ThinPrep® Non-Gyn Lecture Series

Body Fluid Cytology

Benefits of ThinPrep® Technology

The use of ThinPrep Non-Gyn for body fluid specimens:

- Optimizes cell preservation
- Standardizes specimen preparation
- Simplifies slide screening
- Offers the versatility to perform ancillary testing

Required Materials

- ThinPrep® 2000 Processor or ThinPrep 5000 Processor
- ThinPrep Microscope Slides
- ThinPrep Non-Gyn Filters (Blue)
- Multi-Mix™ Racked Vortexor
- CytoLyt® and PreservCyt® Solutions

Required Materials

- 50 ml capacity swing arm centrifuge
- 50 ml centrifuge tubes
- Slide staining system and reagents
- 1 ml plastic transfer pipettes
- 95% alcohol
- Coverslips and mounting media

Optional: glacial acetic acid and saline for troubleshooting

Recommended Collection Media

- CytoLyt®
- Plasma-Lyte®
- Polysol®
- Balanced electrolyte solutions

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Non-recommended Collection Media

- Normal Saline
- Culture Media
- RPMI
- PBS
- Solutions containing formalin

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Hologic® Solutions

- CytoLyt®
- PreservCyt®

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Hologic® Solutions

CytoLyt® Solution

- Methanol-based, buffered preservative solution
  - Lyses red blood cells
  - Prevents protein precipitation
  - Dissolves mucus
  - Preserves morphology for 8 days at room temperature
- Intended as transport medium
- Used in specimen preparation prior to processing

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Hologic® Solutions

PreservCyt® Solution

- Methanol based, buffered solution
- Specimens must be added to PreservCyt Solution prior to processing
- PreservCyt Solution cannot be substituted with any other reagents
- Cells in PreservCyt Solution are preserved for up to 3 weeks in a temperature range between 4 -37 C

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Sample Collection

Fluid Specimens

- Concentrate fresh sample by centrifugation before adding CytoLyt® Solution
  - If not possible, collect samples directly into CytoLyt Solution
    - The minimum CytoLyt to sample ratio should be 1:3

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Sample Preparation

1. Sample collection
2. Concentrate by centrifugation 600g for 10 min - alternately you may centrifuge 1200g for 5 min
3. Pour off supernatant and vortex to resuspend cell pellet
4. Add 30mls of CytoLyt® Solution. Repeat centrifugation, pour off supernatant
5. Evaluate cell pellet . If cell pellet is not free of blood, do a CytoLyt wash and repeat steps 2-4
6. Add recommended # of drops of specimen to PreservCyt® Solution Vial
7. Allow to stand for 15 minutes
8. Prepare slide on ThinPrep® 2000 using Sequence 2 or ThinPrep 5000 using Sequence Non-gyn
9. Fix, Stain, and Evaluate

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Sample Collection

- Fresh – recommended

  -For extremely bloody fluids, start processing with only 10 ml of fresh fluid

- CytoLyt®

  -If fresh collection is not possible, collect samples directly into CytoLyt Solution

  -The minimum CytoLyt to sample ratio should be 1:3 and is not considered a wash step, but only a collection step. A CytoLyt wash is required prior to instrument processing

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Sample Preparation Techniques

- Centrifugation-600g for 10 minutes or 1200g for 5 minutes

  - Concentrate cellular material in order to separate the cellular components from the supernatant

Refer to Centrifuge Speed Chart in the ThinPrep® 2000 or ThinPrep 5000 Owner's Manual, Non-Gynecologic section to determine the correct speed for your centrifuge to obtain force of 600g or 1200g

Sample Preparation Techniques

- Pour off supernatant

  - Invert the centrifuge tube 180 in *one smooth movement*, pour off all supernatant and return tube to its original position

  (Note: Failure to completely pour off the supernatant may result in a sparsely cellular sample due to dilution of the cell pellet).

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Sample Preparation Techniques

- Vortex to re-suspend cell pellet

  - Randomize the cell pellet and to improve the results of the CytoLyt® solution washing procedure

  - Place the centrifuge tube onto a vortexor and agitate the cell pellet for 3 seconds or vortex manually by syringing the pellet back and forth with a plastic pipette

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Sample Preparation Techniques

- CytoLyt® Solution Wash

  - Preserve cellular morphology while lysing red blood cells, dissolving mucus and reducing protein precipitation

  - Add 30 ml of CytoLyt Solution to a cell pellet, concentrate by centrifugation, pour off the supernatant, vortex and evaluate cell pellet

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Sample Preparation Techniques

- Evaluate cell pellet
  - If cell pellet is white, pale pink, tan or not visible, calculate number of drops of specimen to be added to the PreservCyt® Solution Vial *(will be discussed in detail on future slides)*
  - If cell pellet is distinctly red or brown indicating the presence of remaining blood, conduct a second CytoLyt® Wash.

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Sample Preparation Techniques

- Calculate how many drops of specimen to add to PreservCyt® vial:

  - If pellet volume is > 1ml *(if not consider next 2 slides)*

    - Add 1ml of CytoLyt® Solution into the tube and vortex briefly to resuspend the cell pellet
    - Transfer 1 drop of the specimen to a fresh PreservCyt Solution Vial

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Sample Preparation Techniques

- Calculate how many drops of specimen to add to PreservCyt® vial:

  - If pellet is clearly visible and pellet volume is < 1ml *(if not consider next slide)*

    - Vortex pellet and transfer 2 drops to a fresh PreservCyt solution vial

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Sample Preparation Techniques

- Calculate how many drops of specimen to add to PreservCyt® vial:
  - If pellet is not visible or scant
    - Add contents of a fresh PreservCyt Solution Vial into the tube and vortex briefly to mix the solution
    - Pour entire sample back into the vial

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Sample Preparation Troubleshooting

- Due to the biological variability among samples and variability in collection methods, standard processes may not always yield a satisfactory and uniformly distributed preparation on the first slide.

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Sample Preparation Troubleshooting

- After staining, you may observe the following irregularities:
  - Non-uniform distribution of cells in the cell spot *without* a “sample is dilute” message
  - Uneven distribution in the form of a ring or halo of cellular material and/or white blood cells
  - A sparse cell spot lacking in cellular component and containing blood, protein and debris – may be accompanied by a “sample is dilute” message

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Techniques Used in Troubleshooting

- Diluting the Sample 20 to 1
- Glacial Acetic Acid Wash for Blood and Non-Cellular Debris
- Saline Wash for Protein

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Techniques Used in Troubleshooting

- Diluting the Sample 20 to 1

  - Add 1ml of the sample that is suspended in PreservCyt® Solution to a new PreservCyt Solution vial (20ml). This is most accurately done with a calibrated pipette.

