td>
<td>13 (1%)</td>
<td>24 (2%)</td>
</tr>
<tr>
<td>Follicular/Hurthle cell adenoma</td>
<td>145 (18%)</td>
<td>770 (43%)</td>
<td>316 (25%)</td>
</tr>
<tr>
<td>Benign (hyperplasia+)</td>
<td>449 (21%)</td>
<td>595 (34%)</td>
<td>387 (31%)</td>
</tr>
</tbody>
</table>

No. of cases classified as NIFTP compared to FVPTC was highly variable (A=14%; B=39%, C=12%); the avg. ↓ ROM after the exclusion of NIFTP for all TBSRTC categories (A, 9.8%; B, 3.9%; C, 1.3%)

Contributing Factors in Indeterminate Thyroid FNAs

- Nature of the lesion
- Quality/Quantity of the specimen
- Limited clinical/radiographic information
- Inter/intraobserver variability in interpretation
- Availability/limitations of ancillary studies
- Cannot assess capsular/vascular invasion
Strategies for Improvement

- Improve quality of specimen-collection
  - Ultrasound guidance/operator expertise
  - Immediate assessment with additional sampling
- Clinical history/radiologic correlation
- Second opinion cytopathology review
- Ancillary tests
  - Immunohistochemistry (limited use)
  - Mutational Analysis
## Thyroid FNA Results by Pathologist Case Volume

<table>
<thead>
<tr>
<th>Source of Results</th>
<th>Benign</th>
<th>Atypical</th>
<th>Malignant</th>
<th>Non-Dx</th>
</tr>
</thead>
<tbody>
<tr>
<td>High-volume pathologists</td>
<td>68</td>
<td>13</td>
<td>1.5</td>
<td>17</td>
</tr>
<tr>
<td>Low-volume pathologists</td>
<td>50</td>
<td>32</td>
<td>0.6</td>
<td>17</td>
</tr>
<tr>
<td>Gharib &amp; Papinil &amp; Gharib &amp; Goellner</td>
<td>70</td>
<td>10</td>
<td>5</td>
<td>15</td>
</tr>
</tbody>
</table>

Summary of Concordance to Institutional Diagnosis

<table>
<thead>
<tr>
<th>Diagnosis</th>
<th>Concordance</th>
</tr>
</thead>
<tbody>
<tr>
<td>AUS/FLUS</td>
<td>36.4%</td>
</tr>
<tr>
<td>SFN/FN</td>
<td>36.5%</td>
</tr>
<tr>
<td>Suspicious</td>
<td>46.2%</td>
</tr>
<tr>
<td>Benign</td>
<td>62.4%</td>
</tr>
</tbody>
</table>

Baylor College of Medicine, Univ. of Colorado Hospital, Loyola University, Cleveland Clinic, Mayo Clinic, North Shore, PeaceHealth Lab., Univ. of Wisconsin
Results of 2\textsuperscript{nd} Thyroid FNA After AUS/FLUS Dx

- Unsatisfactory (5.2%)
- Benign (42.7%)
- AUS/FLUS (38.5%)
- SFN (5.2%)
- Suspicious (4.2%)
- Malignant (4.2%)

Ho AS, et al. Thyroid. 2014:24; 832-839
Cost-Effectiveness Analysis of Repeat FNA in AUS Thyroid FNAs: Decision Tree

Repeat FNA was cost-effective in AUS thyroid FNAs.

Immunophenotyping: Limited Use in Thyroid FNAs

- Galactin-3, HBME-1, CK-19, CD44, Cyclin D1 & 3 have wide range of sens, spec, PPV, & NPV; ? use in Indeterminate FNAs
- BRAF just validated on FNA cell block preparations at MDACC (select cases)
- Useful to confirm dx of uncommon tumors such as MTC, parathyroid lesions, metastatic carcinoma, & lymphoma

Zhang X. Arch Path Lab Med. 2015;139:1484-1490
Follicular vs Parathyroid

Intrathyroid parathyroid
Impact of Mutational Testing

- Molecular testing has a ↑PPV of 87-95% for predicting thyroid ca (exceeds the 50-75% by cytology)

- **BRAF V600E, RET-PTC, and PAX8-PPARγ** are ↑↑associated with malignancy

- **RAS** mutation has lower risk of thyroid malignancy (seen in adenomas & NIFTP)

- ~70% of thyroid carcinomas have mutations
ATA Thyroid Nodule/DTC Guidelines

- Use of molecular marker on indeterminate FNA specimens should not be intended to replace other sources of information or clinical judgment
- If molecular testing is considered, pt should be counseled regarding the potential benefits & limitations of testing & uncertainties in long-term clinical implications

