Pulmonary Pathology Journal Club, November 2020

Jennifer Boland, MD; Associate Professor of Laboratory Medicine and Pathology
Lacey Schrader, MD; Pulmonary Pathology Fellow
Mayo Clinic, Rochester, MN

Table of Contents

Articles for Discussion

Articles for Notation

Non-neoplastic

Neoplastic

Multicenter, Double-Blinded, Randomized Study of 2045 Cases. Archives Lab Med Pathol 2020;144:1245-1253.

Page 12 Durm GA, SK Jabbour, SK Althouse, et al. A Phase 2 Trial of Consolidation Pembrolizumab Following Concurrent Chemoradiation for Patients With Unresectable Stage III Non–Small Cell Lung Cancer: Hoosier Cancer Research Network LUN 14-179. Cancer 2020;126:4353-4361.

Page 12 Herbst RS, G Giaccone, F de Marinis, et al. Atezolizumab for First-Line Treatment of PD-L1–Selected Patients with NSCLC. New Engl J Med 2020;383(14):1328-1339.

Page 13 Hernandez A, TC Brandler, F Chen, et al. Scoring of Programmed Death-Ligand 1 Immunohistochemistry on Cytology Cell Block Specimens in Non–Small Cell Lung Carcinoma. Am J Clin Pathol 2020; 154;517-24.

Page 13 Kimura T, H Nakamura, A Omura, et al. Novel imprint cytological classification is correlated with tumor spread through air spaces in lung adenocarcinoma. Lung Cancer 2020;148:62-68.

Page 14 Krigsfeld GS, EA Prince, J Pratt, et al. Analysis of real-world PD-L1 IHC 28-8 and 22C3 pharmDx assay utilisation, turnaround times and analytical concordance across multiple tumour types. J Clin Pathol 2020;73:656-664.

Page 14 Marin I, E Ofek, J Bar, et al. MiR-21, EGFR and PTEN in non-small cell lung cancer: an in situ hybridisation and immunohistochemistry study. J Clin Pathol 2020;73:636-641.

Page 15 Nakashima Y, K Inamura, H Ninomiya, et al. Frequent expression of conventional endothelial markers in pleural mesothelioma: usefulness of claudin-5 as well as combined traditional markers to distinguish mesothelioma from angiosarcoma. Lung Cancer 2020;148:20-27.

Page 15 Negrao MV, VM Raymond, RB Lanman, et al. Molecular Landscape of BRAF-Mutant NSCLC Reveals an Association Between Clonality and Driver Mutations and Identifies Targetable Non-V600 Driver Mutations . J Thorac Oncol 2020;15(10):1611-23.

Page 16 Parra ER, M Jiang, J Machado-Rugolo, et al. Variants in Epithelial-Mesenchymal Transition and Immune Checkpoint Genes Are Associated With Immune Cell Profiles and Predict Survival in Non–Small Cell Lung Cancer. Arch Lab Med Pathol 2020;144:1234-1244.

Page 16 Ray MA, MP Smeltzer, NR Faris, et al. Survival After Mediastinal Node Dissection, Systematic Sampling, or Neither for Early Stage NSCLC. J Thorac Oncol 2020; 15(10):1670-81.

Page 16 Schultz KAP, A Nelson, AK Harris, et al. Pleuropulmonary blastoma-like peritoneal sarcoma: a newly described malignancy associated with biallelic DICER1 pathogenic variation. Mod Pathol 2020;33:1922-1929.

Page 17 Thomas A, I Mian, C Tlemsani, et al. Clinical and Genomic Characteristics of Small Cell Lung Cancer in Never Smokers- Results From a Retrospective Multicenter Cohort Study. Chest 2020;158(4):1723-1733.

Page 17 Yang H, Z Zhou, L Lin, et al. Characterization of *MET* exon 14 alteration and association with clinical outcomes of crizotinib in Chinese lung cancers. Lung Cancer 2020;148:113-121.

Page 18 Yoon J, R Nayyar, G Quest, et al. PD-L1 lineage-specific quantification in malignant pleural effusions of lung adenocarcinoma by flow cytometry. Lung Cancer 2020;148:55-61.

Page 18 Zoborawski M, AC Vargas, J Pulvers, et al. When used together SS18–SSX fusion-specific and SSX C-terminus immunohistochemistry are highly specific and sensitive for the diagnosis of synovial sarcoma and can replace FISH or molecular testing in most cases. Histopathol 2020;77:588-600.

Page 18 Zaffiri L, A Long, ML Neely, et al. Incidence and outcome of post-transplant lymphoproliferative disorders in lung transplant patients: Analysis of ISHLT Registry. J Heart Lung Transplant 2020; 39:1089-99.

