Pulmonary Journal Club- January 2022

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Articles for Discussion

Postdeployment Respiratory Syndrome in Soldiers with Chronic Exertional Dyspnea. Gutor et al. Am J Surg Pathol. 2021;45(12):1587-1596.

Introduction: An increased frequency of respiratory symptoms has been documented in military personnel deployed to Southwest Asia compared with nondeployed personnel. In a recent study in 380 deployed military members with respiratory symptoms postdeployment, most had evidence of definable entities (e.g. asthma), but a large percentage (32.1%) had unexplained exertional dyspnea. In other study, 77% of soldiers with unexplained symptoms were diagnosed with constrictive bronchiolitis based on findings from lung Bx, despite the lack of fixed obstruction on PFTs.

Methods: Comprehensive, in-depth histopathologic evaluation of lung Bx from 50 soldiers developed persistent unexplained respiratory symptoms (exercise intolerance and exertional dyspnea) post-deployment to Southwest Asia. 17 age-matched, nonsmoking controls (lungs rejected for lung Tx). Pathologic evaluation + quantitative histomorphometry was performed for evaluation of inflammation and pathologic remodeling of small airways, pulmonary vasculature, alveolar tissue and visceral pleura. Scanning HRCT (prone and expiratory phase) and PFTs: spirometry, lung volumes, and DLCO

Criteria for inclusion: at least 2 FFPE blocks available with 3 or more small airways in cross section, along with minimal hemorrhagic / artifactual damage. Each patient had lung Bx findings reported to be consistent with a diagnosis of constrictive bronchiolitis without evidence of other distinct lung pathologies, including: granulomatous diseases, interstitial lung disease, or COPD.

Histopathological analysis and morphometry: 4 serial sections, stained for: H&E, PAS, VVG, PicroSirius red. Additional serial sections for IHC targeting immune inflammatory cells (e.g. CD4, CD8 T cells), structural cells (smooth muscle or endothelial cells), and elastin. Small airways <2mm in diameter were selected for examination. Cartilaginous or larger airways with submucosal glands were excluded. They measured multiple variables: 1) epithelial height, (2) lamina propria thickness, (3) smooth muscle thickness, (4) adventitia thickness, (5) collagen and elastin content, and (6) immune inflammatory cell (CD4 and CD8 lymph and neut) infiltrations. Distal pulmonary were assessed for media and adventitia thickness, and wall-to-lumen ratio on VVG. Alveolar tissue was analyzed for blood capillary density, collagen content, and elastin content.

Results: Most soldiers were young (mean: 36 y), male (94%), nonsmokers. All affected soldiers reported exposure to environmental hazards during deployment. 46% reported exposure to smoke from a sulfur-mine fire in Iraq. Soldiers also reported exposures to burn pits (42%), dust storms (16%), diesel exhaust (6%), human waste and/or combat smoke (16%). Initial PFTs: 60% Normal… 34% DLCO reduction (<80% pred), 4% mild obstruction (FEV1/FVC< 0.7) and 8% had mild restriction (TLC<80%). Follow up (58% available info, up to 15y), all had persistent symptoms, one died of ILD (unspecified). At follow up all had reduction in TLC, of those 48% TLC reduction >10% ($p<0.001$); 24% reduction in FVC.

Non-morphometric pathological evaluation: All cases → fibrous remodeling of bronchiolar walls c/w constrictive bronchiolitis. 24% cases macrophage infiltration within the bronchiolar lumen and peribronchiolar alveoli suggested a component of respiratory bronchiolitis.

Morphometric results: Bronchioles from soldiers showed ↑ thickness of the lamina propria ($p<0.05$), hypertrophy of smooth muscle ($p<0.001$), and increased collagen density in the subepithelium($p<0.001$). No differences in: epithelial height, adventitial thickness, or elastin content within the subepithelium. >3-fold increase in CD4 and CD8 T cells within the airway walls ($p<0.001$). 64% had B cell-containing lymphoid follicles in close proximity to small airways ($p<0.001$). Distal pulmonary arteries: ↑ medial thickness due to smooth muscle hyperplasia/hypertrophy ($p<0.001$). ↑adventitial thickness due to edema and fibrosis ($p<0.001$), wall-to-lumen ratio ($p<0.001$). Interalveolar septa (IAS) revealed a reduction in blood capillary density ($p<0.001$). In addition, the IAS showed a diffuse fibrotic phenotype with increased deposits of collagen and elastin($p<0.001$), but without distortion of alveolar architecture. Pathologic changes of visceral pleura, (92%) of soldiers($p<0.001$): pleural thickening (46%), mononuclear inflammatory cells (74%), increased collagen deposition (88%).

Unsupervised hierarchical clustering based on morphometric parameters showed complete separation of these 2 groups. Clinical parameters (↓DLCO and air trapping on CT scans) and reported exposures (sulfur-mine or burn pits) were equally represented within the subclusters of affected soldiers. They did not identify any specific correlations between clinically detected abnormalities (↓DLCO or air trapping) or environmental exposures (tobacco smoke, sulfur-mine, or burn pits) and individual pathologic features identified in the lungs of affected soldiers. Logistic regression analysis parameters with the most discriminatory power for separating symptomatic soldiers from controls: (1) CD4 and CD8 T cell infiltration of small airway walls, (2) medial thickness in distal pulmonary arteries, (3) reduced density of blood capillaries in alveolar septa, and (4) pleura pathology.

