Georgia Association of Pathology

GAPNEWS

MAY 2020

Welcome from the EDITOR

We are happy to launch our periodic GAPNews Newsletter. We hope this newsletter service will keep you updated on everything happening behind the scenes within GAP, in between the annual meetings. It is during these times more than ever, that we hope to rely on a network that connects us all together. We would love to keep building our community; we all share so much in common.

In our newsletter, we will provide updates on what is happening within the Georgia Association of Pathology, including planning for events and meetings. We hope to spotlight an active member of our community within each edition. We will highlight diagnostic updates within Pathology as a specialty, including a diagnostic multiple-choice case question. Finally, we will keep you informed about the legislative actions happening within our state and across the country, and their impact on our specialty.

Why have a State Pathology Association? The goal of the GAP is to bring together the community of pathologists so that we can share scientific advances in our field, promote awareness of our specialty, and identify and address advocacy opportunities for membership in the political landscape both on the state and national level. We also want to build membership by including fellows and residents in our activities. We believe that remaining engaged with membership by meetings, emails and newsletters will help us accomplish our goal.

HISTORY OF RE-LAUNCH

For many years, Georgia had an active pathology society. But, over time and due to a variety of reasons, the society became dormant. In 2018, the CAP learned of legislative issues potentially affecting pathologists in Georgia. They inquired about pathologist leadership/organization in Georgia and found it lacking. Those pathologists identified as current officers in the Georgia Association of Pathologists (old GAP) met and believed it was time for a management change in our state society. After several conference calls and vetting of management options, a new management company was selected, a contract signed, and the Georgia Association of Pathology (new GAP) was re-launched under a new name (Pathology instead of Pathologists).

Partnered with WJ Weiser, GAP leadership is excited about what we accomplish together. The GAP has a diverse leadership that strives to represent all pathologists in the state. Currently the GAP board has strong representation of private practice and academic institutions. We will strive to continue that in the future.

We are particularly lucky to have within our Board of Directors, Dr. Pat Godbey, who is currently the president of The College of American Pathologists and a staunch legislative advocate for all of us in the field.

RECENT GAP LEGISLATIVE/POLITICAL ACTIVITY

GAP officers actively participated in the Out of Network Physician Coalition.

By participating in a MAG established Out of Network Physician Coalition, GAP officers and their CAP partners were able to influence out of network legislation and insert language to protect pathologist interests. We believe the end result was fair and in some ways superior to what has passed in other states. Unfortunately, due to the COVID pandemic the legislative session was suspended before passage of the bill. Currently the out of network bill sits in limbo as we wait to see what happens at the Georgia capitol.

GAP officers played a role in getting the temporary waiver from CLIA provisions so that pathologists can work remotely.

With the help and guidance of the CAP, the GAP president contacted every member of the Georgia delegation to the US House of Representatives and requested their assistance in getting a CLIA waiver from Sec. Azar and the Centers for Medicare and Medicaid Services. The Honorable Buddy Carter (R) from Georgia's 1st district (lead signatory) and the Honorable Jody Hice (R) from Georgia's 10th district signed the letter asking for the waiver. On March 26th, the Trump administration announced the temporary waiver allowing pathologists to work remotely.

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CAP and GAP supported Georgia network adequacy disclosure legislation (HB 789).

The bill passed the House but did not receive Senate consideration because of the early adjournment for the public health emergency. The bill (HB 789) would have compelled health plans to disclose to consumers whether they had contracted with various specialty physicians at in-network hospitals. Plans that fail to indicate contracts with specialty physicians would be given a lower rating that consumers could rely upon when selecting their insurance provider.

RECENT GAP EDUCATIONAL ACTIVITY

Since the re-launch, we successfully held an annual meeting for state pathologists in October 2019. This meeting included great updates and presentations from our local academic and community pathologists. We had 50 attendees in total and would love for our community to keep growing. It was nice to see a lot of familiar faces and we welcomed new faces who joined us. We were happy to see a good number of future pathologists/trainees participate as well. A group of residents and fellows presented a wide range of case presentations, followed by great discussions.