Page 19 Zhang EW, I Dagogo-Jack, A Kuo, et al. Association Between Circulating Tumor DNA Burden and Disease Burden in Patients With ALK-Positive Lung Cancer. Cancer 2020;126:4473-4484

Case Reports, Letters, Editorials, and Reviews (listed on Table of Contents Only, PDFs available in Dropbox)

Coleman N, D Woolf, L Welsh, et al. EGFR Exon 20 Insertion (A763_Y764insFQEA) Mutant NSCLC Is Not Identified by Roche Cobas Version 2 Tissue Testing but Has Durable Intracranial and Extracranial Response to Osimertinib. J Thorac Oncol 2020;15(10):e162-165.

Cornelissen R, HJ Dubbink, JH von der Thusen, et al. ALK in Mesothelioma: To FISH or Not to FISH? J Thorac Oncol 2020;15(10):e168-169.

Jonas AM, R Raj. Vaping-Related Acute Parenchymal Lung Injury: A Systematic Review. Chest 2020;158(4):1555-1565.

Kunimasa K, Y HIrotsu, Y Miyashita, et al. Multiregional sequence revealed SMARCA4 R1192C mutant clones acquired EGFR C797S mutation in the metastatic site of an EGFR-mutated NSCLC patient. Lung Cancer 2020;148:28-32.

Kuriyama K, M Tanagawa. CT Diagnosis of Lung Adenocarcinoma: Radiologic- Pathologic Correlation and Growth Rate. Radiology 2020;297:199-200.

Li M, Z Xu, C Zhan, et al. A Few Clouds Over the Eighth Edition T Categorization System. J Thorac Oncol 2020; 15(10):e159-160.

Liu S, G Giang, EH Wang, et al. Are there preinvasive lesions of pulmonary large cell carcinoma? Lung Cancer 2020;148:166-69.

Matsuda K. The Isoform Matters in NUT Carcinoma: A Diagnostic Pitfall of p40 Immunohistochemistry. J Thorac Oncol 2020; 15(10):e176-177.

McCarthy C, MP Keane, A Fabre. Lipid-Laden Macrophages Are Not Diagnostic of Pulmonary Alveolar Proteinosis Syndrome and Can Indicate Lung Injury. Am J Respir Crit Care Med 2020; 202(8):1197-98.

Roy-Chowdhuri, S. A New Guideline from the College of American Pathologists to Improve the Adequacy of Thoracic Small Specimens for Ancillary Studies. Cancer Cytopathol 2020;690-92.

Severson DT, A De Rienzo, R Bueno. Mesothelioma in the age of “Omics”: Before and after The Cancer Genome Atlas. J Thorac Cardiovasc Surg 2020;160(4):1078-1083.

Sharma P, A Zeki. Does Vaping Increase Susceptibility to COVID-19? Am J Resp Crit Care Med 2020;202(7):1055-56.

Sinozaki-Ushiku A, S Kohsaka, H Kage, et al. Genomic profiling of multiple primary cancers including synchronous lung adenocarcinoma and bilateral malignant mesotheliomas: Identification of a novel BAP1 germline variant. Pathol Int 2020:70:775-780.

Singh Sj, EK Bergsland, CM Card, et al. Commonwealth Neuroendocrine Tumour Research Collaboration and the North American Neuroendocrine Tumor Society Guidelines for the Diagnosis and Management of Patients With Lung Neuroendocrine Tumors: An International Collaborative Endorsement and Update of the 2015 European Neuroendocrine Tumor Society Expert Consensus Guidelines. J Thorac Oncol 2020;15(10):1577-98.

Stock CJ, C Conti, A Montero-Fernandez, et al. Interaction between the promoter *MUC5B* polymorphism and mucin expression: is there a difference according to ILD subtype? Thorax 2020;75:901-903.

Swaminathan AC, JL Todd. A Fresh Look at Idiopathic Pulmonary Fibrosis Biomarkers in the Antifibrotic Era. Chest 2020;158(4):1321-22.

Weatherald J, L Bondeelle, MC Chaumais, et al. Pulmonary Complications of Bcr-Abl Tyrosine Kinase Inhibitors. Eur Resp J 2020;56(4):2000279.

Yang H, SRR Hall, F Yao. The Value of PD-L1 Expression in Metastatic Lymph Nodes of Advanced Non-Small Cell Lung Cancer. Chest 158(4):1786-7.

Yoshimura M, M Hamasaki, Y Kinoshita, et al. Utility of highly expressed EZH2 in pleural effusion cytology for the diagnosis of mesothelioma. Pathol Int 2020;70:831-33.

US Preventive Services Task Force Issues New Draft Recommendation Statement Regarding Lung Cancer Screening. Cancer 2020;4269.

Articles for Discussion

Further support for the identification of a new tumor category in the classification of lung neuroendocrine neoplasms.