Discussion: Previous report that focused on pathologic changes in bronchioles underestimated the nature and extent of lung pathologic remodeling in this cohort. They propose **postdeployment respiratory syndrome (PDRS)**: (1) history of deployment in Southwest Asia and Afghanistan, (2) inhalational exposures, (3) chronic respiratory symptoms (↓exercise tolerance and exertional dyspnea) that develop and persist in the postdeployment period, and (4) lung pathology that affects all distal lung compartments. In contrast to classic constrictive bronchiolitis, which can result from exposure to toxic gases or occur secondary to autoimmune disorders, constrictive bronchiolitis in PDRSs was relatively mild in most cases and not accompanied by fixed airways obstruction and/or air-trapping on chest CT.

Similar findings of normal spirometry and chest-CT scans were described in Bx-confirmed case series of constrictive bronchiolitis from Iranian survivors of sulfur mustard gas exposure and studies of flavoring/popcorn factory workers. They hypothesize that changes in pulmonary vasculature, along with diffuse fibrotic remodeling, may explain the reduced exercise tolerance and exertional dyspnea in these soldiers. The reported environmental exposures were varied. Hard to determine which factor contributed the most to the pathology.

Take home point: Clinical syndrome is relatively homogenous as cluster analysis separates the syndrome from controls. Persistent immune cell infiltration, along with pathologic changes in blood vessels and pleura, are conserved aspects of lung pathology in affected soldiers.

Potential risk for bias: Case control study. No disease controls. Retrospective. Sample size. Follow up is not homogeneous, and incomplete, therefore it is hard to evaluate progression. Biased samples as all had constrictive bronchiolitis, milder phenotypes may had been excluded.

Cryobiopsy as a reliable technique for the preoperative identification of micropapillary/solid components in early-stage lung adenocarcinoma. Suzuki et al, Lung Cancer 162 (2021) 147-153.
Dr. Yasushi Yatabe research group

Introduction: Micropapillary (MIP) and solid (SOL) subtypes of early-stage lung adenocarcinomas (ADC) have been found to be associated with:

1) High risk of occult lymph node (LN) metastasis: cN0 ADC with MIP/SOL > 5%
2) Postoperative lymph node upstaging: in cN0 diseases, and/or
3) Local recurrence: resulting in ↓ disease free (DFS) and overall (OS) survival in stage I ADC

MIP and SOL are classified as high-grade (HG) subtypes. Recently, the IASLC proposed a new grading system and defined tumors with 20% or more of HG subtypes as grade 3 (poorly differentiated), which is associated with poor DFS and OS in stage I lung adenocarcinoma. Therefore, preoperative identification of these components may influence the decisions of treatment strategy:

1) additional LN evaluation
2) indication for limited resection
3) the extent of LN dissection

Problem: Conventional Bx specimens are insufficient for identifying HG subtypes, especially MIP components. The performance of CT-guided needle Bx and R-EBUS-guided TBBx for detecting HG ADC, has shown: 1) High specificity (MIP, 97.4%; SOL, 97.7%), but very low sensitivity (MIP, 7.8%; SOL, 14.6%). CryoBx can collect larger tissue samples with fewer crush artifacts than conventional forceps Bx.

Objective: To evaluate the feasibility of using cryoBx for the preop identification of MIP/SOL components

Methods: Single center study. Retrospective. 115 consecutive patients with clinical IA lung AD (Dx by cryoBx). Included cases had transbronchial brushing, forceps Bx, and/or needle aspiration before cryoBx (at the end of the examination).

- Pathological review. 2 independent readers. Blinded to clinical data and outcomes. Results by consensus. Threshold for MP/SOL components: 5%. The % of each component recorded in 5% increments. Primary predominant subtype was selected (highest %).
- Concordance rate of the HG subtypes between the cryoBx specimens and surgical specimens
- Clinical factors for predicting HG subtypes
- Clinicopathological parameters that affect the ability of cryoBx to accurately identify HG ADCs.
- Classification the % of MIP/SOL components in surgical specimens: < 5%, 5–39%, or ≥ 40%

Results: The median age was 68 years (range 33–86), with slight female predominance (57.4%). Median tumor size on preop CT was 23.1 mm (range 10.1–59.8), and median consolidation size was 15.6 mm (range, 3.3–29.9). Distribution of c-stages IA1(12.1%), IA2(55.7%), and IA3 (32.2%). The predominant histologic subtypes (surgical spec): papillary (34.8%), acinar (33.0%), lepidic (26.1%), SOL (6.1%), No MIP predominant. Patients with severe bleeding: 1.7%. No cases of life-threatening bleeding.

Component		# of cases (n=115)	Sensitivity	Specificity
Primary predominant pattern				
	MIP (no cases)	0	-	-
	SOL	7	86% [CI]: 57–96%	99% CI: 97–99.7%
Secondary predominant				

	MIP	6	83% CI: 46–97%	89% CI: 87–89.7%
	SOL	5	0% CI: 0–38%	94 CI: 93.6–95.4%
Either 1ary or 2ary predominant				
	MIP	6	83% CI: 46–97%	89% CI: 87–89.7%
	SOL	12	50% CI: 31–57%	99% CI: 97–99.8%
	All HG components	18	72% CI:53–86%	89% CI: 86–92%
Sole presence of pattern (≥5%)				
	MIP	26	65% CI: 55–65%	100% CI: 97–100%
	SOL	14	50% CI: 34–50%	100% CI: 98–100%
	All HG components	40	66% CI: 58–66%	100% CI: 97–100%

Clinical factors predictive of presence of MIP/SOL: heavy smoker (p=0.012, OR= 5.077) and larger consolidation size (p=0.04, OR=1.149). Influencing factors of concordance: MIP: no factors; in SOL two factors were associated with higher concordance: 1) ↑tumor size (p=0.04), and 2) ↑% of SOL component (p=0.03).