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Techniques Used in Troubleshooting

- Glacial Acetic Acid Wash for Blood and Non-Cellular Debris

  - If sample is bloody, it can be further washed using a solution of 9 parts CytoLyt® Solution and 1 part Glacial Acetic acid.

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Techniques Used in Troubleshooting

- Saline Wash for Protein

  - If sample contains protein, it can be further washed with saline solution in place of CytoLyt® Solution.

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Troubleshooting
Bloody or Proteinaceous Specimens

"Sample is Dilute" message

Yes

Check to see if cellularity is adequate. If not, use more of the pellet, if available and process new slide.

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No, continue to next slide

Troubleshooting
Bloody or Proteinaceous Specimens

Does the slide have a “halo” of cellular material and/or white blood cells?

Yes

Dilute 20:1 using a calibrated pipette to add 1ml of residual sample to a new PreservCyt® Solution Vial. Prepare new slide.

If halo is present on the new slide, contact Hologic® Technical Service.

No, continue to next slide

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Troubleshooting
Bloody or Proteinaceous Specimens

Is the slide sparse and does it contain blood, protein or non-cellular debris?

Yes-blood or non-cellular debris

Centrifuge remaining specimen from PreservCyt® vial, pour off and vortex. Add 30ml of a 9:1 CytoLyt® to glacial acetic acid solution to the sample, centrifuge, pour off and vortex. Add appropriate number of drops to PreservCyt vial and prepare new slide. *If resulting slide is sparse, contact Hologic Technical Service.*

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No

Yes-protein

Centrifuge remaining specimen from PreservCyt vial, pour off and vortex. Add 30 ml of saline to sample, centrifuge, pour off and vortex. Add appropriate number of drops to PreservCyt vial and prepare new slide. *If resulting slide is sparse, contact Hologic Technical Service.*

Contact Hologic® Technical Service

Troubleshooting Common Artifacts

- Smudged Nuclear Detail
- Compression Artifact
- Staining Artifact
- Edge of the Cylinder Artifact

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Troubleshooting Common Artifacts

- Smudged Nuclear Detail
  - May occur if specimen is collected in saline, PBS or RPMI
  - To avoid this, collect the sample either fresh, in CytoLyt® or in PreservCyt® solution

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Troubleshooting Common Artifacts

- Compression Artifact
  - Appears as “air dry” artifact on the perimeter of the cell spot
  - Due to the compression of cells between the edge of the filter and the glass of the slide

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Troubleshooting Common Artifacts

- Staining Artifact
  - Mimics air-drying
  - Appears as a red or orange central staining primarily in cell clusters or groups.
  - Due to the incomplete rinsing of counterstains.
  - To eliminate this artifact, fresh alcohol baths or an additional rinse step after the cytoplasmic stains is required.

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Troubleshooting Common Artifacts

- Edge of the Cylinder Artifact
  - Narrow rim of cellular material just beyond the circumference of the cell spot.
  - Result of cells from the outer edge of the wet filter cylinder being transferred to the glass slide.

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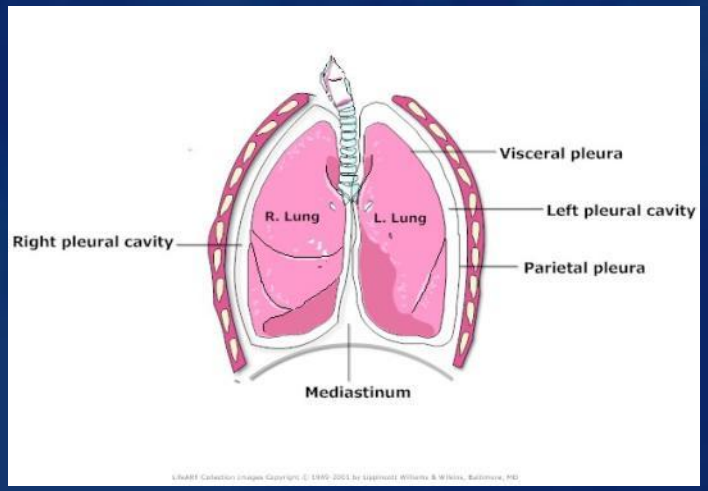
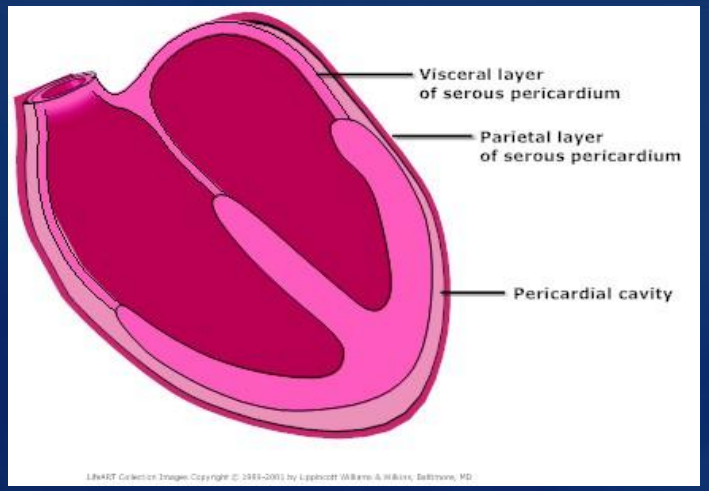
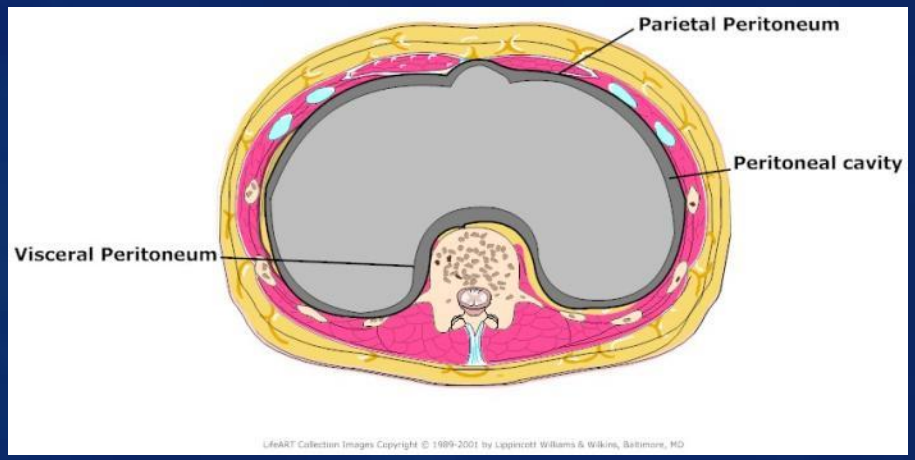
Anatomy of Body Cavities







