Anatomic Pathology Abstracts

Editors: Rouzan Karabakhtsian, MD, PhD, associate professor of pathology, Albert Einstein College of Medicine, Montefiore Medical Center, Bronx, NY; Rachel Stewart, DO, PhD, molecular genetic pathology fellow, University of Utah/ARUP Laboratories, Salt Lake City; Nicole Panarelli, MD, associate professor of pathology, Albert Einstein College of Medicine, Montefiore Medical Center; and Shaomin Hu, MD, PhD, pathology resident, Albert Einstein College of Medicine, Montefiore Medical Center.

Confirmation of ProMisE: a clinical classifier for endometrial cancer

Classification of endometrial carcinomas by morphologic features is irreproducible and imperfectly reflects tumor biology. The authors developed the Proactive Molecular Risk Classifier for Endometrial Cancer (ProMisE) molecular classification system, based on The Cancer Genome Atlas genomic subgroups, and sought to confirm its feasibility and prognostic ability in a new, large cohort of endometrial carcinomas (ECs). They performed immunohistochemistry (IHC) for the presence or absence of mismatch repair proteins (to identify MMR deficiency [MMR-D]), sequencing for polymerase-ε exonuclease domain mutations (POLE EDMs), and IHC for p53 (wild type versus null/missense mutations; p53 wt and p53 abn, respectively) on 319 new EC samples. They then characterized and assessed subgroups relative to outcomes. The authors compared the prognostic ability of ProMisE with that of current risk-stratification systems (European Society of Medical Oncology [ESMO]). ProMisE decision-tree classification categorized all cases and identified four prognostic subgroups with distinct overall, disease-specific, and progression-free survival ($P < .001$). Tumors with POLE EDMs had the most favorable prognosis and those with p53 abn the worst. Separation of the two middle survival curves (p53 wt and MMR-D) was observed. There were no significant differences in survival rates between the ESMO low-risk and intermediate-risk groups. ProMisE improved the ability to discriminate outcomes compared with ESMO risk stratification. Substantial overlap (89 percent) was noted between the p53 abn and high-risk ESMO subgroups, but there were no predictable associations between molecular and ESMO risk groups. The authors concluded that molecular classification of ECs can be achieved using clinically applicable methods and provides independent prognostic information beyond established clinicopathologic risk factors available at diagnosis. Consistent, biologically relevant categorization enables stratification for clinical trials and targeted therapy, as well as identification of women who are at increased risk of having Lynch syndrome. It may also guide clinical management.

Talhouk A, McConechy MK, Leung S, et al. Confirmation of ProMisE: A simple, genomics-based clinical classifier for endometrial cancer. *Cancer.* 2017;123(5):802–813.

Correspondence: Dr. J. N. McAlpine at jessica.mcalpine@vch.ca

Leiomyoma with bizarre nuclei: a morphological, IHC, and molecular analysis

Leiomyomas associated with hereditary leiomyomatosis and renal cell carcinoma syndrome and leiomyomas with

bizarre nuclei often show overlapping morphological features, in particular cells with prominent eosinophilic nucleoli, perinucleolar halos, and eosinophilic cytoplasmic inclusions. Although hereditary leiomyomatosis and renal cell carcinoma syndrome is defined by fumarate hydratase (FH) germline mutations, resulting in S-(2-succino)-cysteine (2SC) formation, it is unknown whether leiomyomas with bizarre nuclei show similar alterations. The authors of this study evaluated the morphology and FH/2SC immunoprofile of 31 leiomyomas with bizarre nuclei. They subjected DNA from tumor and normal tissues of 24 cases to massively parallel sequencing targeting 410 key cancer genes. Somatic genetic alterations were detected using state-of-the-art bioinformatics algorithms. No patient reported a personal history of renal neoplasia or cutaneous leiomyomas, but one had a family history of renal cell carcinoma and another had a family history of uterine leiomyomas. Aberrant FH/2SC expression was noted in 17 tumors (16 FH negative/2SC positive, one FH positive/2SC positive). On univariate analysis, staghorn vessels, eosinophilic cytoplasmic inclusions, diffuse distribution of prominent eosinophilic nucleoli with perinucleolar halos, and an alveolar pattern of edema were associated with an abnormal immunoprofile, but only staghorn vessels remained significant on multivariate analysis. Massively parallel sequencing analysis (n=24) revealed that 13 of 14 tumors with aberrant FH/2SC immunoprofile harbored somatic FH somatic genetic alterations, including homozygous deletions (n = 9), missense mutations coupled with loss of heterozygosity (n = 3), and a splice site mutation (n = 1), whereas no somatic FH mutations/deletions were found in tumors with normal immunoprofile (n = 10; P<.0001). Leiomyomas with bizarre nuclei with normal FH/2SC staining pattern more frequently harbored TP53 or RB1 alterations, or both, than those with aberrant FH/2SC immunoprofile (60 versus 14 percent; P = .032). These data demonstrate that leiomyomas with bizarre nuclei are morphologically and genetically heterogeneous and that hereditary leiomyomatosis and renal cell carcinoma syndrome-related morphological features, abnormal FH/2SC staining, and somatic FH mutations/deletions can be seen in a subset of sporadic tumors.