M. Rubino et al, Lung Cancer. 2020;148:149-158

Purpose: The category of neuroendocrine tumor grade 3 (NET G3) was first introduced in 2017 World Health Organization (WHO 2017) pancreatic classification of neuroendocrine neoplasms. NET G3 are well differentiated NETs with a mitotic count >10/2 mm^2 and/or a Ki-67 index >20%. This study investigates if there are cases of lung NETs with carcinoid morphology that correspond to the NET G3 category described in other tissue sites.

Methods: Retrospective review of a total of 630 cases of NET from 2000 to 2017 at two NET referral centers. Cases were excluded if there was a diagnosis of poorly differentiated lung neuroendocrine carcinoma, unavailability of tissue for microscopic review, or there was no available clinical/follow-up data. 514 cases qualified using the above criteria, which were examined for frequency of highly proliferative lung carcinoids (HPLC) using the NET G3 criteria, clinical behavior, pathological features and response to therapy.

Results:

- 30/514 (6%) HPLC identified based on Ki-67 index >20% and/or mitotic count >10/ 2 mm^2
- 30/302 (10%) among tumors with available Ki-67 index were classified as HPLC
- 12/18 HPLC cases with local disease later developed metachronous distant metastases
- Median progression free survival in months of HPLC under platinum/Etoposide 5.1, Everolimus 12.1 and PRRT 14.2

HPLC	**Localized**	**Metastatic**
Case #	18	12
Ki67	23% (15-65)	25% (8-60)
Mitotic count	4.5/2 mm^2 (1-11)	6/2 mm^2 (3-15)

	Conventional Lung Carcinoid		**HPLC**
Ki-67	≤5%	6-20%	>20%
Cases #	157	115	30
Recurrence rate	9% (33/352)		66% (12/18)
Median RFS in months	288 (141-NR)	NR (148-NR)	24 (10-NR)
median OS in months for metastatic dz	203 (83-NR)	101 (79-NR)	53 (39-NR)

Discussion:
The WHO 2015 classification of lung carcinoid tumors did not recognize the HPLC category. This study highlighted a group of well-differentiated lung NETs with proliferative rates higher than

currently accepted for TC and AC. It is speculated that the HPLC defined in this article is the lungs equivalent to the GEP NET G3. Additionally, the clinical behavior and profile of HPLC mimics those of atypical carcinoid versus neuroendocrine carcinoma. HPLC is less responsive to platinum/Etoposide therapy (1st line therapy for NEC) and more response to Everolimus and PRRT.

Comment: Will the next WHO pulmonary classification of neuroendocrine neoplasms include a NET G3 like category and how should this category be defined?

Osimertinib in Resected EGFR-Mutated Non-Small-Cell Lung Cancer.
Yi-Long Wu et al. N ENGL J. MED. 2020 October;383(18):1711-1723.

Purpose: ADOURA trail—is there benefit in adjuvant Osimertinib therapy for EGFR mutated non-small cell lung cancer in patients who have undergone complete resection.

Methods: Phase 3, double-blind, placebo-controlled, randomized international trail. A total of 682 patients with stage IB-IIIA completely resected NSCLC harboring an EGFR activating mutation (Ex19del or L858R) were randomized on a 1:1 basis to receive 80 mg/day Osimertinib or placebo for 3 years. Adjuvant chemotherapy was allowed. The primary endpoint was disease-free survival (DFS) in stage II to IIIA disease. The secondary endpoints were overall survival (OS) and safety.

Results:
- At 24 months 90% of patients with stage II to IIIA disease in the osimertinib group and 44% of those in the placebo group were alive and disease-free.
- The hazard ratio for disease recurrence or death for stage II to IIIA only and when including stage IB was 0.17 and 0.20 respectively.
- DFS benefit with osimertinib was also observed in subgroups across the entire population, regardless of race, stage of disease or type of EGFR mutation.
- Overall survival (OS) data was immature.

Discussion:
There was an 80% reduction in the risk of disease recurrence or death, indicating significant improvement in DFS in patients with stage IB/II/IIIA EGFRm NSCLC after complete tumor resection and adjuvant chemotherapy, when indicated. The DFS was met with such a strong degree that upon recommendation by the Independent Data Monitoring Committee, the study was unblinded at a trail level (two years earlier than planned).

Comment: ASCO 2020 –presented at the plenary session and got a lot of discussion about the potential clinical impact. The trail was positive for the primary end point. However, did unblinding the trail early compromise OS data? Are EGFR TKIs in this setting improving cure rates or merely delaying recurrence without improving OS? Is this going to lead to overtreatment and patients receiving years of therapy when they would have done just as well

if the therapy was started at time of relapse? What does this mean for the feature of molecular testing on stage IB/II/IIA specimens?

Diffuse alveolar damage (DAD) Resulting from Coronavirus Disease 2019 Infection is Morphologically Indistinguishable from Other Causes of DAD.

Konopka et al, Histopathology

Background: As we are learning the histologic features of fatal COVID-related diffuse alveolar damage, it has not been clear if there are specific histological features (some have been proposed such as prominent vascular thrombosis/necrosis/inflammation, etc.), or if it may be histologically the same as every other cause of DAD.