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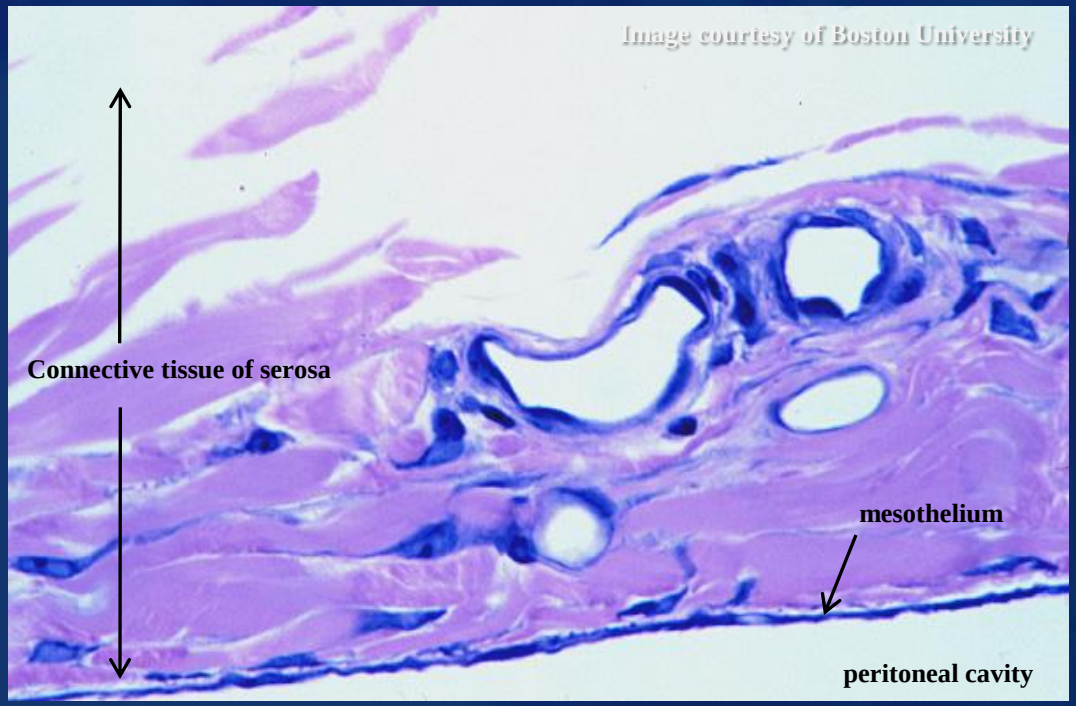
Histology of the Epithelium



- Serous membranes consist of connective tissue that is normally lined by a single layer of mesothelial cells
- Clear, watery fluid (serous fluid) is produced, which lubricates the organs
- Any excess of serous fluid is an effusion and indicates a disease state

Biological Nature of Effusion

- An effusion is any excess amount of serous fluid in a body cavity
- Always caused by a pathologic process
- Fluid is reabsorbed after successful treatment

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Biological Nature of Effusion

- Effusions are classified into four categories
  - Hydrostatic
  - Infectious
  - Noninfectious inflammatory
  - Malignant
- Each category can be one of two types
  - Transudate
  - Exudate

Biological Nature of Effusions

- Hydrostatic Effusions
  - Due to imbalance of intravascular pressure resulting in increased flow of plasma into body cavities
  - Higher fluid to cell ratio
  - Low protein level
  - Cardiac failure is common cause

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Biological Nature of Effusions

- Infectious Effusions
  - Due to direct effects of organism or as by-product of inflammation
  - Varying types of inflammatory cells can be present
  - Cultures and/or gram and acid fast stains may be useful to identify an organism

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Biological Nature of Effusions

- Noninfectious Inflammatory Effusions
  - Due to autoimmune conditions or reaction to a stimulus
  - Autoimmune conditions include rheumatoid arthritis and systemic lupus
  - Reactive conditions include tissue necrosis or radiation therapy

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Biological Nature of Effusions

- Malignant Effusions
  - Due to a primary tumor (such as mesothelioma) or a metastatic tumor
  - Patient history is extremely helpful in rendering a diagnosis
  - Knowledge of the appearance of the fluid is also helpful (clear, bloody, partially clotted)

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Biological Nature of Effusion

- Transudate
  - Due to physiomechanical factors, i.e. congestive heart failure or cirrhosis
  - Typically clear, pale yellow fluid
  - Low protein content (<3.0 g/dL)
  - Does not clot
  - Low specific gravity (<1.015)
  - Low cellularity
  - Unlikely to contain malignant cells

Biological Nature of Effusion

- Exudate
  - Indicates damage to the serous membrane, i.e. inflammation or tumor
  - Usually cloudy, yellow or bloody
  - High protein content (>3.0 g/dL)
  - Tends to clot
  - High specific gravity (>1.015)
  - High cellularity

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Biological Nature of Effusions

- Additional testing may aide in diagnosis:
  - **Glucose** – Low in TB, rheumatoid disease and bacterial infections
  - **Amylase** – High in pancreatitis, esophageal perforation
  - **pH** – Low in rheumatoid disease, hemothorax, acidosis
  - **Ammonia** –Intestinal necrosis, perforation, effusion(-) vs. urine(+)
  - **Creatinine** – Negative in effusion, positive in urine
  - **Bile** – Included in the differential dx of green appearing effusion
  - **Cell Block and Immunochemistry*–** Tumor typing
  - **Electron Microscopy** – Differentiate adenoca vs. mesothelioma
  - **Flow Cytometry** – Useful in hematopoietic malignancies

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Immunochemistry panels will be discussed in later slides for specific lesions

Specimen Types

- Pleural fluid
- Pericardial fluid
- Peritoneal (Ascites) fluid
- Pelvic/Peritoneal Washing

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Pleural Fluid

- Collected via thoracentesis
  - Removal of pleural fluid from the pleural space with a needle and syringe
  - Procedure can be used as a sampling technique or to alleviate pressure on the lungs
  - Catheter may be used for larger amounts of fluid or to drain recurrent accumulations

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Pleural Fluid

- Sources of benign conditions
  - Primary pulmonary infection or infarction
  - Secondary due to abdominal disease, i.e. cirrhosis
- Sources of malignant conditions
  - Most common sources are lung, breast, lymphoid neoplasms, GI and mesothelioma