Part of our efforts to become an active state association included creating a GAP Education Committee, to help keep updates in all specialties of Diagnostic Pathology at your fingertips. We sent a survey to hear your opinion and formulate our next steps. Thanks to all who have submitted their opinions. If you did not have the chance to complete the survey, you will have many chances in the future. We plan to send a yearly survey to make sure we are listening to our members and get their input.

The GAP Education committee has also put together a series of virtual online presentation for our members starting in May. This effort was in the planning, and was propelled to the spotlight by the COVID19 situation and the exponential growth of virtual platform usage. This is an experimental offering and we would like to hear from you if this is an activity you would be interested in participating in moving forward in the future. We are also working on securing CME credit for all attendees.

We welcome your feedback and would like to hear from you. Please do not hesitate to reach out to any of the board members anytime regarding anything, including educational and leadership opportunities in the GAP.

Lara Harik, MD
Board of Directors Member, Georgia Association of Pathology
Chair, GAP Education Committee
Anatomic Pathologist, Emory University Department of Pathology
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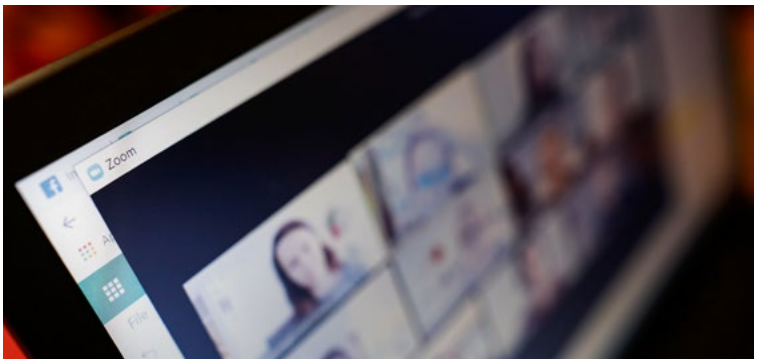
Mathew Fries, MD
President of Georgia Association of Pathology

GAP VIRTUAL PRESENTATIONS



Please join us for a series of GAP Virtual Presentations On Wednesday at Noon (12 pm), encompassing a variety of Pathology topics. We are working on securing CME credit for these presentations and we will keep you posted as things develop. The presentations can be accessed on Zoom with the following details:

Join Zoom Meeting:
https://us02web.zoom.us/j/89512348559?pwd=QmtLazdaRWVwLzlDa05pSTNEcm0xZz09

Meeting ID: 895 1234 8559
Password: 084693

June 11, 2020

Natasha M. Savage, MD
Associate Professor of Pathology & Residency Program Director, Medical College of Georgia at Augusta University Medical Director of Hematology & Hematopathology, AU Health

EOSINOPHILIA: GRAPPLING WITH GRANULES

Join Dr. Natasha M. Savage and the Georgia Association of Pathology to review eosinophilia. We will briefly discuss normal eosinophil morphology, origin, development, migration, and function. Then the differential diagnosis and work-up of eosinophilia will be discussed in detail with a case based approach. We will review the constellation of clinical and morphologic findings. In addition, we will discuss the appropriate, cost effective ancillary diagnostic evaluation for each entity. Do not miss this great educational opportunity.

THE LABORATORY DURING COVID-19

Jeannette Guarner, MD
Professor of Pathology and Vice Chair of the Department of Pathology and Laboratory Medicine
Emory University School of Medicine

During the COVID-19 pandemic we have heard the media talk about testing extensively. Obviously as laboratorians we know something about testing as we are the people doing this routinely. In the mind of many, testing refers to obtaining the sample for diagnosis of the disease. The majority of the population does not realize that in the mind of a laboratorian this is detection of viral nucleic acids from the nasopharynx and we know there are several platforms in which the testing can be performed. Unfortunately not all platforms are created equal though because of the race to get testing done we have been forced to forgo our usual motto of using only one platform in order to accommodate the needs. Independent to testing for diagnosis there have been some very interesting shifts in other requests for the laboratory:

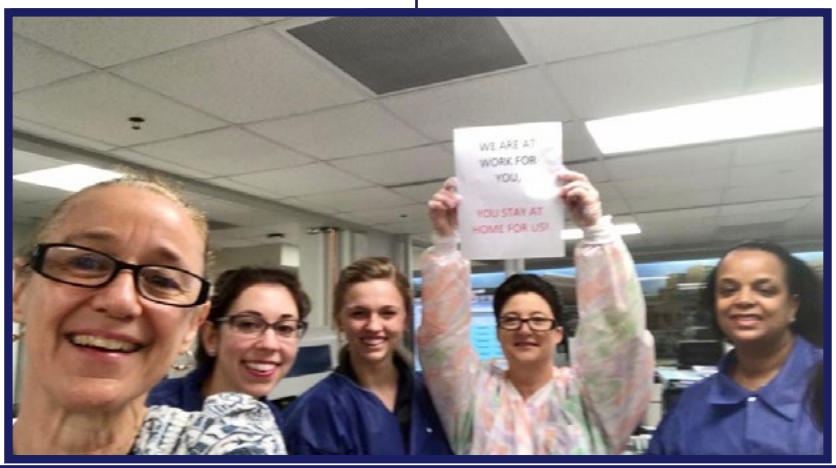
One of the bigger shifts has been an increase in D-dimer testing. Since we started receiving patients at Emory Healthcare, the number of requests for D-dimer have increased to double the usual amount. The Chinese used D-dimer as a marker of risk severity. What we have found is a major increase in thrombotic events in patients with COVID-19. They have pulmonary emboli, their lines clot and have higher risk of embolic events. Some have suggested it is a hypercoagulable state; however, one needs to realize the situation these patients are in: They are immobilized as they are on ventilators and they have multiple comorbidities. In addition, they have an infection thus their inflammatory markers are up. Then it is not surprising their coagulation is activated. The increase in D-dimer requests has been accompanied by other coagulation requests such as fibrinogen and fibrin monomers. Fortunately, many centers have realized that an important component of treating these patients include anticoagulation.

Three other tests that have increased in volume include ionized calcium, C-reactive protein and ferritin. The increase in ionized calcium is from the dialysate of patients receiving continuous renal replacement treatment as many of the COVID-19 patients have renal failure. In any particular day we may have in the hospital several patients in intensive care units receiving this treatment and we have to test the dialysate several times in a given day each patient. The increase in C-reactive protein is not surprising as this a marker of inflammation. Ferritin has also been used as a risk marker in these patients. Many believe that there is macrophage activation syndrome which is characterized by fever, increased ferritin, cytopenias, and hypertriglyceridemia. Further studies are needed to define if this is the reason why there is increase in this marker.

Lastly, there are many reasons to implement serological tests for detection of antibodies against SARS-CoV-2 as these can define who has been exposed to the disease. In general serological tests should primarily be performed for epidemiological purposes: define healthcare workers that have been exposed and determine the seroprevalence of the disease and the true mortality. However, serology could potentially be used for diagnosis of those patients that presented with symptoms of COVID-19 and tested negative when using the nucleic acid test or in the extreme case in which we run out of nasopharyngeal swabs and the only possibility of diagnosis is serology.

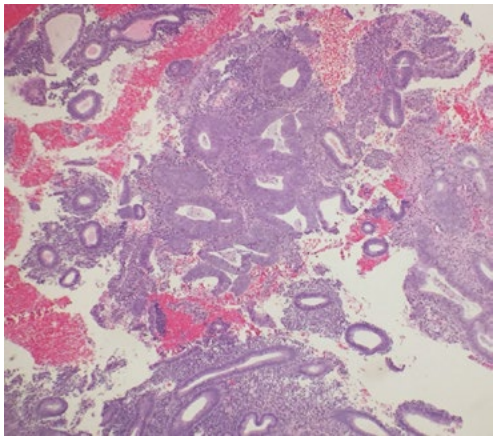
During this uncertain times, my role as the medical director of the laboratory has been to explain to lab techs what SARS-CoV-2 is, how it is transmitted, and calm laboratory staff as we are using universal precautions when handling specimens. This is not a blood-borne virus and the risks to laboratorians handling blood specimens is very small. Even when handling nasopharyngeal specimens the risk is extremely small as this is done in biosafety hoods with adequate personal protective equipment. It is our mission as pathologists to be available to answer questions and be present. I have been told by my techs in the core lab and blood bank that seeing me in the laboratory gives them strength to come and do their job.

