PULMONARY PATHOLOGY JOURNAL CLUB
(September 2024 Articles)
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Discussion articles

Hariri LP et al. Endobronchial optical coherence tomography as a novel method for in vivo microscopic assessment of interstitial lung abnormalities. Am J Respir Crit Care Med 2024; 210:671-677.

Purpose: Investigate if the presence of definite fibrosis on endobronchial optical coherence tomography (EB-OCT) predicts for progressive fibrosis in patients with interstitial lung abnormalities (ILAs) incidentally discovered on CT in patients undergoing resection of lung nodules.

Methods:

- Single-center prospective study
- Inclusion criteria: 1) incidentally discovered ILA and concurrent lung nodule on thin-section CT, 2) undergoing elective bronchoscopy and surgical resection, 3) presence of ILA within and away from planned resection
    - Exclusion criteria: Pre-existing diagnosis of interstitial lung disease (ILD)
- EB-OCT performed during bronchoscopy, sited selected based upon areas of CT abnormality
- Pathologist 1, blinded to histology, classified EB-OCT images, using descriptive terms
    - "Microscopic definite fibrosis" = subpleural and/or airway-centered fibrosis with microscopic traction bronchiectasis or honeycombing
- EB-OCT then compared against histology that was interpreted by another pathologist(s?), who was blinded to EB-OCT
- Patients clinically followed for at least 5-years

Results:

- 8 patients met inclusion criteria
- Patient characteristics listed in Table 1
    - All patients ≥ 60 years old
    - Males (n = 7) >> Females (n = 1)
    - All former smokers
- 6 of 8 patients suffered disease progression (CT +/- respiratory symptoms) within 3 years
    - All had "definite fibrosis" on EB-OCT; presence is 100% predictive of disease progression
    - Histology reported as collection of descriptive terms that were also used in EB-OCT interpretation
    - 2 had "early UIP" on both EB-OCT and histology and later diagnosed as IPF
- 2 patients without "definite fibrosis" showed no radiographic of symptomatic progression
    - Traction bronchiolectasis and honeycombing not seen on histology

Take-home message: EB-OCT "definite fibrosis" (that corresponds with histologic findings of subpleural/airway-centered fibrosis with traction bronchiolectasis and/or honeycombing) predicts for disease progression with asymptomatic ILA.

Liu M et al. Quantitative measurement of HER2 expression in non-small cell lung cancer with a high-sensitivity assay. Mod Pathol 2024; 37:100556.

Purpose: Quantify HER2 expression in NSCLCs using a high-sensitivity HER2 (HS-HER2) assay to establish the limit of detection (LOD = lowest detectable signal that can be distinguished above noise), limit of quantification (LOQ = lowest signal that can be quantified with a reliable degree of precision), and limit of linearity (LOL = signal level beyond which the calibration curve is no longer linear), and ultimately, determine the proportion of cases with detectable HER2 levels

Methods:

- Immunofluorescence antibodies for HER2 and cytokeratin AE1/AE3 applied to NSCLC tissue microarrays (total *n* = 741)
    - 3 TMA cohorts retrospectively collected over different time periods
    - Clinicopathologic characteristics available for TMA cohorts: Demographics, clinical stage at diagnosis, tumor type, PD-L1 status, outcomes
        - *ERBB2* genomic alteration status unknown
    - TMAs stained in batches along with at least 1 calibration cell line microarray (CMA) per batch
        - Calibration CMAs have known expression levels of HER2
- Images acquired on PM-2000 and analyzed by AQUA method of quantitative immunofluorescence (QIF)
- HER2 QIF scores for NSCLC converted to units of attomoles (amol/mm^2) using calibration curves from CMAs associated with each batch
- Calculated LOD and LOQ for NSCLC HER2 expression, using the calibration CMAs as standard curve
    - To test the determined LOD and LOQ, tissue was serially sectioned from the TMAs, test re-run, and the results compared and averaged
- Immunohistochemistry for HER2 also performed using standard (breast cancer) assay and high sensitivity assay

Results:

- Results showed high reproducibility using serial sectioned tissue
- In total, 63% of cases had HER2 levels above the LOD
    - Of these, 17% above LOQ
    - ~0.5% above the limit of linearity
- HER2 expression stratified against clinicopathologic characteristics
    - Adenocarcinomas > squamous cell carcinomas
    - Females > Males
    - PD-L1-low > PD-L1-high
- No association between HER2 expression and outcomes
- HS-HER2 IHC assay shows stronger staining and would be scored differently (S.Fig5)

Take-home message: These findings suggest that a substantial proportion of NSCLCs have detectable and quantifiable levels of HER2 expression, which offers a potential target for HER2 antibody-drug conjugates (e.g. Trastuzumab deruxtecan).

Yamada Y et al. Immunohistochemistry for YAP1 N-terminus and C-terminus highlight metaplastic thymoma and high-grade thymic epithelial tumors by different staining patterns. Virchows Arch 2024; 485:461-469.

Purpose: Evaluate the utility of YAP1 N-terminus (YAP1[N]) and YAP1 C-terminus (YAP1[C]) immunohistochemistry in the diagnosis of metaplastic thymoma

Methods:

- On 11 (of 14) metaplastic thymomas, RNA sequencing for *YAP1::MAML2* fusion product evaluated using ArcherDx FusionPlex Solid Tumor Panel and FISH for *YAP1* break-apart using *MAML2* breakpoint region
- Immunohistochemistry for YAP1(N) and YAP1(C) was applied to metaplastic thymomas (n = 14) and 104 other types of thymic epithelial tumors (tissue microarrays of 95 tumors and 3 additional resection slides each of type A thymoma, type B3 thymoma, and thymic carcinoma)
    - Nuclear staining in ≥50% of cells = positive result
    - Both spindle and epithelial components evaluated in metaplastic thymomas
- POU2F3 antibody applied to metaplastic thymomas only

Results:

- RNA sequencing or FISH successful in 6 (of 11) cases of metaplastic thymoma, showing *YAP1::MAML2* rearrangement and/or *YAP1* breaks
    - Tests failed in 5 cases due to poor DNA/RNA quality (block age ≥ 8 years)
- 14 of 14 (100%) of metaplastic thymomas YAP1(N)-positive (range: 80-100%); YAP1(C)-negative (range: 0-40%)
    - Staining for YAP1(C) limited to spindle cells, which showed weak reactivity
- Table 1 shows patterns of immunoreactivity in various thymic epithelial tumor types
- Sensitivity and specificity for YAP1(N)-positive/YAP(C)-negative profile for metaplastic thymoma: 100% and 95% (99/104), respectively
    - 5 non-metaplastic thymoma cases showed this pattern of immunoreactivity: 1 AB thymoma, 1 B2 thymoma, 1 B3 thymoma, 2 thymic carcinomas
- YAP1(C)-negative only has sensitivity of 100% and specificity of 76%
- All metaplastic thymomas negative for POU2F3

Take-home message: If you are going to invest in one of these antibodies, YAP1(C) is probably sufficient if you ever find that you need to establish a diagnosis of metaplastic thymoma on small biopsies, but you can increase your specificity by performing both and these antibodies may be employed in combination as surrogate markers for *YAP1::MAML2* rearrangement when genetic testing is not possible due to poor DNA/RNA quality.

Gill RR et al. The international association for the study of lung cancer mesothelioma staging project: proposals for revisions of the "T" descriptors in the forthcoming ninth edition of the TNM classification for pleural mesothelioma. J Thorac Oncol 2024; 19:1310-1325.

Purpose: Analyze pleural thickness measurements on CT and the existing 8th edition clinical T staging descriptors to develop recommendations for inclusion in the 9th edition TNM staging of pleural mesothelioma.