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Pericardial Fluid

- Collected via pericardiocentesis
  - Removal of pericardial fluid from the pericardial space using a needle and syringe
  - Procedure can be used as a sampling technique, to relieve pressure on the heart or to administer medication
  - Catheter may be used for larger amounts of fluid or to drain recurrent accumulation
- Benign and malignant processes detected

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Pericardial Fluid

- Sources of benign conditions
  - Congestive heart failure, infection, radiation, hypothyroidism, uremia, hypoproteinemia
- Sources of malignant conditions
  - Commonly associated with lung or breast, followed by lymphoma, sarcoma and melanoma

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Peritoneal Fluid

- Collected via paracentesis
  - Removal of peritoneal fluid using a needle, tubing and a container that may have a vacuum
  - Procedure can be used as a sampling technique as well as a therapeutic technique to alleviate abdominal pressure
  - May need to be repeated periodically with some diseases
- Volume can be as much as 1 gallon or more

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Peritoneal Fluid

- Sources of benign conditions
  - Portal venous hypertension, hypoproteinemia and salt retention
- Sources of malignant conditions
  - Commonly associated with ovary, breast, GI, lymphoid neoplasms and mesothelioma
  - In malignancies of unknown origin, consider genital tract for women and GI tract for men

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Pelvic/Peritoneal Washing

- Collected intra-operatively
  - Surgeon irrigates the peritoneal cavity with sterile fluid that is later recollected and evaluated
- Routinely performed to stage abdominal and pelvic tumors

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Normal Components and Findings

- Mesothelial cells in effusions
  - Nuclei
    - Single or binucleated
    - Centrally located but can be eccentric
    - Round to oval with well-defined, smooth nuclear borders
    - Fine chromatin
    - Inconspicuous nucleoli
  - Cytoplasm
    - Dense center with pale periphery
    - Lacy “skirt” cell borders
    - Blunt cytoplasmic processes due to degeneration

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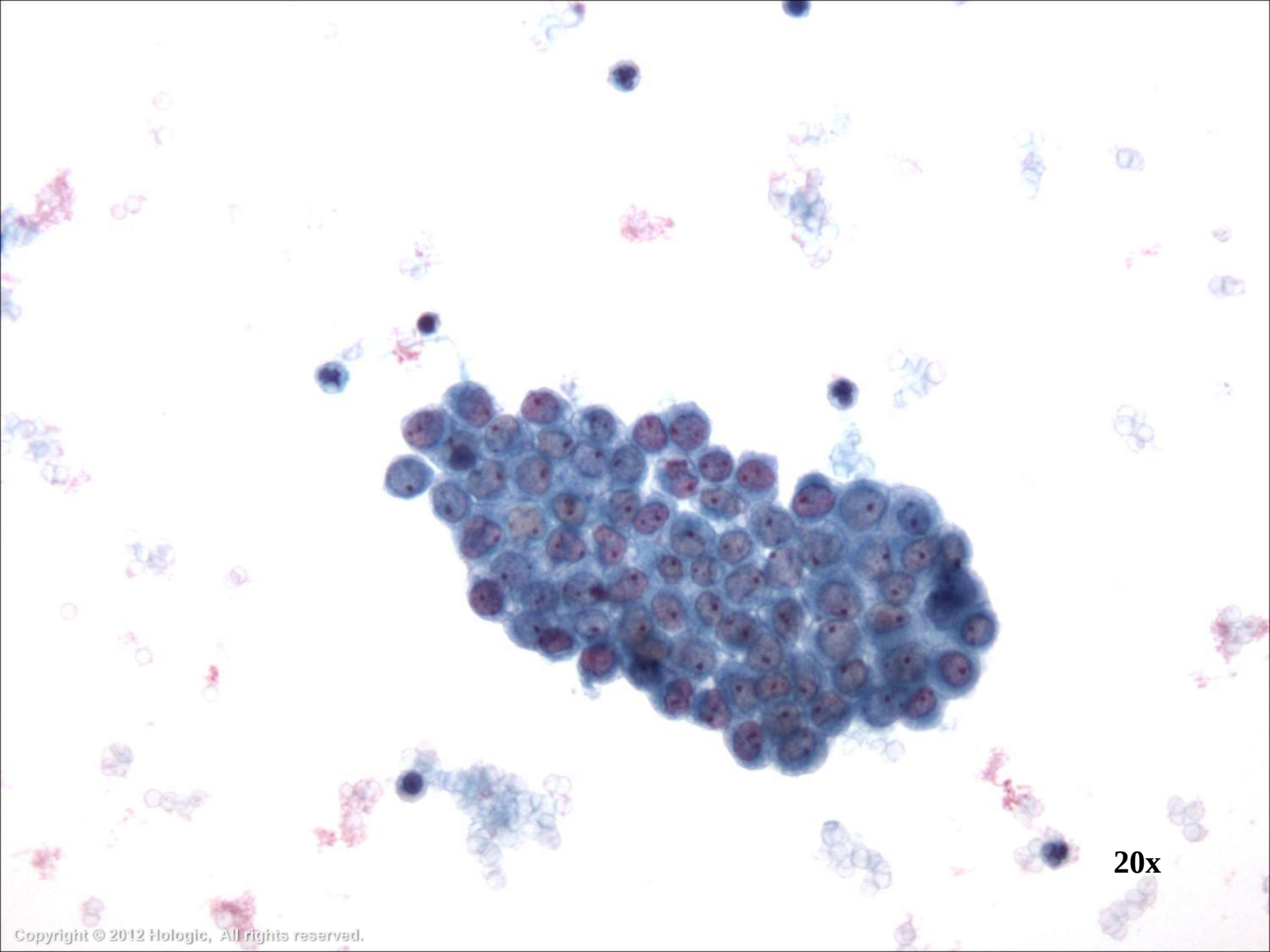