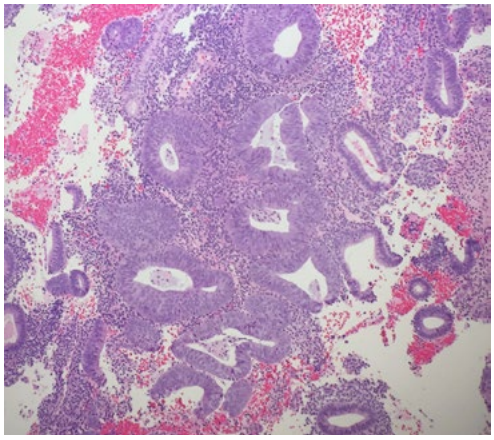
In conclusion, the pandemic of COVID-19 has impacted the laboratory beyond nucleic acid testing for SARS-CoV-2. It has increased testing in some areas that have been unexpected. We need to realize that the presence of the pathologist in the lab cannot be underestimated as we can provide strength to laboratorians performing their jobs every day.

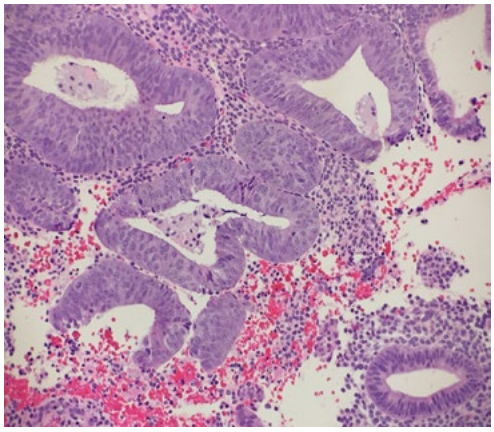


DIAGNOSTIC CASE

A 50 year-old female patient, with a history of uterine fibroids, presents with abnormal uterine bleeding and a thickened endometrial stripe seen on pelvic ultrasound. Endometrial biopsy is performed and shows the histologic findings below.







Which of the following is a diagnostic criterion for the above entity?

A. Minimum size of 1 mm
B. Gland to stroma ratio exceeding 1:1
C. Presence of cytologically distinct glands compared to the background endometrium
D. Option B and C
E. All of the above

Please see page 7 for the answer and explanation.

THE LABORATORY RESPONSE TO SARS-COV-2 IN GEORGIA

Authors: Charles E. Hill, MD, PhD and Charles Meyer, MD from Emory University Department of Pathology and Ravindra Kolhe, MD from Augusta University Department of Pathology

The global pandemic of SARS-CoV-2 has significantly affected essentially every state in the US, with Georgia being one of the medium-sized epicenters. The clinical laboratories in the state of Georgia have responded to this challenge by rapidly implementing testing, both molecular and serologic. Now, a little more than 2 months into the healthcare response, Georgia has had almost 30,000 confirmed cases and over 1300 deaths associated with COVID-19. While barriers remain to increased testing, over 200,000 Georgia residents have now been tested.

In early February, testing for SARS-CoV-2 was limited to the CDC and a small number of public health laboratories. The FDA issued a new policy to facilitate diagnostic test development for SARS-CoV-2 on February 29, 2020. While the regulatory framework that resulted from this policy has expedited some aspects of getting new tests to market, it provided some confusion to individual laboratories that would have normally offered laboratory developed tests without submission of data to the FDA. In an effort to speed the process, the FDA did allow labs to offer testing clinically while awaiting a response from the FDA regarding their test. This process, from data submission to response, was often protracted to two weeks or more and, in some cases, the FDA determined that a new Emergency Use Authorization was not needed after review by FDA staff. By mid-March, a few laboratories in Georgia were offering SAR-CoV-2 testing, including the Georgia Public Health Laboratory, Emory Medical Laboratory, and Augusta University Health.