Discussion: They discuss that few studies have challenged the use of preop Bx for subtyping ADCs, including a couple of studies showing Sensitivity <20% for HG component determination. They also discuss about the advantages of cryoBx: 1) tissue samples can be obtained from lateral side of the tumor, 2) specimen size 3-7 times larger than forceps Bx, 3) structures are less crushed than in forceps Bx. They mention that sampling errors may affect SOL, but interpretation patterns may affect MIP. They discuss about IASLC paper and the MIP and SOL prevalence (22.6% and 12.2%) in that study, which suggest they are not negligible, and then may lead to changes in surgical management. ↑concordance than forceps Bx.

Take Home Point: CryoBx could be a feasible method for detection if HG patterns. However, the sensitivity is still relatively low for solid components, and specificity is not perfect for MIP (perhaps freezing artifact).

Potential risk for bias: Single center. Retrospective. Only two readers. Small number of cases with MIP and SOL components. Small number of cases with SOL predominant (only 14 cases). No cases with MIP predominant. Patient selection bias: not all patients with c-stage IA ADC underwent cryoBx.

Critical review: Did not use IASLC proposed grading system. Relevance? (no randomized prospective data showing clinical utility yet).

Transbronchial mediastinal cryobiopsy in the diagnosis of mediastinal lesions: a randomised trial Zhang et al, Eur Respir J 2021; 58: 2100055.

Introduction: The advent of EBUS-guided TBNA has revolutionized lung cancer staging and obviated the need for more invasive and dangerous mediastinoscopy procedures in most patients. However, while yield is typically very good for metastatic lung carcinoma, other diagnoses may be limited by the scant and/or fragmented nature of TBNA tissue. Diagnostic yield is also important for ancillary genetic and biomarker testing, which can often (but not always) be performed on EBUS tissue. Therefore, especially in select patients/scenarios, there may be a need for alternative procedures to increase tissue yield, and the authors propose studying the utility of cryoprobe biopsies (cryobx).

Methods: 197 patients with mediastinal lesions >1 cm (short axis on CT) were included, and EBUS FNA (4 passes) and EBUS-guided cryobx (3 specimens) were performed sequentially in random order with a 1:1 distribution. Cryobx were obtained through a small incision in the airway wall. Patients with cysts or abscesses were excluded, as were patients that needed an endobronchial biopsy. ROSE was not performed, and pathologists were blinded to the order of tissue procurement. "Diagnostic" biopsies were defined as those with a definitive pathological explanation for the targeted lesion (i.e. suspicious did not count as diagnostic).

Results: Average lesion size was just over 2 cm, and most commonly sampled stations were 4R and 7 (Table 1). 181 patients had a diagnostic biopsy from one or both procedures (diagnostic yield 93.3%). In 78.4% of patients (n=152), both cryobx and TBNA yielded an identical diagnosis. In 26 cases, the cryobx was diagnostic when the TBNA was not; diagnoses included 8 TB, 6 NSCLC, 6 lymphoma (all subtyped), 5 sarcoid and 1 seminoma. In an additional 4 lung cancer cases, cryobx provided more information about the tumor than the TBNA (sarcomatoid features, combined small cell, concurrent infection). In 3 cases, only the TBNA was diagnostic (all 3 NSCLC). Overall diagnostic yield was higher in cryobx compared to TBNA (91.8% vs 79.9%, respectively; p=0.001). Most of this difference seemed to be accounted for by the much higher diagnostic yield for cryobx when considering uncommon tumors (i.e. not NSCLC, 91.7% vs 25% by TBNA; p=0.001) and benign diseases (80.9% vs. 53.2% by TBNA; p=0.004), since there was no difference in diagnostic yield for metastatic carcinoma (94.1% vs 95.6%). Yield for molecular studies was 93.3% for cryobx vs. 73.5% for TBNA (p<0.001). There seemed to be no difference whether cryobx vs. TBNA was performed first, and diagnostic yield of cryobx did not depend on lesion size or station. Complications included 2 pneumothorax and 1 pneumomediastinum, and cryobx took about 2 minutes longer than TBNA. Cryobx samples averaged 10.7 mm^2.

Discussion: They discuss that some centers have implemented EBUS-guided mediastinal forceps biopsies when benign or unusual diagnoses are expected, with increased yield for diagnosis of lymphoma and sarcoid. They have shown that cryobx is possible and safe for mediastinal lesions as well, and produces three times more tissue than forceps. There were 3 cases where TBNA was diagnostic and cryobx was not, so perhaps sequential sampling or targeting patients may be appropriate.

Take Home Point: Cryobiopsy can be done for mediastinal lesions, may increase diagnostic yield compared to TBNA for non-lung cancer diagnoses like lymphoma and sarcoid, and may also provide more tissue for ancillary genetic and biomarker testing. I wish they would have provided a bit more background information about the patient's history/indication for EBUS/cryobx, but this is an interesting paper.