Methods:

- Collect patients with pleural mesothelioma to analyze clinical information, CT data elements, pathologic stage, treatment, and survival
    - Inclusion criteria for clinical T: Histologic/Cytologic diagnosis of mesothelioma and M0 (8th edition TNM)
    - Inclusion criteria for pathologic T: Underwent surgical resection, did not receive neoadjuvant therapy, and information regarding status of fissure available if pT1
- Quantitative image assessment: Maximum fissure thickness (Fmax) and maximum pleural thickness measured by dividing chest into approximate thirds at the upper, middle, and lower hemithorax (Psum = pmax1 + pmax2 + pmax3).
    - Involvement of fissure defined as Fmax > 5 mm
    - Maximum diaphragmatic thickness (Dmax) of interest, but measurement missing in about half of cases, so not included in analysis
- Qualitative image assessment performed, using 8th edition clinical staging criteria of invasion into adjacent structures assessed
    - Excluded assessment of non-transmural invasion into diaphragm, invasion into lung, and endothoracic fascia invasion due to evidence that these cannot be accurately assessed on imaging → loss of distinction between 8th edition cT1/cT2
- Statistical analysis performed to maximize differences in overall survival among the subgroups

Results:

- 1689 patients met inclusion criteria for analysis of cT; 457 met inclusion criteria for pT
- Fissural involvement an independent predictor of worse prognosis regardless of Psum
- Recursive partitioning identified the optimal cutpoints of Psum at 12 and 30 mm, which in combination with extent of invasion yielded 4 prognostic subgroups
- Table 2 data highlights the proposed new T categories

Take-home message: Based upon the authors findings, the pathologic T staging is relatively unchanged, but size criteria as the sum of maximum pleural thickness at 3 levels and presence or absence of fissure involvement on chest imaging are now proposed components for clinical T staging.

Articles for notation

Neoplastic lung disease

Alirezaie N et al. Exomic and epigenomic analysis of pulmonary blastoma. Lung Cancer 2024; 195:107916.

Take-home message: Whole-exome sequencing and DNA methylation profiling was performed on 8 pulmonary blastomas from 6 different patients. At least 1 pulmonary blastoma from all 6 patients had somatic biallelic inactivation of *DICER1* and clustering of pulmonary blastoma with other *DICER1*-mutated tumors using methylation analysis, suggesting that *DICER1* is a key driver. *CTNNB1* pathogenic variants co-occurred in 5 of 6 patients, indicating a possible synergistic relationship between *DICER1* variants and activation of the WNT signaling pathway in the formation of these tumors.

Bille A et al. The international association for the study of lung cancer mesothelioma staging project: proposals for the "N" descriptors in the forthcoming ninth edition of the TNM classification for pleural mesothelioma. J Thorac Oncol 2024; 19:1326-1338.

Take-home message: In preparation for the 9th edition of the TNM classification of diffuse pleural mesothelioma, the IASLC Staging and Prognostic Factors Committee wished to determine whether the nodal subgroupings of the 8th edition required revision. They, therefore, analyzed overall survival in 2836 patients with respect to N subgroups. A novel finding was that pleural thickness (refer to Gill citation below for details) predicts for nodal involvement, and Figures 2 and 4 nicely illustrate the differences in survival between pathologic N categories, which were ultimately deemed as needing no change in the upcoming edition.

Bontoux C et al. Reproducibility of c-Met immunohistochemical scoring (clone SP44) for non-small cell lung cancer using conventional light microscopy and whole slide imagining. Am J Surg Pathol 2024; 48:1072-1081.

Take-home message: As the title reflects, the authors aimed to assess the inter- and intra-observed reproducibility of c-Met expression scoring by immunohistochemistry for 110 cases of non-small cell lung carcinoma (NSCLC) by 6 pathologists, using 1) 3-tiered and 2-tiered scoring systems, and 2) conventional light microscopy (CLM) and whole slide imaging (WSI). Interobserver variability in scoring was moderate to excellent with WSI resulting in better agreement than CLM, and agreement was better for senior pathologists compared to junior pathologists. Discordances in scoring were attributed to staining heterogeneity in large surgical specimens and/or variability in control staining leading to over-interpretation of 3+ staining.