Methods: 4 patients were identified who died in the community with a post-mortem diagnosis of COVID-19, and then matched with 3 different cohorts including 4 patients who died of COVID-19 after hospitalization, 4 DAD patients who died in the hospital before the COVID-19 pandemic began, and 4 patients who died in the community with autopsy finding of DAD. The cases were reviewed independently by 3 pulmonary pathologists, 2 of which were blinded to the cohorts. They reviewed and scored for histologic features including hyaline membranes, thrombi, airspace organization, AFOP, acute bronchopneumonia, perivascular inflammation, endotheliitis, fibrinoid vascular necrosis, and hemorrhage with capillaritis.

Results

- Underlying conditions were common including diabetes, cardiovascular disease, and obesity.

- Cases of death in the ARDS control group included bacteremia/sepsis, brain hemorrhage, and chronic bronchiectasis with pneumonia.

- Presence of hyaline membranes was unanimous in 14 of 16 cases, and in 9 cases there was also unanimous agreement about extent.

- Fibrinous thrombi were identified in 13 of 16 cases, with no differences in presence or extent between COVID cases and control cases.

- Organizing DAD/airspace organization was observed by at least one reviewer in 3 of 4 cases in hospitalized COVID patients but not in community COVID patients.

- Interobserver variability was greatest for AFOP-like changes, and there was no difference between COVID and control patients.

•There seemed to be more acute bronchopneumonia in hospitalized COVID patients than in COVID community patients.

•No cases had viral cytopathic effect, capillaritis with hemorrhage, or fibrinoid vascular necrosis.

Discussion: DAD seems very common at autopsy in patients with COVID, and thus it seems lung involvement is a common cause of death. No meaningful or unexpected differences were identified between the histopathologic patterns observed in hospitalized and community COVID patients, with both showing primarily DAD, and more organization in the hospitalized group, as expected. This indicates the DAD observed in hospitalized patients is very likely due to direct effects of the virus. Furthermore, there were no obvious histological differences between the DAD pattern observed in COVID patients vs. the control group. Specifically, the frequency and extent of fibrin thrombi was similar between these groups.

Take home point: In this small study, there was no difference in pulmonary pathology findings (primarily DAD) between hospitalized and community fatal COVID infection, aside from increased organization that would be expected in hospitalized patients that survived longer. DAD in COVID does not seem morphologically distinguishable from DAD of any other cause.

A Grading System for Invasive Pulmonary Adenocarcinoma: A Proposal From the International Association for the Study of Lung Cancer Pathology Committee.

Moreira et al, JTO

Background: Pulmonary adenocarcinoma shows many different histologic patterns, and the WHO currently recommends listing the predominant pattern. There are some gaps in this system, including the lack of a specific category for cribriform/complex gland pattern, which may be grouped with acinar-predominant tumors despite the fact that it is a high risk pattern based on current data. There have been studies that indicate other features including percentage of high grade patterns, nuclear grade, secondary patterns, mitotic activity, STAS, necrosis, etc. might add additional prognostic value. Despite the increasing evidence that histologic pattern-based grading in lung cancer has prognostic significance, no system has been universally endorsed.

Methods: This is a multi-institutional study of well-annotated adenocarcinoma cohorts with at least 5 years of follow-up. This included a training set of 284 stage I patients, a validation set of 212 stage I patients, and a testing set of 300 stage I-II patients. Comprehensive histologic subtyping was performed by the expert submitting pulmonary pathologists, which included specific consideration of cribriform/fused gland patterns. AIS, MIA, IMA, and multifocal adenocarcinomas were excluded. They also evaluated nuclear grade, mitotic count, cytologic

grade (low vs. high grade based on pleomorphism and cell size), STAS, and necrosis. This included an analysis of the highest grade areas ("hot spots"). They performed a reproducibility study for 23 cases, reviewed by 5 observers via scanned slides.

Results

- Higher nuclear grade, increased mitotic activity, necrosis and STAS were associated with high grade growth patterns. STAS was only observed in tumors with a high grade pattern component.

- High grade cytology was associated with solid and complex glandular growth patterns.

- Various iterations of pattern-based grading were applied (see table 2), and the two that performed the best included predominant pattern plus second most predominant pattern, and predominant pattern plus high grade patterns. They felt predominant + high grade was the most practically applicable.

- They assessed for the minimal amount of high-grade pattern (solid, micropapillary, complex glandular) that was required to influence the performance of the model. A cut-off of 20% performed the best.

- They evaluated whether adding additional histologic features would improve the grading system, including cytologic grade, mitotic activity, STAS, and nuclear grade, in various combinations, and did not find significant improvement in predicting recurrence or death.