Comparison of Nuclear Grade, Necrosis, and Histologic Subtype Between Biopsy and Resection in Pleural Malignant Mesothelioma: An International Multi-Institutional Analysis
Schulte et al, Am J Clin Pathol 2021;156:989-999.

Introduction: While the prognostic significance of stratifying patients with pleura malignant mesothelioma (MM) into epithelioid, biphasic, and sarcomatoid groups has been well established and long practiced, additional histologic parameters to aid in determining prognosis (cytologic atypia, mitoses, necrosis) have recently been added to the WHO classification. It has generally been advocated to record all parameters based on biopsy specimens to aid in making treatment decisions for the patient. However, comparison of these histologic parameters between biopsies and subsequent resection specimens has not been rigorously evaluated.

Methods: This is a multi-institutional study (19 institutions) of 429 patients with pleural MM who had paired biopsy and resection (1995-2018, including peels, pleurectomy, and autopsies). Pathology review was performed by expert authors at each institution without central review. Histological features of the resection were considered the gold standard.

Results: There was a marked male predominance (83.4%), with mean age of 66.4 years (range 25-85). 196 (46%) patients received neoadjuvant therapy. Most (76%) of biopsies were VATS biopsies, with 15% core and 9% open biopsies. Biopsy and resection histological subtypes were concordant in 81% of cases (348 of 429). The number of epithelioid MM was overestimated by biopsy (table 1, 78.6% vs. 63.2%), while the number of biphasic cases was underestimated by biopsy (18.2% vs. 32.9%) and the number of sarcomatoid cases seemed the same (3.3% vs. 3.7%, table 2). Biopsy diagnosis of epithelioid MM was not a very specific indicator of resection diagnosis of epithelioid MM (specificity 54.8%), and biopsy was not particularly sensitive for biphasic morphology (sensitivity 49.6%, table 3). The ability to estimate % of epithelioid vs sarcomatoid components in biphasic MM had only fair agreement between biopsy and resection (kappa=0.27). Most cases that showed epithelioid morphology at biopsy (96%) had at least 50% epithelioid morphology at resection. Overall, there was moderate agreement between biopsy and resection (kappa=0.54). There was agreement in nuclear grade (grade 1, 2 or 3) in 75% of cases (moderate agreement, kappa=0.59). 54 cases had a change in nuclear grade at resection; 93% were upgraded one level. Necrosis was concordant in 81% of cases (moderate agreement, kappa=0.53); necrosis on biopsy was specific (97.1%) but not sensitive (50.7%) for identifying necrosis at resection. Low nuclear grade seemed more common in resection cases with neoadjuvant therapy (42% vs. 20%, p=.0028), but necrosis was not different.

Discussion: About 20% of MM classified as epithelioid on biopsy will be reclassified as biphasic at resection. The opposite does not seem to be true; those classified as sarcomatoid on biopsy remained classified as sarcomatoid at resection. There is a tendency to upgrade on resection compared to biopsy, especially in grade 1 tumors (34% upgraded). This upgrade rate may not apply to cases with neoadjuvant therapy, that seem to have a higher rate of nuclear grade 1 tumors at resection.

Take Home Point: Most pathologic features show moderate agreement between biopsy and resection specimens. About 20% of epithelioid biopsies will be changed to biphasic at resection, but the vast majority will be >50% epithelioid. Identification of sarcomatoid MM on biopsy is highly predictive of sarcomatoid morphology at resection. As you might expect, at resection there is a greater sensitivity for finding necrosis and tendency to up-grade nuclear grade. The most limited capability of biopsies seems to be predicting percentages of sarcomatoid and epithelioid components in biphasic MM.

Articles for Notation

Neoplastic

CD34-negative Solitary Fibrous Tumor : A Clinicopathologic Study of 25 Cases and Comparison with their CD34-positive Counterparts

Dermawan et al, Am J Surg Pathol 2021;45:1616–1625

Summary: CD34 is the historical marker of SFT; while it lacks specificity, it is generally a quite sensitive marker of SFT. However, the sensitive and specific maker STAT6 has largely supplanted CD34 as the maker of choice for SFT, and has led to discovery of CD34-negative examples. The authors describe 25 cases of CD34-negative SFT, which represents 10% of all SFT cases observed (2013-2020; but there is likely referral bias here). All were confirmed by STAT6 expression and/or presence of *STAT6* fusion. Only 4 of the CD34-negative cases were intrathoracic, which is in contrast to the CD34- positive tumors, where the thorax was the most common site. CD34-negative tumors were more likely to occur in the head and neck, and were much more likely to have metastasis at presentation (28% vs. 1% of CD34-positive tumors). They are also more likely to show high grade cytological features including hypercellularity, round cell morphology, and pleomorphism. They were less likely to show classical HPC-like vessels (seen in about half). Interestingly, risk stratification was not different. The authors noted several morphological features unique to CD34-negative tumors, that included alternating hypercellular and hypocellular areas, myxoid areas with curvilinear vessels, pulmonary edema- like microcystic change, and prominent amianthoid collagen fibers.

Take Home Point: CD34-negative SFTs are uncommon in the thorax but can occur. They tend to have high grade cytological features and some other unique histological features. STAT6 is therefore warranted to exclude SFT even if morphological features are not prefect.

Knowledge and Practice Patterns Among Pulmonologists for Molecular Biomarker Testing in Advanced Non-small Cell Lung Cancer

Fox et al, CHEST 2021; 160(6):2293-2303.