Bennett JA, Weigelt B, Chiang S, et al. Leiomyoma with bizarre nuclei: a morphological, immunohistochemical and molecular analysis of 31 cases. *Mod Pathol.* 2017;30:1476–1488.

Correspondence: Dr. J. A. Bennett at jennifer.a.bennett@lahey.org

A new pathogenetic classification for invasive endocervical adenocarcinomas

The authors conducted a study to classify endocervical adenocarcinomas based on morphologic features linked to etiology—that is, human papillomavirus (HPV) infection—unlike the 2014 World Health Organization classification. The International Endocervical Adenocarcinoma Criteria and Classification (IECC criteria), described in the study, distinguishes between human papillomavirus-associated adenocarcinoma (HPVA), recognized by the presence of luminal mitoses and apoptosis seen at scanning magnification, and limited or no HPVA features (nonhuman papillomavirus-associated adenocarcinoma [NHPVA]). HPVAs were subcategorized based on cytoplasmic features, primarily to provide continuity with pre-existing classification schemes, whereas NHPVAs were subclassified based on established criteria, such as gastric type or clear cell. The authors collected complete slide sets for 409 cases from seven institutions worldwide. Tissue microarrays representing 297 cases were constructed, and immunohistochemistry—that is, p16, p53, vimentin, and progesterone receptor—and chromogenic in situ hybridization using an RNA-based probe set that recognizes 18 varieties of high-risk HPV were performed to validate IECC diagnoses. The five most common IECC diagnoses were usual type (HPVA; 73 percent of cohort); gastric type (NHPVA; 10 percent); mucinous adenocarcinoma of HPVA type, including intestinal, mucinous not otherwise specified, signet ring, and invasive stratified mucin-producing carcinoma categories (nine percent); clear cell carcinoma (NHPVA; three percent); and adenocarcinoma, not otherwise specified (two percent). Only three endometrioid carcinomas were recognized, and all were NHPVA. When excluding cases thought to have suboptimal tissue processing, 90 and 95 percent of usual-type IECC cases overexpressed p16 and were HPV+, whereas 37 and three percent of NHPVAs were p16+ and HPV+, respectively. The one HPV+ gastric-type carcinoma was found to have hybrid HPVA/NHPVA features on secondary review. The NHPVA tumors were larger and occurred in significantly older patients compared with HPVA tumors (P<.001). The high-risk HPV chromogenic in situ

hybridization probe set had superior sensitivity, specificity, and positive and negative predictive values (0.955, 0.968, 0.992, and 0.833, respectively) compared with p16 immunohistochemistry (0.872, 0.632, 0.907, and 0.545, respectively) to identify HPV-related usual carcinoma and mucinous carcinoma. IECC reliably segregates endocervical adenocarcinomas into HPVA and NHPVA types using morphology alone. This study confirms that usual-type endocervical adenocarcinomas are the most common type worldwide and that mucinous carcinomas comprise a mixture of HPVA and NHPVA, with gastric-type carcinoma being the major NHPVA type. Endometrioid and serous carcinomas of the endocervix are extraordinarily rare. If clinical outcomes and genomic studies continue to support these findings, the authors recommend replacing the World Health Organization 2014 criteria with IECC 2017.

Stolnicu S, Barsan I, Hoang L, et al. International Endocervical Adenocarcinoma Criteria and Classification (IECC): a new pathogenetic classification for invasive adenocarcinomas of the endocervix. *Am J Surg Pathol.* 2018;42(2):214–226.