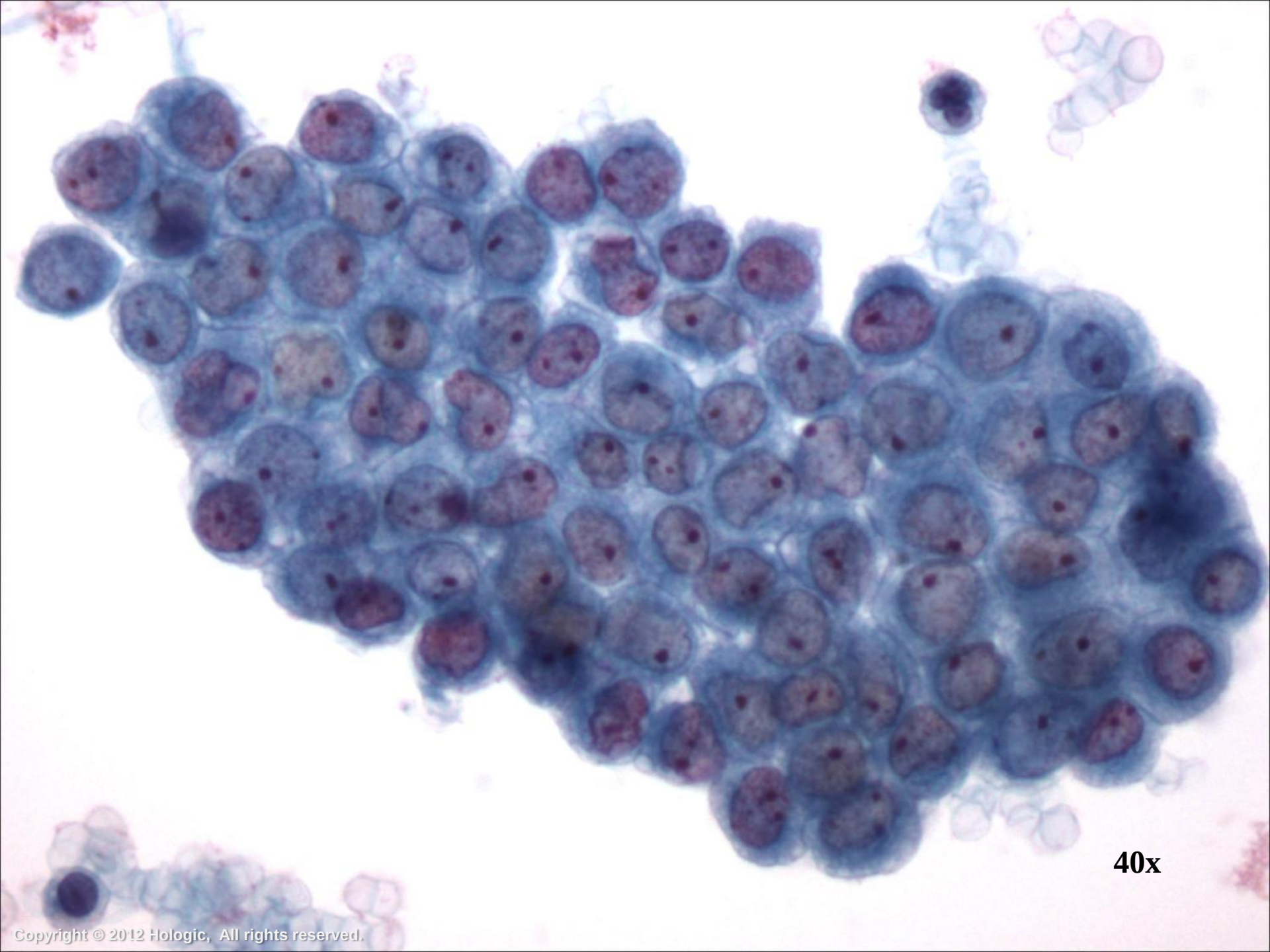

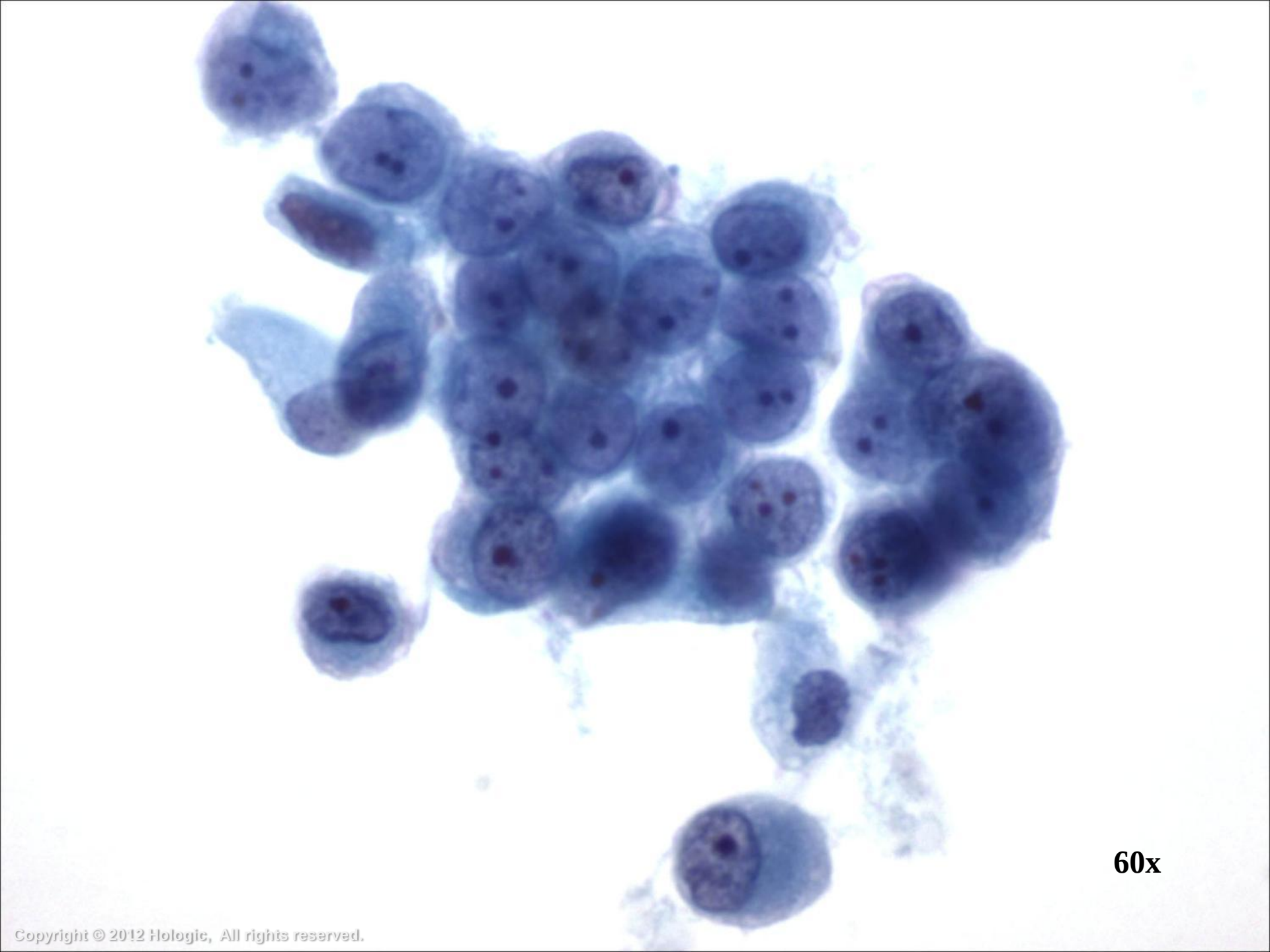

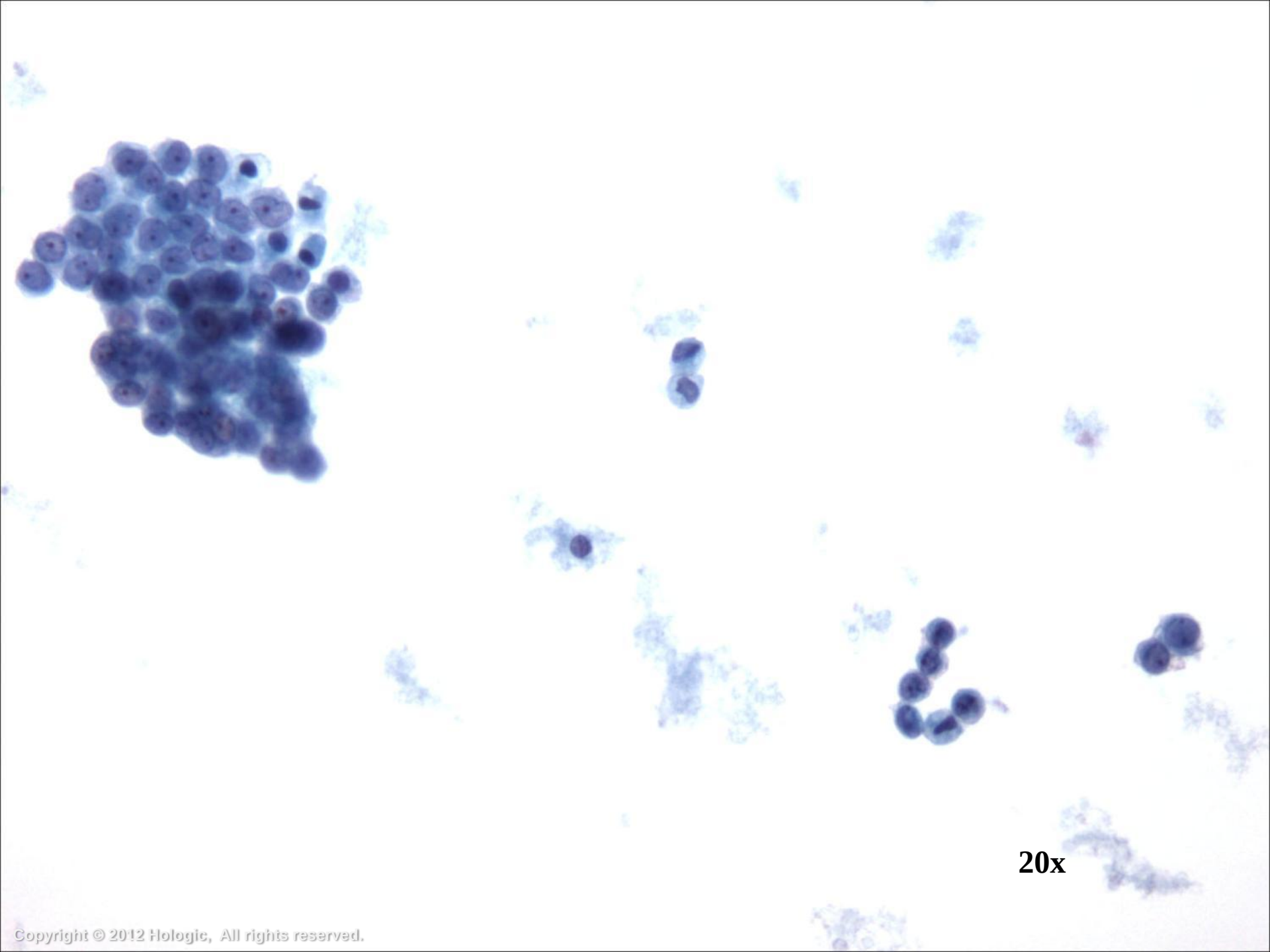