Summary: This is a random survey of 453 pulmonologists of varying practice environments and lung cancer volumes. Increasing knowledge of EBUS-TBNA technical recommendations (number of passes) was associated with academic setting, interventional training and higher volume. Academic pulmonologists were more likely to utilize EBUS-TBNA than those in community practice. Molecular biomarker testing rates were also higher in the academic setting, in pulmonologists that had interventional training, and in institutions that had standard recommendations regarding testing.

Take home point: Academic practices are generally better about ordering biomarker testing in lung cancer, especially if there is a coordinated institutional effort to perform this testing with standard guidelines.

Identification of Molecular Alterations Challenging Initial Pathologic Classification in Cases of Clinician-Initiated Next-Generation Sequencing Testing

Cho et al, Am J Clin Pathol 2021;156:1007-1018.

Summary: This is a report of 9 cases where next generation sequencing changed the pathological diagnosis. This included **mucinous lung adenocarcinoma to metastatic prostate cancer, lung adenocarcinoma to metastatic GE junction adenocarcinoma, lung squamous cell carcinoma to INI1-deficient sinonasal carcinoma, NSCLC to epithelioid sarcoma,** mucoepidermoid carcinoma to hyalinizing clear cell carcinoma, metastatic carcinoma to the brain to glioblastoma, duodenal adenocarcinoma to invasive oncocytic carcinoma, epithelioid MPNST to melanoma, and duodenal adenocarcinoma to metastatic cervical cancer (HPV-mediated).

Take home point: Molecular genetic testing can occasionally provide new information that leading to a change in pathological diagnosis. Nearly half of the cases in this report involved a diagnosis of lung cancer, most commonly misinterpretation of a metastasis as a primary lung cancer. For this reason, it is important for surgical pathologists to always be looped back in on important molecular testing results.

***YAP1–TFE3* gene fusion variant in clear cell stromal tumor of lung: report of two cases in support of a distinct entity**

Dermawan et al, Histopathol 2021 79, 940–946.

Summary: This is an interesting report of two cases of primary lung clear cell tumors with *YAP1–TFE3* fusion and non-endothelial phenotype. These have been reported before, but I have not read about them prior to this. Both patients were men (aged 35 and 77) with solitary lung masses measuring 3.9 and 7.5 cm. These were low-grade appearing spindled tumors with clear to pale to eosinophilic cytoplasm, without high mitotic activity, pleomorphism, or necrosis. They are often endobronchial and circumscribed. They somewhat resemble PEC-omas, and had some stromal chronic inflammation. They were TFE3 positive (this in not a specific marker), but all other markers are negative. The exact fusion characteristics are different from EHE with the same fusion (EHE breakpoint occurs in *YAP1* exon 1 and *TFE3* exons 4 or 6, the fusion breakpoints of these tumors were located in *YAP1* exon 4 and *TFE3* exon 7). Limited follow-up showed very good prognosis.

Take home point: New rare low-grade endobronchial entity with specific fusion and good prognosis-original name may be a bit misleading, as these authors state that cytoplasm is often not very clear. Worth taking a look at the pictures.

Histopathological and molecular profiling of lung adenocarcinoma skin metastases reveals specific features

Fisher et al, Histopathol 2021; 79: 1051–1060 .

Summary: Retrospective study of 42 cases of lung adenocarcinoma metastatic to the skin. The skin metastasis was the initial presenting symptom of lung cancer in 62%. 60% were solid predominant (compared to 46% solid predominant for non-cutaneous metastases). One third showed *KRAS* mutations, while only one had an *EGFR* mutation. Most (27 of 42) cases had a negative PD-L1 (64%) . Interestingly, TTF-1 was more often negative in skin cancer mets (71%) comparted to mets at other sites.

Take home point: Skin mets from lung adenocarcinoma are often solid-predominant, TTF-1 negative, and PD-L1 negative. They often have *KRAS* mutations but *EGFR* mutations are uncommon. The skin met is often the initial presentation of the patient's lung cancer.

Mesenchymal/non-epithelial mimickers of neuroendocrine neoplasms with a focus on fusion gene-associated and SWI/SNF-deficient tumors

Kasajima et al, Virchows Archiv 2021; 479:1209–1219.

Summary: This is a report of 31 mesenchymal (i.e. non-epithelial) neoplasms that expressed the neuroendocrine markers synaptophysin and/or chromogranin. All but one expressed patchy synapto, but one-third co-expressed chromogranin. 42% of these neoplasms were *EWSR1*-rearranged sarcomas, including Ewing sarcoma (6), clear cell sarcoma (5), and DSRCT (1); one tumor showed the related *FUS-CREM* fusion gene. Another 23% of cases were tumors with abnormalities in the SWI/SNF pathway, with deficiency of either SMARCB1 (INI1) or SMARCA4 (BRG1). Other neoplasms included synovial sarcoma, melanoma, ASPS, SFT and chordoma.

Take home point: As I think we are all aware, aberrant neuroendocrine maker expression happens in many types of epithelial and non-epithelial tumors, importantly including *EWSR1*-rearranged sarcomas and tumors with SWI/SNF pathway mutations.

Intimal sarcomas and undifferentiated cardiac sarcomas carry mutually exclusive M*DM2, MDM4*, and *CDK6* amplifications and share a common DNA methylation signature

Koelsche et al, Mod Pathol 2021; 34:2122–2129.