Because of the rapid increases in worldwide demand for testing materials, supply chain shortages emerged as a major barrier to increasing test capabilities. In March, this consisted of reagents and consumables needed to perform viral RNA testing. As that supply chain began to become more secure, collection devices and viral transport medium supplies became limiting. This has led to the validation of saline and alternative collection devices on multiple test platforms, however many components continue to remain in short supply.

In response to many of these challenges, Governor Kemp and the State of Georgia assembled a consortium of universities that includes Augusta University, Emory University, Georgia State University, Georgia Tech, the University of Georgia, the Georgia Department of Public Health, and the Georgia National Guard. The primary focus of this group has been to increase the capacity for testing available to the citizens of Georgia and to coordinate supply chains so that there is a reliable supply of materials and limit fragmented requests for the same supplies from all of the different labs.

At this point, there is now a significantly increased testing capacity, both molecular and serologic across the State of Georgia. These efforts continue to ensure that, as the economy of the State of Georgia attempts to reopen, that testing is widely accessible. These efforts are also likely to yield a more rapid scale up of response if there is an increase in SARS-CoV-2 later in the year. However, as we get closer to the time of year in which seasonal respiratory virus is more common, a change of strategy may need to be implemented. Multiplex respiratory to include SARS-CoV-2 in conjunction with the more common causes of influenza like illness may need to become the norm instead of individual virus testing for SAR-CoV-2 alone.

INITIAL EMORY EXPERIENCE WITH COVID-19 SEROLOGICAL TESTING

As the world and medical community negotiate the COVID-19 pandemic, one notes the daily news covering specifics of medical laboratory testing at a level of granular detail usually only seen at specialty conferences. Discussions of strengths and weaknesses of PCR based nucleic acid testing have become common talking points, and as the trajectory of the pandemic moved on, attention has turned to the potential characterization of immunity in recovered patients, identification of putatively asymptomatic individuals, and calculation of the true population prevalence of the disease, all issues that can be addressed, if imperfectly, by classical serological methods. Herein, I discuss the initial period of development and implementation of a serological test for COVID-19 in the Immunopathology laboratory at Emory University Department of Pathology.

SARS-CoV-2 is a novel human beta-coronavirus, recognized as the causative agent of pandemic COVID-19 disease. The virus emerged in Wuhan, China in late 2019. Closely related to SARS-associated coronavirus (SARS-CoV) isolated in 2003, SARS-CoV-2 is more distantly related to MERS-CoV and the seasonal disease-causing human coronaviruses (HKU1, OC43, NL63, 229E) generally associated with mild upper respiratory infections. The pathogen is an enveloped positive sense single stranded RNA (+ssRNA) virus harboring a 29.9 kB genome, which encodes viral structural and non-structural proteins, including a large (140 kDa) homotrimeric glycoprotein (spike or S-protein). The S-protein of the virus extends beyond the viral envelope and is necessary for interaction with cells of the respiratory tract and other organ systems.

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A receptor binding domain (RBD) is the site of direct interaction between the S-protein and its cellular receptor, angiotensin converting enzyme 2 (ACE-2). Importantly, sequence comparisons with SARS-CoV-2 and other coronaviruses show limited homology in the RBD region, especially to the seasonal coronaviruses. Furthermore, test designers reasoned that antibodies to this region may have neutralizing properties, meaning that antibodies binding to this epitope *in vivo* may prevent or reduce viral entry or inhibit the viral life cycle. Thus, the qualitative serological assay now in utilization is a manual enzyme-linked immunosorbent assay (ELISA) adapted from methods developed by viral immunologists at the Emory Vaccine Center (1), utilizing the RBD fragment of SARS-CoV-2 S-protein as capture antigen. The recombinant protein is immobilized to a polystyrene 96 well plate, with subsequent detection of bound immunoglobulin by an HRP conjugated anti-human IgG. Initial validation studies utilized over 900 pre-2020 archived sera as representative negative controls and over 200 nasopharyngeal swab PCR positive and clinically confirmed COVID-19 cases as positive controls. At currently utilized testing cut-off, the assay has a sensitivity and specificity were calculated at 100% and 98%, respectively. Cross reactivity is not seen with sera positive for other viral infections (HIV, HBV, HCV, CoV HKU1, CoV OC43, CoV 229E, RSV, Rhinovirus, influenza A and B, and H. influenzae) and confounders (autoimmune antibody and rheumatoid factor) often seen in a clinical immunology laboratory. Recombinant RBD protein is produced in facilities within Emory University, a process assisted greatly by the institutional expertise of Emory biochemists and vaccinologists. The decision to pursue an in-house developed assay was strongly influenced by concern over lack of a stable and consistent supply chain for key reagents, a situation that has plagued full implementation of nucleic acid testing.