- Based on these data, the following model was proposed (table 4):

    Grade 1: well-differentiated adenocarcinoma: lepidic predominant tumors with less than 20% of high- grade patterns (solid, micropapillary, and/or complex glandular).

    Grade 2: moderately differentiated adenocarcinomas: acinar or papillary predominant tumors with less than 20% of high-grade patterns.

    Grade 3: poorly differentiated: any tumor with 20% or more of high-grade patterns.

- In the validation set, the proposed grading system (predominant + high grade) was the best indicator of recurrence. Other factors were associated with recurrence including predominant pattern, nuclear/ cytological grade, mitoses, and STAS, but addition of these factors did not significantly improve the model.

- In the test cohort, they looked at the predominant + high grade grading system compared to predominant pattern only, with a more evident stratification of survival in the predominant + high grade system (figure 2).

•They performed a reproducibility assessment including 5 observers using WSI from 23 cases (all slides for each case). There was total agreement in about half, with kappa of 0.617 (substantial agreement). Most disagreements were between grades 1 and 2 (specifically between lepidic and papillary predominant). Disagreements between grades 2 and 3 were mainly due to disagreement between the percentage of high grade patterns present. When only 2 pathologist pairs were considered, the agreement was almost perfect.

Discussion: This grading system is designed to formulate a common language for prognostic groups based on architectural patterns for lung adenocarcinoma. This is a multi-institutional study which will hopefully reduce pathologist and institutional biases. The AUC for histological grade in predicting prognosis is about 0.7, so it is not perfect and there are other factors at play, but it would be a good benchmark to use standardized grading for future studies investigating other important prognostic factors. This system is not meant to replace the current system of assigning predominant pattern, but to complement it. Of note, this system did not include invasive mucinous adenocarcinoma, so future study would be needed to figure out what to do with those tumors.

Take home point: The IASLC is recommending a grading system based on predominant pattern plus high grade patterns: grade 1 lepidic predominant with <20% solid/micropapillary/complex glandular patters, grade 2 acinar or papillary predominant with 20% solid/micropapillary/complex glandular patters, and grade 3 any tumor with >20% solid/micropapillary/complex glandular patters.

Articles for Notation

Non-Neoplastic

Immunohistochemical expression of Napsin A in normal human fetal lungs at different gestational ages and in acquired and congenital pathological pulmonary conditions

Giordano et al, Virchows.

Summary: Napsin is a protease expressed on type 2 pneumocytes responsible for cleavage of surfactant protein B into its active form. The authors found napsin was expressed throughout the epithelium in pseudoglandular fetal lungs. 30 week fetuses and newborns both show a pattern similar to adults, with only scattered napsin positive cells in the alveolar epithelium. Napsin expression was increased in the pathologic setting (chorio, acute pneumonia, hypoplasia, etc.).

Take home point: Napsin is diffusely expressed in early fetal life and has an adult-like pattern by 30 weeks. It seems to be overexpressed in the setting of repair/ injury in fetuses and newborn infants.

Neoplastic

Digital Whole Slide Imaging Compared With Light Microscopy for Primary Diagnosis in Surgical Pathology: A Multicenter, Double-Blinded, Randomized Study of 2045 Cases.

Borowski et al, Archives.

Summary: 19 pathologists reviewed 5849 slides digitally via whole slide imaging and compared to glass reads, enriched for more diagnostically difficult cases. Washout was a minimum of 31 days. The gold standard was the original clinical diagnosis. Discrepancy rates were similar (3.64% for digital review, 3.2% for glass slides). Time to review slides was average of 5.2 minutes for digital review and 4.95 for glass.

Take home point: Digital WSI are non-inferior to glass slide review in primary diagnosis.

A Phase 2 Trial of Consolidation Pembrolizumab Following Concurrent Chemoradiation for Patients With Unresectable Stage III Non–Small Cell Lung Cancer: Hoosier Cancer Research Network LUN 14-179

Durm et al, Cancer.

Summary: This is a study of 93 patients receiving pembro as consolidation therapy after definitive chemorads for unresectable stage III NSCLC, who did not show progression during chemorads. Median follow-up was 32 months, and 43% were able to complete 12 months of therapy. Symptomatic pneumonitis occurred in 17% of patients. Improvement was observed in time to metastasis or death, progression free and overall survival. PDL1 status was assessed with 22C3 using modified proportion score, which counts tumor infiltrating immune cells in addition to tumor cell staining. PDL1 status (positive vs. negative) was not a significant predictor of time to metastasis or death, overall or progression free survival.

Take home point: Consolidation therapy with pembro after definitive chemorads improves outcome for patients with unresectable stage III NSCLC, and at least in this study, PDL1 IHC did not predict response with the caveat that they used a different score including tumor infiltrating immune cells, not TPS.