Summary: Report of 35 cases including 25 pulmonary artery intimal sarcomas, one intimal sarcoma of the renal artery, and 9 undifferentiated pleomorphic sarcomas (UPS) of the left atrium. All cases had a morphology similar to UPS at other soft tissue sites. All cases demonstrated very complex karyotypes. Most cases (83%) showed mutually exclusive amplifications of *MDM2* (25), *MDM4* (2), and *CDK6* (2). There was frequent co-amplification of *PDGFRA* (21), *CDK4* (15), *TERT* (11), *HDAC9* (9), and *CCND1* (4). *CDKN2A/B* was also co-deleted in 10 cases. DNA methylation showed a unique profile, which was similar among all of the PA and cardiac tumors.

Take home point: Genetic similarities between intimal sarcoma and UPS of the left atrium supports that they are the same entity.

Comparison of solid tissue sequencing and liquid biopsy accuracy in identification of clinically relevant gene mutations and rearrangements in lung adenocarcinomas

Lin et al, Mod Pathol 2021; 34:2168–2174.

Summary: This is a head-to-head comparison study of tissue-based NGS molecular testing and plasma-based NGS (i.e. liquid biopsy), which included 100 consecutive patients with lung adenocarcinoma. 83% had stage IV disease, and clinically relevant mutations were found in 78 cases by one or both methods. Tissue- based NGS testing was significantly more sensitive than liquid biopsy in the identification of clinically relevant mutations (74 vs. 41 cases, sensitivity for targetable mutaions 94.8% vs. 52.7%). Liquid biopsy was especially inaccurate in patients with non-metastatic disuse. There were some examples of actionable targets (*EGFR, ALK, ROS1, BRAF, MET, NTRK1*) which were missed by liquid biopsy. There were a few examples of relevant mutations found by liquid biopsy only (3 *EGFR* and 1 *KRAS*). But the authors note that there were more numerous examples of mutations that actually raise questions about the specificity of the liquid biopsy assay- 6 *JAK2* mutations were found in patients without underlying myeloproliferative neoplasms, and one tumor was reported to have both *KRAS* and *EGFR* mutations in plasma (which are generally considered mutually exclusive), and only the *EGFR* mutation was found on tissue-based testing (implying a false-positive *KRAS* test).

Take home point: Tissue NGS has significantly better sensitivity compared to liquid biopsy, especially in the non-metastatic setting. Therefore, tissue testing should be the test of choice whenever possible. While liquid biopsies may be helpful in some cases, negative results should be viewed with healthy skepticism and repeated on tissue if possible.

***KEAP1* and *TP53* Frame Genomic, Evolutionary, and Immunologic Subtypes of Lung Adenocarcinoma with Different Sensitivity to Immunotherapy**

Scalera et al, J Thorac Oncol 2021;16(12): 2065–2077.

Summary: This is an in-depth molecular study of *KEAP1* single mutant lung adenocarcinoma compared to *KEAP1/TP53* double mutants ,and response of these tumors to immune checkpoint inhibitors. There is a lot of data in this paper and much of it is pretty detailed. In a nutshell, *KEAP1* single mutant tumors had the worst response to immunotherapy with the shortest survival, followed by *KEAP1/TP53* double mutants (which was very similar to the entire *TP53* mutant cohort), and tumors with wild type *KEAP1* and *TP53* had the best response/longest survival.

Take home point: Underlying tumor genetic landscape including evaluation of *KEAP1* and *TP53* can help predict response to immune checkpoint inhibitors.

Differential Immune-Related Microenvironment Determines Programmed Cell Death Protein-1/ Programmed Death-Ligand 1 Blockade Efficacy in Patients with Advanced NSCLC

Shirasawa et al, J Thorac Oncol 2021;16(12): 2078–2090.

Summary: 228 patients were divided into 4 tissue microenvironment groups based on PD-L1 TPS and number of CD8-positive tumor infiltrating lymphocytes (TILs): type 1 was PD-L1 high (TPS>50) and TIL high (n=73), type 2 was PD-L1 low (TPS<50) and TIL low (n=70), type 3 was PD-L1 high and TIL low (n=37), and type 4 was PD-L1 low and TIL high (n=48). Response rate to checkpoint inhibitor therapy and progression free survival were different between groups (type 1>type 4>type 3>type 2). When comparing the PD-L1 high tumors broken down by low vs. high TILs, those with high TILS (type 1) clearly had more favorable response.

Take Home Point: High rates of CD8-positive TILs is a favorable indicator of response to checkpoint inhibitor therapy, even in the PD-L1 low setting.

Non-Neoplastic

Follicular Helper–like T Cells in the Lung Highlight a Novel Role of B Cells in Sarcoidosis

Bauer et al, Am J Respir Crit Care Med 2021; 204 (12):1403–1417.

Summary: Previous studies have shown that pulmonary sarcoidosis is driven by a population of CD4+ T-helper type 1 cells with produce interferon gamma and drive granuloma formation. In a prior mouse model of inflamed lung, this group had discovered a population of cells resembling follicular T-helper cells that drive local B-cells to differentiate into follicular center-like B-cells and plasmablasts, which produce large amounts of antibodies into the inflamed tissue. The cells defer from true follicular T helper cells in that they lack expression of BCL 6 andCXCR4, but do express follicular T-cell markers CD40L and IL-21. They studied BAL fluid from 18 sarcoid patients to see if they could find these cells using flow, and they found them; they were also present in peripheral blood. They did transcriptome analysis and confirmed these cells were follicular T-cell like, and resident to the lung tissue. These T-cells strongly activated B-cells into plasmablasts *in vitro*. In sarcoid lung tissue, they found IgA positive plasmablasts in peribronchial tissue associated with lots of B-cells and T-cells, without true germinal centers.