Devins KM et al. Large and extensive multilocular peritoneal inclusion cysts lack genomic alterations and follow an indolent clinical course despite rare recurrences. Am J Surg Pathol 2024; 48:1177-1184.

Take-home message: This work details the histologic and patient clinical characteristics of 20 peritoneal inclusion cysts (PICs), including 8 large solitary lesions and 12 multifocal lesions. Two patients, both with multifocal PICs, experienced recurrence following resection,

but all patients are otherwise alive with no evidence of disease. BAP1 was only performed on the recurrent cases and was retained and 5 cases (2 solitary, 2 multifocal) underwent genetic testing with no fusions/genetic alterations detected, and for these reasons, the authors concluded that these lesions are associated with benign outcomes.

Gorbokon N et al. PAX8 expression in cancerous and non-neoplastic tissue: a tissue microarray study on more than 17,000 tumors from 149 different tumor entities. Virchows Arch 2024; 485:491-507.

Take-home message: This is a study employing *monoclonal* PAX-8 antibodies. Relevant to our interests, no pulmonary squamous cell carcinoma, neuroendocrine tumors, thymomas, or epithelioid mesotheliomas shows staining, while <1% of lung adenocarcinoma showed staining (moderate) and 18.5% of biphasic mesotheliomas showed staining, including approximately 4% with strong staining (The component with staining was not indicated… I am assuming that it was the sarcomatous component since epithelioid mesotheliomas were negative).

Naso J et al. Predictive value and molecular correlates of MYC immunohistochemistry and copy number gain in non-small cell lung carcinomas treated with immunotherapy. Lung Cancer 2024; 195:107927.

Take-home message: Here, the authors retrospectively assessed *MYC* copy number gain and/or MYC immunoreactivity in 82 patients with non-small cell lung carcinomas who were treated with immunotherapy in an effort to predict response to immunotherapy. Eleven percent of cases showed *MYC* copy number gain, while 70% of cases show positive MYC staining by immunohistochemistry and these results correlated poorly with one another. MYC staining (≥40%) was significantly associated with current or former smoker status, *KRAS* mutation, and *KRAS* and *TP53* co-mutation, and the combination of positive MYC staining and negative PD-L1 (<1%) was associated with poor overall and progression free survival following immunotherapy.

Nowak AK et al. The international association for the study of lung cancer pleural mesothelioma staging project: proposal for revision of the TNM stage groupings in the forthcoming (ninth) edition of the TNM classification for pleural mesothelioma. J Thorac Oncol 2024; 19:1339-1351.

Take-home message: For me, this article essentially provided a summary of clinical rationale detailed in the related articles about pleural mesothelioma T and N staging for the 9th edition (Gille and Bille references above, respectively).

Rerkpichaisuth V et al. The utility of the lineage specific immunohistochemical stains SATB2, CDX2, and villin, and the mucin glycoproteins MUC2, MUC5AC, and MUC6 to distinguish pulmonary invasive mucinous adenocarcinoma from metastatic colorectal carcinoma. Hum Pathol 2024; 151:105627.

Take-home message: The authors applied the above listed antibodies to tissue microarrays of 34 pulmonary non-mucinous adenocarcinomas, 31 pulmonary mucinous

adenocarcinomas, and 32 colorectal carcinomas. Table 1 details the frequency with which each stain was identified with any expression, >50%, and >90% for each tumor type. The authors conclude that cytokeratin 7 is the "most useful" marker in distinguishing between mucinous adenocarcinoma of the lung and metastatic colorectal carcinoma, but as you well know from practice, a panel of stains is most helpful (and if you're lucky, clinical history) in deciding between these possibilities.

Roma L et al. Tracing tumor heterogeneity of pleomorphic carcinoma of the lung. J Thorac Oncol 2024; 19:1284-1296.

Take-home message: Forty-two histologically distinct tumor areas from 20 patients with pleomorphic carcinomas underwent comprehensive molecular analysis. The authors found that sarcomatoid carcinomas from adenocarcinoma- and squamous cell carcinoma-like tumors have different genetic landscapes and are more similar to their matched epithelial components than each other. My 2 cents: While a tumor may qualify for a diagnosis of "pleomorphic carcinoma" based upon the WHO classification, consider avoiding this generic term in your reporting when there is clear morphologic or immunohistochemical evidence of a more differentiated component.