Correspondence: Dr. R. A. Soslow at soslowr@mskcc.org

Prognostic value of PD-L1, p53, and Ki-67 for advanced-stage colorectal cancer

Prognostic indicators are ineffective for identifying advanced-stage colorectal cancer patients with high risk of recurrence after surgical resection. The authors investigated the prognostic value of p53, Ki-67, and programmed death ligand 1 (PD-L1) in 254 patients with stage II and III colorectal cancer. Expression of p53 was positive in 63 percent of cases. Upregulation of p53 was associated with smaller tumor size ($P = .001$) and higher Ki-67 labeling index ($P = .031$). The tumor Ki-67 labeling index was high (20 percent or more) in 197 (78 percent) of the patients. A high Ki-67 labeling index was associated with higher tumor-node-metastasis (TNM) stage ($P = .031$), positive p53 expression ($P = .031$), and negative PD-L1 expression ($P = .003$). The five-year relapse-free survival rates were 53 and 89 percent, respectively, for the p53-positive and Ki-67 labeling index-high patients and the p53-negative and Ki-67 labeling index-low patients ($P<.001$). In univariate analysis, negative p53 ($P = .001$), low Ki-67 labeling index ($P = .006$), low PD-L1 expression ($P = .044$), low TNM stage ($P<.001$), rectosigmoid location ($P = .026$), and small size ($P = .013$) were significantly related to relapse-free survival. In multivariate Cox regression analysis, positive p53 expression (hazard ratio [HR], 2.48; 95 percent confidence interval [CI], 1.34–4.59; $P = .004$), high Ki-67 labeling index (HR, 2.62; 95 percent CI, 1.12–6.14; $P = .027$), and high TNM stage (HR, 2.598; 95 percent CI, 1.55–4.37; $P <.001$) were independent predictors of unfavorable prognosis. The authors concluded that PD-L1, Ki-67, and p53 staining, individually, had significant prognostic value for patients with stage II and III colorectal cancer. Moreover, combining a p53 H-score of 35 or more and a Ki-67 labeling index of 20 percent or more identified patients with poor clinical outcome.

Wang L, Liu Z, Fisher KW, et al. Prognostic value of programmed death ligand 1, p53, and Ki-67 in patients with advanced-stage colorectal cancer. *Hum Pathol.* 2018;71:20–29.

Correspondence: Dr. X. Du at dx2008cn@163.com

Frequent homozygosity in mature and immature ovarian teratomas

Although homozygosity is well documented in mature teratomas, the genetic zygosity of ovarian immature teratomas and mixed germ cell tumors is less well studied. The authors conducted an analysis of 10 cases of mature cystic teratomas, 11 cases of grade 2 or 3 immature teratomas, and seven cases of mixed germ cell tumors with an immature teratoma component in which they used short tandem repeat genotyping to interrogate their genetic zygosity. DNA genotyping was informative in eight mature teratomas, seven immature teratomas, and six mixed germ cell tumors. Of the eight mature teratomas, five (63 percent) showed partial or complete homozygosity, with two (25 percent) cases demonstrating complete homozygosity. Of the immature teratomas, six

(86 percent) showed partial or complete homozygosity, with two (29 percent) demonstrating complete homozygosity. For the mixed germ cell tumors, two (33 percent) showed partial homozygosity and none displayed complete homozygosity. Long-term clinical follow-up was available for five immature teratomas (mean follow-up, 110 months) and five mixed germ cell tumors (mean follow-up, 66 months). None of the five patients with pure immature teratoma had a recurrence, whereas four of five mixed ovarian germ cell tumors recurred between four months and eight years ($P = .048$). The authors concluded that both immature and mature teratomas harbor frequent genetic homozygosity, suggesting a common cellular origin involving germ cells at the same developmental stage. The difference in the rate of homozygosity and tumor recurrence between pure immature teratomas and mixed germ cell tumors suggests that the two entities may involve different pathogenetic pathways and likely pursue different biological behaviors.

Snir OL, DeJoseph M, Wong S, et al. Frequent homozygosity in both mature and immature ovarian teratomas: a shared genetic basis of tumorigenesis. *Mod Pathol.* 2017;30:1467–1475.

Correspondence: Dr. P. Hui at pei.hui@yale.edu