Take Home Point: Sarcoid may not be driven only by CD4-positive T-helper type 1 cells, but may also have an important pathogenic follicular T cell-like and B cell mediated antibody response. This could have therapeutic implications, and could potentially be used to follow disease severity/activity.

Chronic respiratory diseases are predictors of severe outcome in COVID-19 hospitalized patients: a nationwide study

Beltramo et al, Eur Respir J 2021;58(6):2004474.

Summary: Population study of 89,530 COVID-19 patients compared to 45,819 influenza patients. COVID patients were actually less likely to have an underlying chronic respiratory disease (16%) compared to patients with influenza (20%). The chronic lung diseases that seemed to be most overrepresented in COVID patients included lung cancer, ILD, emphysema and sleep apnea. In contrast, some other chronic lung diseases were underrepresented in COVID patients, including asthma, CF, COPD, and pulmonary

hypertension. Compared to influenza patients, COVID patients were more likely to suffer a PE and get ventilator-associated pneumonia. Patients with COVID and an underlying chronic lung disease had a higher need for ICU care and higher mortality compared to those without underling chronic lung disease, and compared to those with influenza.

Take Home Point: Certain underlying lung disease seem to particularly increase risk of severe COVID and COVID-related death, including lung cancer, emphysema, ILD and sleep apnea.

Interstitial lung disease increases susceptibility to and severity of COVID-19

Lee et al, Eur Respir J 2021; 58: 2004125.

Summary: This is a large case: control study of patients with COVID-19 (n=8070) in Korea, focusing on those with ILD. Included ILDs included IPF, collagen vascular disease associated ILD (most were not taking immunosuppressive agents), HP, sarcoid, and "others." The control cohort was 1:15 age-, sex- and residential area-matched (n=121,050). The rate of COVID was higher in patients with ILD (0.8%) vs controls (0.4%, p<.001) (higher infection rate vs. higher symptomatic rate? Not sure). The odds ratio for ILD in the COVID cohort was 2.02. Importantly, patient with ILD were much more likely to have severe COVID compared to those without ILD (47.8% vs. 12.6%) and much more likely to die of COVID (13.4% vs. 2.8%), with an odds ratio of severe COVID of 2.23.

Take home point: Not surprisingly, underlying ILD leads to an increased risk of severe COVID and death from COVID.

Bronchoalveolar lavage cytokine-based risk stratification of minimal acute rejection in clinically stable lung transplant recipients
Levy et al, J Heart Lung Transplant 2021;40:1540–1549.

Summary: While pretty much all lung transplant clinicians agree that A2 rejection and higher should be treated by increasing immunosuppression, treatment vs. observation in A1 rejection is much more controversial if the patient is clinically stable. The authors do not treat clinically stable patients (<10% drop in FEV1) with A1 rejection at their facility, and they have similar outcomes to patients with A0 rejection, but their rate of chronic allograft dysfunction (CLAD) and death are still high in their population. Therefore, they wanted to try and identify patients at high risk that might benefit from aggressive treatment of A1 rejection. They did this by looking at BAL specimens at the time of first incidence of A1 rejection in 75 bilateral lung transplant recipients (2010-16). They assessed the BAL fluid for 21 cytokine proteins that have been previously associated with CLAD, fibrosis, or acute cellular rejection, using a multiplex bead assay. 39% of patients went on to develop CLAD, and 24% experienced death or re-transplant. Several proteins including MCP1/CCL2, S100A8, IL10, TNF-receptor 1, and pentraxin 3 were associated with both CLAD development and death ($p < 0.05$). BAL levels of pentraxin 3 remained significant after multi variable analysis.
Take Home Point: The authors described a protein signature in BAL, specifically elevated levels of pentraxin 3, that may be helpful to identify which clinically stable patients with A1 rejection are at increased risk of CLAD or death, and therefore may benefit from increased immunosuppression.

Peripheral blood leucocyte telomere length is associated with progression of interstitial lung disease in systemic sclerosis

Liu et al, Thorax 2021;76:1186–1192.

Summary: Retrospective study of 213 patients with systemic sclerosis (SS)- findings were validated in a separate set of 61 SS ILD patients. In patients with SS, peripheral blood telomeres were shorted in patients with ILD compared to those without ILD. Conversely, shorter telomeres also predicted the presence of ILD (odds ratio of 2). Telomeres were shorter in patients that lacked classic autoantibodies seen in SS, compared to those with antibodies. Shorter telomeres were also associated with decreasing lung function.

Take home point: Shorter telomeres seem to be associated with presence of ILD in SS patients, as well as risk of decreasing lung function over time.

Pulmonary fibrosis 4 months after COVID-19 is associated with severity of illness and blood leucocyte telomere length

McGroder et al, Thorax 2021;76:1242–124.