Atezolizumab for First-Line Treatment of PD-L1–Selected Patients with NSCLC

Herbst at al, NEJM

Summary: This is a study of 572 non-treated metastatic NSCLC, randomized to atezo vs. chemo. They used inclusion criteria of PDL1 (clone SP142) TPS $\geq$ 1 OR $\geq$ 1 tumor infiltrating immune cells expressing PDL1. In EGFR and ALK wild-type patients with positive PDL1, there was a 7 month increase in overall survival in the atezo group vs chemo (20 vs 13 months, HR 0.6, p=0.01). Overall rate of adverse events was similar (90% in atezo, 94% in chemo), with more severe adverse events in the chemo group (52% vs. 30% in the atezo group).

Take home point: Atezo demonstrated improved OS compared to conventional chemo in first-line therapy of NSCLC, for patients that had $\geq$ 1 TPS OR $\geq$ 1 tumor infiltrating immune cells.

Scoring of Programmed Death-Ligand 1 Immunohistochemistry on Cytology Cell Block Specimens in Non–Small Cell Lung Carcinoma: An Interobserver Agreement Study

Hernandez et al, AJCP

Summary: This is a reproducibility study of PDL1 (223C) expression determined in 54 NSCLC cell blocks, scored by 7 cytopathologists. This included 21 pleural fluids, 17 lung FNAs, 14 lymph node FNAS, 1 mediastinal FNA, and one bronchial brush. Scores were grouped as <1%, 1-49%, or 50% and higher. Total agreement between all 7 observers was observed in 48% of cases, with a majority opinion achieved in 98% of cases. Kappa was 0.6 (substantial agreement). Three of their cytopathologists had expertise in pulmonary pathology, and they agreed in 67% (kappa 0.63) compared to the non-pulmonary experts who agreed in 56% of cases (kappa 0.62). This was not significantly different. There was also no difference in agreement based on cellularity of the tumor block or collection method.

Take home point: Cell block PDL1 (22C3) interpretation agreement is "substantial", and does not seem effected by specific pulmonary expertise, collection method, or cellularity of the cell block.

Novel imprint cytological classification is correlated with tumor spread through air spaces in lung adenocarcinoma

Kimura et al, Lung Cancer

Summary: The authors have designed and previously reported a classification system for intraop cytology evaluation to detect STAS in lung adenocarcinoma, based on cell cluster shape, cell size, nucleus size, and presence of necrosis. These features are used to sort cases into 5 risk groups. This is a study of 164 intraoperative imprint cytology preps. STAS was present in about 17% of cases based on exam of the resected tumors, and this was significantly associated with

the group assigned based on intraop cytologic smear interpretation, with increasing presence of STAS with group (0% in group 1, 10.5% in group 2, 26% in group 3, and 43% of group 4-5).

Take home point: Cytological features seem to be associated with STAS, which can be detected on introp smears, although I am not sure this system is robust enough to make intra-operative decisions regarding proceeding from sublobar resection to lobectomy.

Analysis of real-world PD-L1 IHC 28-8 and 22C3 pharmDx assay utilisation, turnaround times and analytical concordance across multiple tumour types

Krigsfeld et al, J Clin Pathol

Summary: This is a large concordance study including 3,050 PDL1 tests, which had both 28-8 and 22C3 clones which could then be compared. This included multiple tumor types: lung cancer, urothelial carcinoma, melanoma, and squamous carcinoma of the head and neck. PDL1 values were grouped as follows: 0%, 1-4%, 5-49%, $\geq$50%. Strong correlation was observed between the 28-8 clone and the 22C3 clone, with Kendall's tau correlation of 0.94 for the overall population and 0.92-0.98 across the various tumor types.

Take home points: The 28-8 and 22C3 clones seem to perform in a similar fashion.

MiR-21, EGFR and PTEN in non-small cell lung cancer: an in situ hybridisation and immunohistochemistry study

Marin et al, J Clin Pathol

Summary: mir-21 is an oncogenic micro-RNA that is overexpressed in NSCLC. In this study they looked at 22 lung SQCC and 22 ADCA, and looked for mir-21 by ISH, and evaluated possible downstream targets or proteins that maybe responsible for its up-regulation by IHC on TMA tissue (EGFR, PTEN, surfactant protein A, p53), as well as doing a KI67. Expression of mir-21 was noted in tumor cells and associated stromal cells, and was most prominent in stromal cells immediately adjacent to tumor cells, and decreased as stromal cells got further from the tumor. It was more commonly expressed in ADCA than SQCC, but this was not significant. Expression of mir-21 was associated with increased PTEN expression but not with the other tested markers.

Take home points: mir-21 appears to be a microenvironment signaling molecule in NSCLC, possibly important in tumor characteristics relying on stromal-tumor interaction such as invasiveness and EMT. Its effects may be mediated by PTEN.