To date (5/6/2020), we have performed over 5000 assays, with a maximal daily capacity of 300 samples daily using the manual ELISA. Efforts are underway to adapt the method to automated instrumentation that could expand testing capacity more than tenfold, allowing practical community based sero-surveys. In a pilot survey of Emory health care workers, 8-10 percent have been shown to be positive with our IgG assay.

As with any new serological assay there are a number of known unknowns, and in this rapidly evolving situation, much concern has been expressed in the scientific and lay press regarding the quality and predictive value of many the serological assays now appearing. Critical questions include the duration and strength of immunity, which is limited in seasonal coronavirus infections, but likely more durable with MERS/SARS, which are more closely related to SARS-CoV-2 (2). Early studies indicate that host IgG and IgM antibodies are produced against SARS-Cov-2 within 5-7 days post symptom onset, a time when patients are positive by nucleic acid amplification test (NAAT) (indeed, a dangerous feature of COVID-19 is virus shedding and infectivity several days prior to symptom onset, thus illustrating a fundamental and inherent limitation of the serological assay in acute phase diagnostic testing). Thus, our test reporting to ordering physicians indicates explicitly that seropositivity does not necessarily equate with functional immunity and notes that seropositivity is not a guarantee that the patient has ceased shedding virus. It is anticipated that future iterations of in house testing may include confirmatory sequential assay for IgM and IgA, or an alternative platform that detects IgG to S-protein or another viral antigen such as nucleocapsid, and will show continued improvement in performance. Beyond traditional serology, *in vitro* cellular based viral-entry neutralization assays are being developed as a potential clinical test that would represent a powerful method to ascertain the level of immunity associated with seropositivity.

A validated and highly sensitive serological test for SARS-CoV-2 may have diagnostic utility when employed with appropriate history taking and timed correctly, though the gold standard for diagnosis of infection remains nucleic acid based testing. Persons of interest for COVID-19 that could not be tested or were negative on the molecular test, could be evaluated for seropositivity to attribute symptoms to SARS-CoV-2. Additionally, idiosyncratic sequelae, such as post infection Kawasaki disease, would require serological confirmation of past infection. Serological assays serve the unique purpose of providing a “retrospectoscope” to determine past infection, and as such have tremendous public health value in their ability to assess population percentages that have been exposed to a given pathogen and allow calculation of true morbidity and mortality from the disease. More controversially, seropositivity has been viewed in some quarters as an “immunological passport” that will allow those with antibodies (whether they were symptomatic or not) to consider themselves immune and return to work. Such hopes must be tempered by the nascent understanding of the immune response to SARS-CoV-2 (a rapidly changing situation).

There is great interest is the potential utilization of convalescent plasma as a therapeutic modality. Broad based serological testing is vital for detection of seropositive individuals whose plasma could be further characterized for neutralizing activity and used in a passive immunity treatment strategy. Finally, anticipating development of an effective vaccine, serological assays with the appropriate viral antigen will ultimately be vital to confirm immunity post vaccination, as is currently performed for Hepatitis B and Measles/Mumps/Rubella/VZV. The rapid development of high quality, scalable serological testing with a secure supply chain of reagents will be useful component in the armamentarium against the current pandemic.

Andrew Neish, MD
Professor of Pathology
Medical Director, Immunopathology Laboratories, Emory Medical Laboratories

The author would like acknowledge the outstanding and unprecedented effort of the virologists, biochemists, clinical chemists, pathologists, medical technicians and students who were vital to this effort.