Summary: The authors assessed CT findings in 76 COVID patients requiring hospitalization, 4 months after discharge. 42% of patients required mechanical ventilation. The most common CT abnormality was GGOs (43%), followed by reticulation (39%) and traction bronchiectasis (28%). They found fibrosis was much more common in patients requiring mechanical ventilation (72% vs. 20%). Other independent risk factors for fibrosis included duration of mechanical ventilation, severity of COVID and telomere length. Patients with fibrosis complained of more cough but not dyspnea, and fibrosis was also associated with frailty and decreased lung function.

Take home point: Risk factors for post-COVID fibrosis on CT include mechanical ventilation, duration of mechanical ventilation, severity of COVID, and telomere length.

Fibroblasts positive for meflin have anti-fibrotic property in pulmonary fibrosis

Nakahara et al, Eur Respir J. 2021 Dec 23;58(6):2003397.

Summary: This is a basic science study of fibroblasts that express meflin. While the methods of this study are a bit over my head, the upshot seems to be that the that meflin-positive fibroblasts are increased in IPF lungs compared to controls, and that it is often co-expressed with pro-fibrotic genes. They were most prevalent in fibroblast foci and not in the dense mature fibrosis. They used a mouse bleomycin mouse model and demonstrated that meflin protein expression increased after bleomycin administration and was localized to fibrotic lesions. They applied the bleomycin to meflin-deficient mice, who experienced more severe fibrosis than the meflin wild type mice. They demonstrate evidence to indicate that meflin has a protective anti-fibrotic role in mice exposed to bleomycin. Lack of meflin seems to lead to aberrant fibrogenesis mediated by TGF-beta and cellular senescence.

Take home point: The protein meflin seems to have anti-fibrotic properties that could hypothetically be exploited to slow progression of IPF.

Dynamics of the Upper Respiratory Tract Microbiota and Its Association with Mortality in COVID-19

Ren et al, Am J Respir Crit Care Med 2021; 204(12):1379–1390.

Summary: They performed serial megatranscriptome testing to monitor the oropharyngeal microbiome in 192 patients with COVID, of which 39 had fatal disease, compared to 95 healthy controls. The microbiome in COVID patients differed from controls, and was associated with increased serum levels of inflammatory cytokines. Interestingly, the patients who died had a distinct microbiome both at the time of admission and just prior to death. *Streptococcus*-dominated microbiome was more prevalent in patients who recovered, and this microbiome was very stable and resistant to pathogenic organisms. Patients who died had more deviation of their microbiome from normal and experienced secondary infections. *Veillonella, Actinomyces*, and *Rothia* were more prevalent in patients with COVID-19, whereas *Streptococcus* and *Capnocytophaga* were more prevalent in controls. *Candida* and *Enterococcus* were more enriched in patients who died, while *S. parasanguinis* was associated with better outcome when it was the dominant organism at admission with COVID.

Take home point: Interesting evidence that the microbiome plays an important role in COVID-19 infections and may be able to be used to predict prognosis even at the time of admission.

Transcriptomics of bronchoalveolar lavage cells identifies new molecular endotypes of sarcoidosis

Vukmirovic et al, Eur Respir J 2021; 58: 2002950.

Summary: RNA sequencing study of BAL fluid from 215 patients with pulmonary sarcoidosis, validated on an independent cohort of 50 sarcoid patients. They found that T-helper type 1 and type 17 pathways were associated with hilar lymphadenopathy, TGFB1 and MTOR abnormalities were associated with parenchymal involvement, and IL-7 and IL-2 were associated with airway involvement. Unsupervised gene clustering led to four phenotypic groups: hilar lymphadenopathy associated with increased acute T-cell response; extraocular organ involvement associated with PI3K activation; chronic and multiorgan disease associated with increased immune response pathways; and multiorgan involvement associatd with increased IL-1 and IL-18 response.

Take Home Point: Specific gene profiles can be detected in BAL which are associated with disease severity and pattern of systemic involvement in sarcoid.

Case Reports, Reviews and Letters (Listed for notation only; Full PDFs in Dropbox folder)

Molecular testing in stage I–III non-small cell lung cancer: Approaches and challenges. Aggarwal et al, Lung Cancer 2021; 162: 42–53.

Localized ALK-positive histiocytosis in a Chinese woman: report of a case in the lung with a novel EML4-ALK rearrangement. Bai et al, Virchows Archiv 2021; 479:1079–1083.

Pulmonary Arterial Hypertension. Hassoun, PM. N Engl J Med 2021;385:2361-76.

A primary thymic adenocarcinoma with two components that traced distinct evolutionary trajectories Ishida et al, Pathol International 2021;71:849–855.

Immune checkpoint inhibitor therapy for malignant pleural mesothelioma. Nowak et al, Lung Cancer 2021;162:162–168.

A contemporary practical approach to the multidisciplinary management of unclassifiable interstitial lung disease. Ryerson et al, Eur Respir J 2021; 58: 2100276

POINT: Should Surgical Lung Biopsy Still Be Performed for Interstitial Lung Disease Evaluation? Yes. Ryerson et al, Chest 2021; 160(6):2007-2011.

COUNTERPOINT: Should Surgical Lung Biopsy Still Be Performed for Interstitial Lung Disease Evaluation? No. Tomassetti et al, Chest 2021;160(6): 2011-2014.

An Alert to Possible False Positives with a Commercial Assay for *MET* Exon 14 Skipping. Teishikata et al, J Thor Oncol 2021; 16(12): 2133–2138.

Endobronchial Clear-Cell Sugar Tumor. Xiao et al, Am J Resp Critical Care Med 2021; 204 (12):1476-77.