Frequent expression of conventional endothelial markers in pleural mesothelioma: usefulness of claudin-5 as well as combined traditional markers to distinguish mesothelioma from angiosarcoma

Nakashima et al, Lung Cancer

Summary: Sarcomatoid mesos often only express keratin without expression of other mesothelial markers, which can make them difficult to distinguish from other sarcomas that can express keratin, like angiosarcoma. This study looked at endothelial marker expression (CD31, CD34, ERG, factor VII and claudin-5) in 93 epithelioid, 29 sarcomatoid, and 25 biphasic mesotheliomas, compared to 41 angiosarcomas. Keratin positivity rate was over 90% for mesothelioma and less than 20% in angiosarcoma. Mesotheliomas expressed CD31 in 10.3% (although honestly their photo looks a lot like tumor-infiltrating macrophage staining), CD34 in 3.5%, ERG in 29%, and factor VIII in 3.4%. All mesos were negative for claudin-5.

Take home point: Endothelial marker expression can occur in meso, and in particular, this study further calls the specificity of ERG into question. Personally I see CD31 overcalled a lot due to tumor infiltrating macs, it is a hard stain to read. I wish they had included FLI1, which is my preferred "rule out angiosarc" stain- in my hands perhaps somewhat less sensitive than ERG, but more specific.

Molecular Landscape of BRAF-Mutant NSCLC Reveals an Association Between Clonality and Driver Mutations and Identifies Targetable Non-V600 Driver Mutations

Negrao et al, JTO.

Summary: *BRAF* mutations occur in about 4% of NSCLC, and only about half of those are V600E. This is a study of the Guardant 360 database which identified 305 unique non-V600E *BRAF* mutations; however, many of these (45%) were VUS. 90% were missense mutations. Two novel activating mutations were identified. Known activating mutations had higher clonality than VUS. Three patients were treated with MEK with or without BRAF inhibition, and only one had a durable response (*BRAF* L597R). Drug sensitivity testing using BRAF mutant cell lines showed favorable response to trametinib with or without dabrafenib, LXH254, and lifirafenib.

Take home point: Non-V600E BRAF mutations occur in NSCLC, and a subset of them may respond to targeted therapy. The clonality tends to be higher in true oncogenic mutations, which may help to distinguish them from mutations that are not contributing to oncogenesis.

Variants in Epithelial-Mesenchymal Transition and Immune Checkpoint Genes Are Associated With Immune Cell Profiles and Predict Survival in Non–Small Cell Lung Cancer

Parra et al, Archives.

Summary: This is a study of 164 NSCLC TMA cases that were subjected to multiplex immunofluorescence and image analysis to evaluate PDL1 expression in concert with a host of immune markers (CD68, CD3, CD20, PD-1, CD8, CD57, CD45RO, FOXP3). Low PDL1 expression was associated with smoking and ADCA histology. Low density of NK T-cells was also associated with smoking. High density of antigen-experienced T-cells was associated with brain mets. A high density of B-cells was more common in patients that did not receive adjuvant therapy. Genetic analysis showed mutations in the EMT-related gene ZEB2 were associated with immunologic ignorance and immune tolerance microenvironments, which suggests those tumors might be susceptible to immune checkpoint therapy. Lower risk of death was associated with high density of memory T-cells, a specific allele of CTLA4, and lack of ZEB2 mutations.

Take home point: A patient's genetic background regarding alleles regulating EMT and immune checkpoints are associated with different kinds of tumor immune microenvironments, and therefore might predict response to immune checkpoint inhibition.

Survival After Mediastinal Node Dissection, Systematic Sampling, or Neither for Early Stage NSCLC

Ray et al, JTO

Summary: This study looked at outcome for patients receiving mediastinal nodal dissection (which includes stations 2R, 4R, 7, 8, 9 and 10R on the right, and 5, 6, 7, 8, 9,and 10L on the left vs. systematic LN sampling (minimum of 2R, 4R, 7 and 10R on the right, and 5, 6, 7,and 10L on the left), or mediastinal nodal sampling that did not fulfill criteria for either of these. The study included 1942 patients; 18% had nodal dissection, 6% had systematic LN sampling, and 75% had neither. Interestingly, in teaching hospitals, nodal dissection was associated with decreased risk of death compared to the "neither" group, but not compared to the systematic sampling. There was no significant difference between any of the groups at non-teaching institutions. Periop complications were not significantly different between the groups.

Take home point: Get your mediastinal LN dissection done at a teaching hospital ☺.

Pleuropulmonary blastoma-like peritoneal sarcoma: a newly described malignancy associated with biallelic DICER1 pathogenic variation

Schultz et al, Mod Pathol

Summary: This is a series of 7 cases describing a very interesting peritoneal sarcoma that is essentially histologically indistinguishable from PPB. It occurs in kindreds with inherited *DICER1* mutations. All occurred in kids younger than 14, and primary sites included fallopian tube (4), serosa of colon (1), pelvic side wall (2).