1. Suthar, MS, et al. Rapid generation of neutralizing antibody responses in COVID-19 patients, submitted
2. Altman, DM et al. What policy maker need to know about COVID-19 protective immunity, Lancet April 27, 2020 S0140-6736(20)30985-5

DIAGNOSTIC CASE ANSWER

CORRECT ANSWER

The correct answer is Option E.

Explanation of the correct answer:

The endometrial biopsy demonstrates a focus of glandular crowding (defined as an increase in the normal gland to stromal ratio, with only a small amount of stroma separating individual glands. These crowded glands are cytologically altered as compared to the surrounding background endometrial glands, and display nuclear enlargement, vesicular chromatin, prominent nucleoli, pseudostratification of the nuclei, and mitotic figures. The focus measures approximately 1.5 mm in greatest dimension.

Together, these findings fulfill the 5 diagnostic criteria for endometrial intraepithelial neoplasia (EIN), which are as follows: 1) architectural glandular crowding; 2) altered cytology relative to background glands; 3) minimum size of 1 mm; 4) exclusion of adenocarcinoma; and 5) exclusion of mimickers.

Endometrial Intraepithelial Neoplasia (EIN) – Brief Summary:

EIN is defined as a monoclonal, premalignant proliferation of cytologically altered endometrial glands that reflects the molecular progression to endometrial endometrioid adenocarcinoma.

The primary risk factor for the development of EIN is prolonged, unopposed estrogenic endometrial stimulation. Unopposed estrogen may occur in various clinical settings, such as obesity, anovulatory cycles, and polycystic ovarian disease.

The EIN classification system is two-tiered, with cases designated either as EIN or benign hyperplasia (BH). This two-tiered system is in contrast the WHO 1994 (WHO94) system, which categorizes lesions into four subtypes based on glandular complexity and cytologic atypia (i.e., simple or complex hyperplasia with or without atypia). The WHO94 system has shown poor interobserver reproducibility. The poor reproducibility, as well as increased understanding of the genetics of EIN and endometrial carcinoma, have driven an ongoing discussion on ways to improve or replace WHO94. Notably, independent investigators have demonstrated superior diagnostic reproducibility of the EIN system as compared to WHO94.

In general, use of immunohistochemistry is not necessary for the routine diagnosis of EIN. However, in some cases, immunohistochemical stains for *PTEN* and *PAX2* may be helpful in establishing the diagnosis. Genetic alterations involving *PTEN*, *PAX2*, *KRAS*, and microsatellite instability have all been associated with endometrial adenocarcinoma and its precursor EIN.

In normal endometrium, *PTEN* displays nuclear and cytoplasmic staining and PAX2 displays nuclear staining. *PTEN* expression is lost in 83% and 63% of endometrial endometrioid adenocarcinomas and EIN lesions, respectively, and likewise *PAX2* loss of expression is seen in 77% of adenocarcinomas and 74% of EIN cases. It is good to keep in mind that sometimes normal background endometrium may also display small foci of *PTEN* or *PAX2* loss and these stains should be used and interpreted with caution.

For patients wishing to preserve fertility or those whom may not be surgical candidates, EIN may be managed conservatively with a trial of hormonal therapy and clinical follow-up including endometrial re-sampling. Hysterectomy is generally recommended for patients who fail conservative treatment or are at high risk for developing endometrial adenocarcinoma.

Authors

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References

Kurman RJ, Kaminski PF, Norris HJ. The behavior of endometrial hyperplasia. A long-term study of “untreated” hyperplasia in 170 patients. Cancer 1985;56:403-12. PMID: 4005805

Baak JP, Mutter GL. EIN and WHO94. J Clin Pathol 2005;58:1-6. PMID: 15623473

Owings RA, Quick CM. Endometrial intraepithelial neoplasia. Arch Pathol Lab Med 2014;138:484-91. PMID: 24678678

We hope you enjoy the inaugural edition of GAP News! In the future this content will be available to GAP Members only. If you are not already a member of the GAP, we invite you to join our community of professionals today. We strive to serve a voice for all pathologists in the state of Georgia and are made stronger by your involvement!