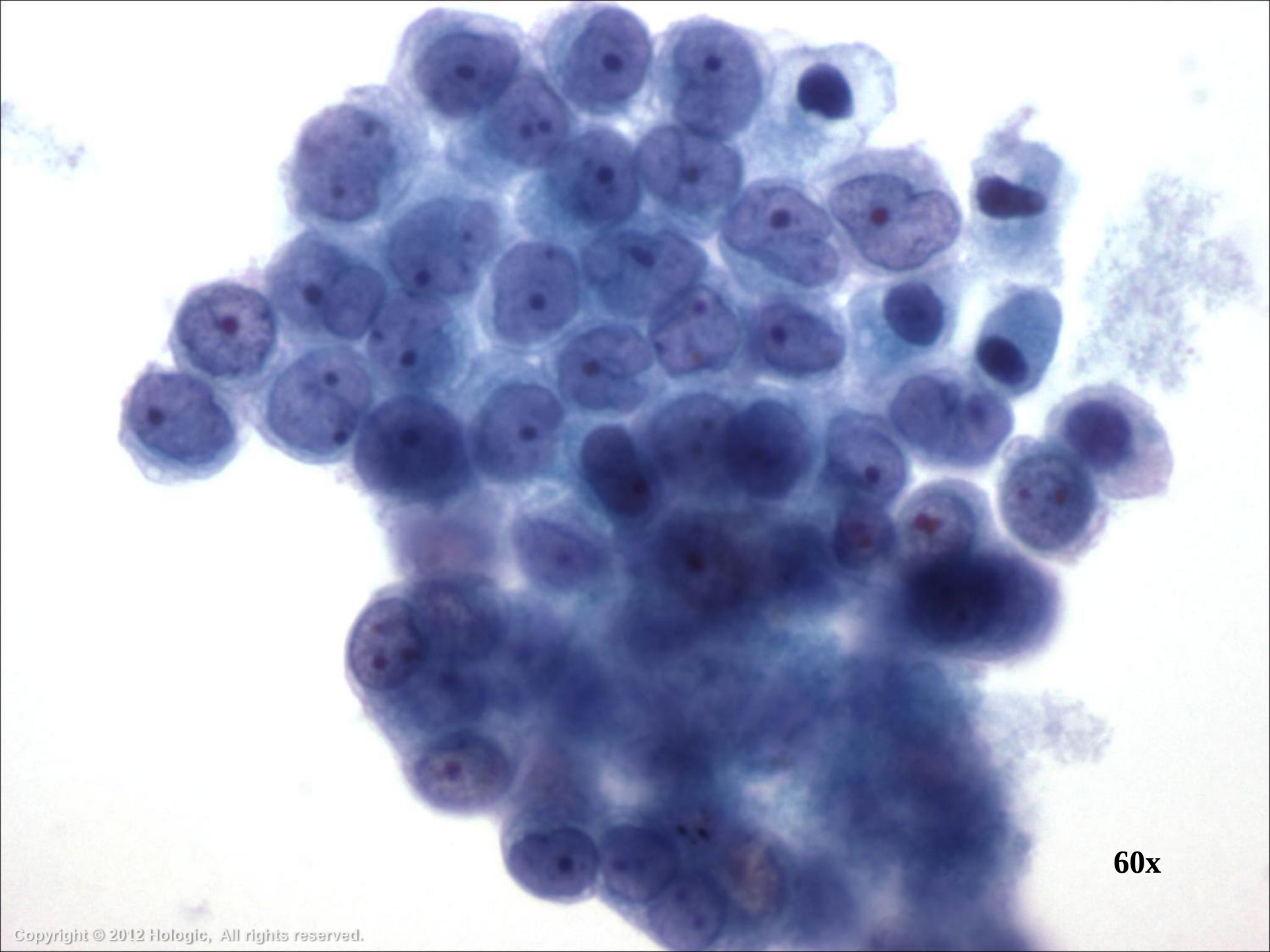

Normal Components

- Mesothelial cells in peritoneal washings
  - Broad and flat sheets