Take home point: Tumors indistinguishable from pleuropulmonary blastoma can occur in the peritoneal/pelvic cavity in kids with *DICER1* mutations.

Clinical and Genomic Characteristics of Small Cell Lung Cancer in Never Smokers Results From a Retrospective Multicenter Cohort Study.

Thomas et al, Chest

Summary: In their cohort of 5,632 small cell carcinoma patients, only 1.8% were never smokers. Compared to smokers, never smokers with small cell lung cancer were more likely to be female, and present at extensive stage. Interestingly, never smokers were more likely to be at the extremes of age (35- 49 or >80). Genetic analysis of 9 tumors in never smokers showed lower mutation burden, lower frequency of *TP53* mutations, and absence of tobacco-associated signature. There was no difference in outcome between smokers and never smokers.

Take home point: Never smokers can rarely get small cell carcinoma, but constitute <2% of small cell patients. They are more likely to be women at the extremes of age, and show different genetic background than small cell in smokers, suggesting a different pathogenesis.

Characterization of *MET* exon 14 alteration and association with clinical outcomes of crizotinib in Chinese lung cancers

Yang et al, Lung Cancer

Summary: Large study looking for *MET* exon 14 skipping in 11,306 Chinese lung cancer patients, which found a frequency of 1.1%. These mutations were more common in older patients. Not surprisingly, crizotinib therapy was associated with improved PFS compared to chemo, but there was no difference in OS.

Take home point: *MET* exon 14 skipping mutations seem less common in Chinese patients compared to Western populations, and there is some evidence for improved PFS with crizotinib therapy.

PD-L1 lineage-specific quantification in malignant pleural effusions of lung adenocarcinoma by flow cytometry

Yoon et al, Lung Cancer

Summary: This study looked at assessing several antigens by flow [EpCAM, D2-40, PD-L1 (clone MIH1), CD3, CD20, CD45] in TTF1-positive lung adenocarcinoma present in pleural effusion. The PD-L1 could then be assessed by flow in a lineage-specific manner, and TPS could be calculated based on the flow findings. This was compared to PD-L1 (22C3) immunostains read manually on the cell block. The R value for correlation between flow-determined TPS and cell block IHC determined TPS was 0.8.

Take home point: Flow is a promising tool to determine PD-L1 in a non-manual way using pleural fluid from malignant effusions. It has the added benefit of knowing exactly which cells are tumor cells based on EpCam expression, which can sometimes be a struggle in effusion cytology.

When used together SS18–SSX fusion-specific and SSX C-terminus immunohistochemistry are highly specific and sensitive for the diagnosis of synovial sarcoma and can replace FISH or molecular testing in most cases

Zaborowski et al, Histopathology

Summary: There are 2 recently described antibodies aimed to help in diagnosis of synovial sarcoma. One targets the SS18-SSX fusion protein, and one targets the C-terminus of SSX. In this study they stained 39 synovial sarcoma samples from 25 patients with molecularly confirmed synovial sarcoma. 87% were positive for the fusion antibody IHC, and 92% were positive for the C-terminus antibody. False negatives were associated with decal. They also stained 580 other tumors which were all negative for the fusion antibody, and 7% were positive for the C-terminus antibody.

Take home point: The SS18-SSX fusion protein antibody for IHC had perfect specificity and good sensitivity at 87%. The C-terminus SSX antibody IHC was more sensitive at 92%, but not perfectly specific. False negatives occur in the setting of decalcification, but these antibodies may obviate the need for molecular testing in most cases of synovial sarcoma.

Incidence and outcome of post-transplant lymphoproliferative disorders in lung transplant patients: Analysis of ISHLT Registry.

Zaffiri et al, Histopathology.

Summary: This is a study of 454 patients with PTLD from the ISHLT database. Cumulative incidence of PTLD from database patients was 1.1% at one year and 4% at 10 years. About half occurred in the first year post-transplant. Independent risk factors for PTLD included age, EBV positive status, restrictive lung disease, induction, HLA types A1 and A24. HLA type DR11 was protective.

Take home point: About half of case of PTLD occurs in the first year, with 1% incidence at 1 year and 4% incidence at 10 years.

Association Between Circulating Tumor DNA Burden and Disease Burden in Patients With ALK-Positive Lung Cancer

Zhang et al, Cancer.

Summary: This is a study of 97 plasma samples from 75 patients with *ALK* rearranged lung cancer. *ALK* fusion was able to be detected in 79%, and *ALK* mutation was detected in 76%. Higher disease burden was associated with increasing amount of circulating tumor DNA, correlating most with disease burden in liver, bones and adrenals. Plasma *ALK* fusion allele frequency did not perform as well as maximum plasma alteration allele frequency or maximum *ALK* alteration allele frequency for predicting tumor burden.

Take home point: Disease burden correlates with genetic alterations that can be detected in plasma, including maximum plasma alteration allele frequency or maximum *ALK* alteration allele frequency.