Shang Z et al. Single-cell transcriptome analysis reveals 2 subtypes of tumor cells of sclerosing pneumocytoma with distinct molecular features and clinical implications. Mod Pathol 2024; 37:100560.

Take-home message: This work offers a detailed look into the cuboidal surface cells and round stromal cells of sclerosing pneumocytomas obtained from 4 patients with no nodal disease, recurrence, or metastasis, using single-cell RNA sequencing and spatial transcriptomics, to characterize the morphologic manifestations and biological behavior. In brief, they found that surface cells are reminiscent of type I alveolar epithelial cells (AT2C) and round cells show a mesenchymal-epithelial dual-phenotype (MEDP), results that are perhaps not surprising based upon what we already know from their distinct immunophenotypes. Both cell types exhibit prominent stemness and MEDP possess a propensity for epithelial-mesenchymal transition, while AT2C are indeed clonal and neoplastic rather than entrapped benign, reactive pneumocytes.

Non-neoplastic lung disease

Varghese NP et al. An interdisciplinary consensus approach to pulmonary hypertension in developmental lung disease. Eur Respir J 2024; 64:2400639.

Take-home message: This document is intended to provide details of a multidisciplinary approach to the diagnosis and management of neonates and young infants suffering from pulmonary hypertension. The role of lung biopsy is discussed as is the optimal size/processing of an adequate surgical lung biopsy, but the histologic features of developmental lung diseases associated with pulmonary hypertension are not detailed here.

Zen Y et al. Idiopathic hyalinizing fibrosclerosis: a systemic steroid-resistant condition distinct from IgG4-related disease. Hum Pathol 2024; 151:105638.

Take-home message: This small case series described the clinical, serologic, and outcome data for 3 patients, who underwent biopsy of mass-forming fibroinflammatory lesions involving multiple sites, including the lung and mediastinum. While the histologic findings overlapped with IgG4-related disease by showing hyalinizing fibrosis, there was no increase in serum of IgG4 concentrations or IgG4-positive plasma cells by immunohistochemistry, and the biopsies lacked storiform fibrosis and obliterative phlebitis. The authors conclude by proposing the term "Idiopathic hyalinizing fibrosclerosis" for this lesion, which to my eye, looks like pulmonary hyalinizing granuloma.

Case Report

Alawi S et al. IgG and plasma viscosity as markers of disease activity in primary Sjogren's syndrome-related lymphocytic interstitial pneumonia. Thorax 2024; 79:886-888.

Take-home message: A 33-year-old woman was diagnosed with primary Sjogren's syndrome-associated lymphocytic interstitial pneumonia following lung wedge biopsy and multidisciplinary discussion of her clinical and radiographic data and results of serologic testing. She was treated with immunomodulatory therapy. Serial IgG levels and plasma viscosity levels mirrored her physiologic and radiographic improvement, indicating a potential role for these blood biomarkers in monitoring disease activity and treatment response.

Letter to the Editor

Ding CC et al. Regarding NF2 (Merlin) status in mesothelioma of uncertain malignant potential (MUMP) or complex mesothelial tumor of the tunica vaginalis. Am J Surg Pathol 2024; 48:1198-1200.

Take-home message: In this follow-up letter to their April 2024 article on MUMP/complex mesothelial tumor of the tunic vaginalis, the authors report the results of Merlin immunohistochemistry in 9 cases. Seven cases showed intact staining, while the antibody failed in 2 cases. The authors use this data as further support that MUMP is an indolent tumor.

Review Article

Klebe S et al. Lung cancer caused by asbestos: what a reporting pathologist needs to know. Lung Cancer 2024; 195:107849.

Take-home message: The author's message to the pathologist can be summed up in a single sentence from the discussion, "Morphologic assessment in isolation has limited to no utility to diagnose [asbestos-induced lung cancer]."