  - Slightly rounded up
  - Cuboidal

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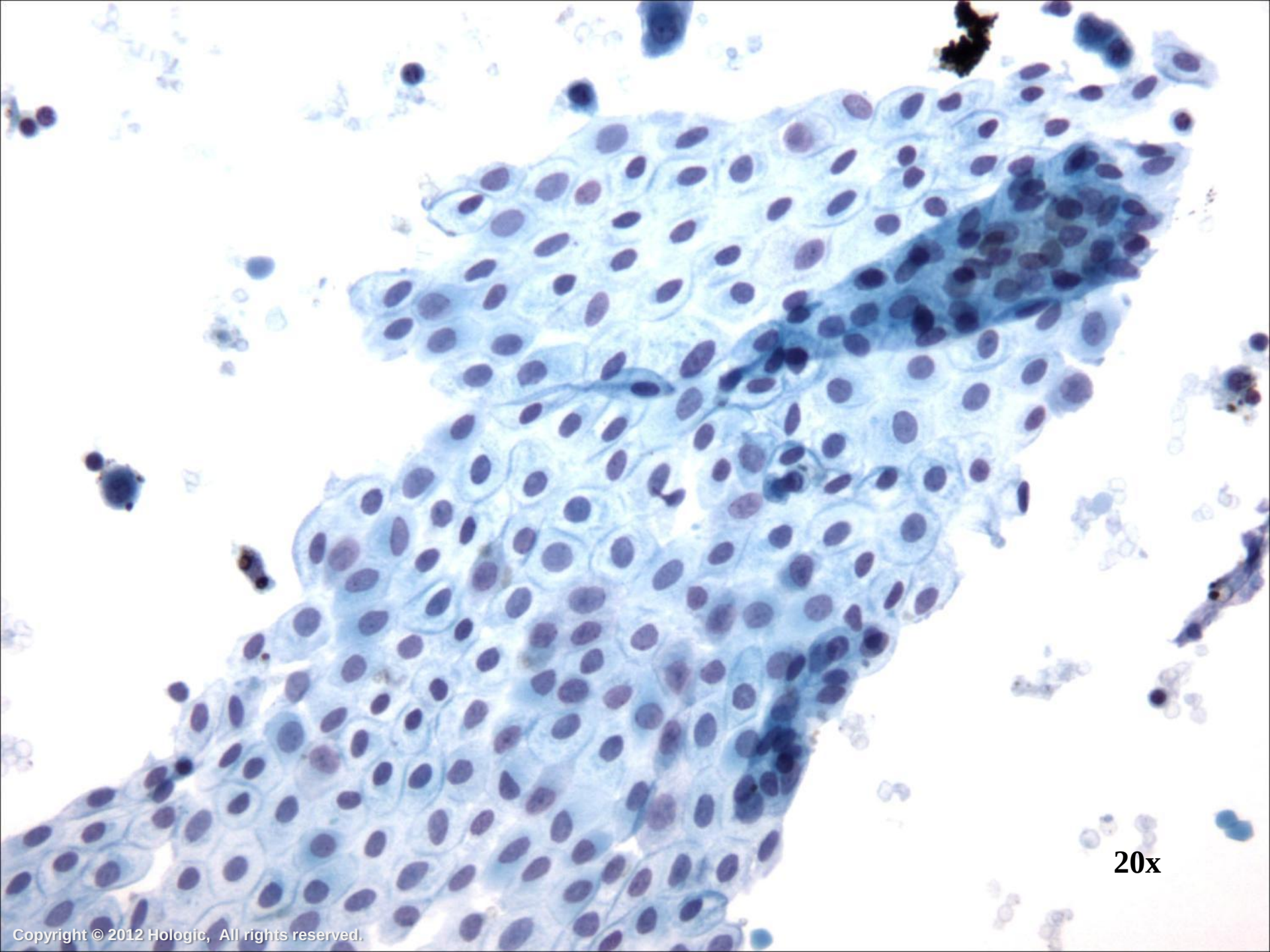