Thyroid Molecular Tests

- Commercially Available Tests (Not inclusive)
  - Asuragen - MiRInform test - rule IN
  - ThyroSeq - rule IN
  - Rosetta GX Reveal - rule IN
  - Interpace Diagnostics - rule IN / rule OUT
  - Afirma-Veracyte - rule OUT
## Molecular Mutations in Thyroid Neoplasms

<table>
<thead>
<tr>
<th>Molecular</th>
<th>FA</th>
<th>FTC</th>
<th>NIFTP</th>
<th>PTC</th>
<th>PD Ca</th>
<th>Anaplastic Ca</th>
<th>All Thyroid Ca</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>BRAF</strong></td>
<td>&lt;1%</td>
<td>&lt;1%</td>
<td>0%</td>
<td>45%</td>
<td>15%</td>
<td>10%</td>
<td>42%</td>
</tr>
<tr>
<td><strong>RAS</strong></td>
<td>20-40%</td>
<td>45%</td>
<td>46%</td>
<td>10%</td>
<td>35%</td>
<td>55%</td>
<td>16%</td>
</tr>
<tr>
<td><strong>PAX8-PPARγ</strong></td>
<td>7%</td>
<td>30%</td>
<td>-</td>
<td>5%</td>
<td>0</td>
<td>-</td>
<td>4%</td>
</tr>
<tr>
<td><strong>RET-PTC</strong></td>
<td>0</td>
<td>0</td>
<td>-</td>
<td>20%</td>
<td>-</td>
<td>-</td>
<td>8%</td>
</tr>
</tbody>
</table>

*Courtesy of Dr. Gregg Staerkel & Thyroid (2013) 23:1256-1262*
Comparison of Currently Available Molecular Tests for Indeterminate Thyroid FNA Specimens

<table>
<thead>
<tr>
<th></th>
<th>Afirma</th>
<th>ThyGenX</th>
<th>ThyroMIR</th>
<th>ThyroSeq</th>
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<tr>
<td><strong>Methodology</strong></td>
<td>mRNA gene expression</td>
<td>Multiplex PCR by sequence-specific probes</td>
<td>MicroRNA expression</td>
<td>Next-Generation sequencing</td>
</tr>
<tr>
<td><strong>Test report</strong></td>
<td>Benign/ suspicious</td>
<td>Specific gene mutation/ translocation</td>
<td>Negative/ Positive</td>
<td>Specific gene mutation/ translocation</td>
</tr>
<tr>
<td><strong>Specimen collection</strong></td>
<td>2 dedicated FNA passes</td>
<td>1 dedicated FNA pass; at least 50 ng of cellular material</td>
<td>Same as ThyGenX</td>
<td>1-2 drops from 1st pass, if adequate cellularity on smear slide</td>
</tr>
<tr>
<td><strong>Strength</strong></td>
<td>High NPV</td>
<td>High PPV</td>
<td>Good NPV &amp; PPV when combined with ThyGenX</td>
<td>High NPV &amp; PPV</td>
</tr>
<tr>
<td><strong>Limitation</strong></td>
<td>Low PPV $4875 for Afirma GEC &amp; MTC $975 Afirma MTC $475 Afirma BRAF</td>
<td>Low NPV $1675 for ThyGenX alone ($300 pt)</td>
<td>Limited validation data $3300 for ThyraMIR (reflex test)</td>
<td>Limited validation data $3200</td>
</tr>
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*Zhang M, Lin O. Arch Pathol Lab Med. 2016;140:1338-1344*
## Insurance Payments for Molecular on Thyroid FNAs

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Things to Consider

- Is lab CLIA certified & state licensed
- How easy is it to incorporate into the workflow of the lab & ~ number of cases tested annually
- Is there special rinse sol’n, shelve life of the media, shipping requirements
- What is the turn-around time
- Is there a pathology interpretation of the cytology
- Cost, Billing, Coding
- Is it cost effective
Proced With Caution

- Pts who are not surgical candidates do not need molecular testing
- Remember ~50% of AUS are reclassified as benign on repeat FNA
- Not all thyroid ca have molecular mutations or rearrangements
- Molecular testing may be useful in select AUS/FLUS & FN, but no significant benefit in suspicious for malignancy
Conclusions

- Thyroid FNA is the first line test for evaluating thyroid nodules
- 10-25% of thyroid FNAs are Indeterminate
- IPOX has limited role in Indeterminate FNAs
- Molecular testing may be considered in pts with indeterminate FNAs to help guide management
- Molecular testing should not be REFLEX test
- Molecular testing on thyroid FNAs should complement clinical, radiographic, & cytology interpretations
Thank You