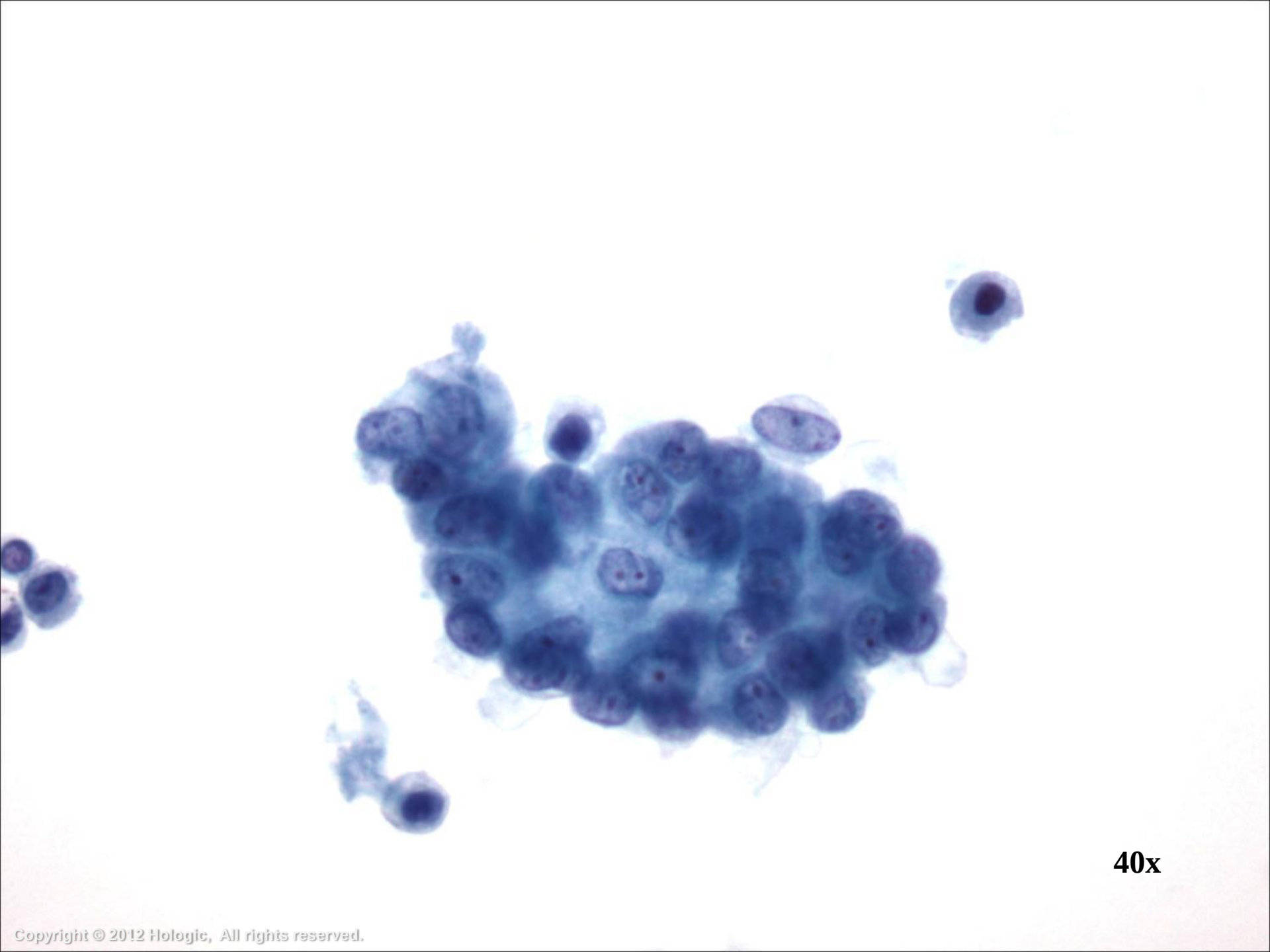

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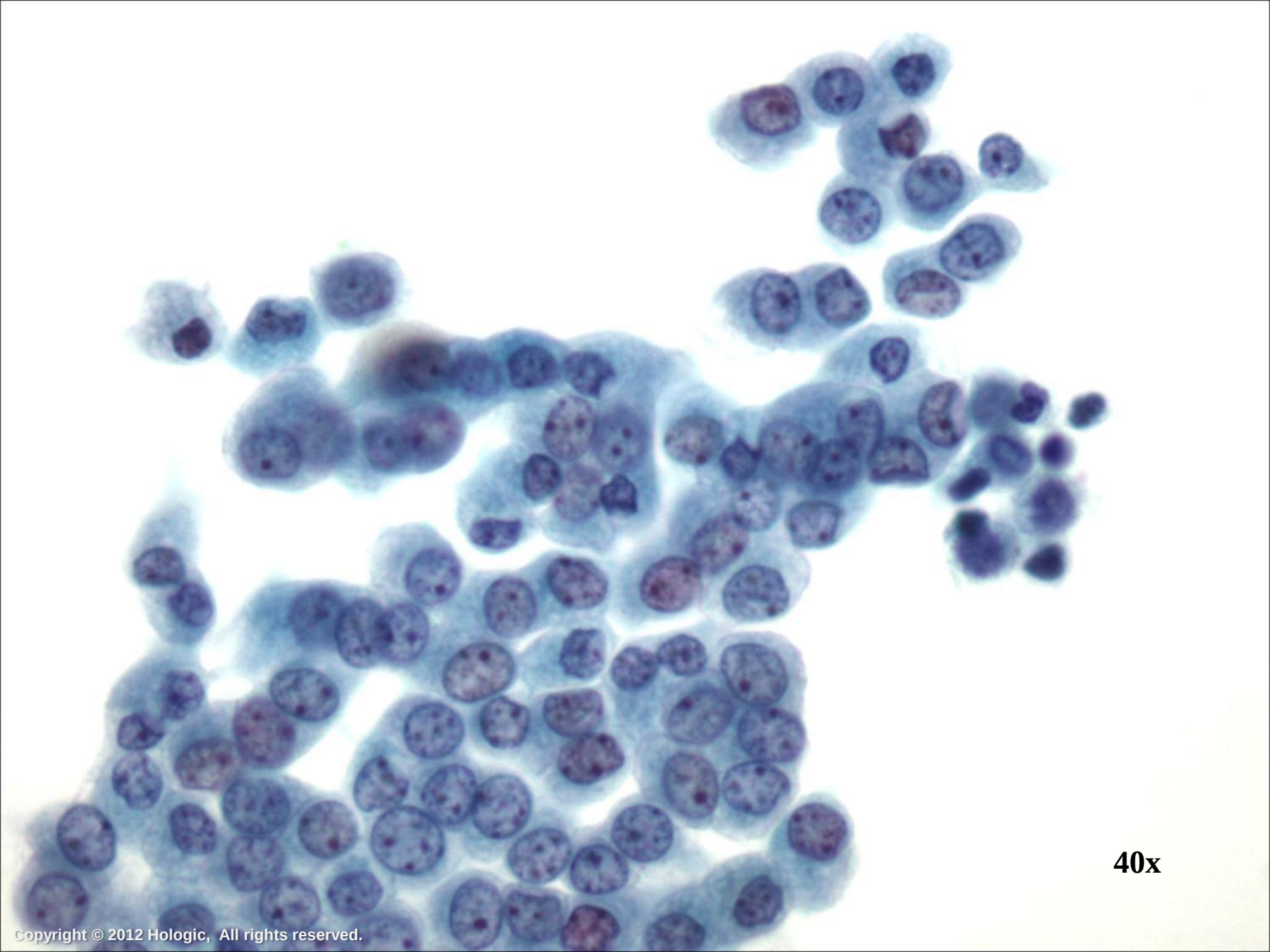
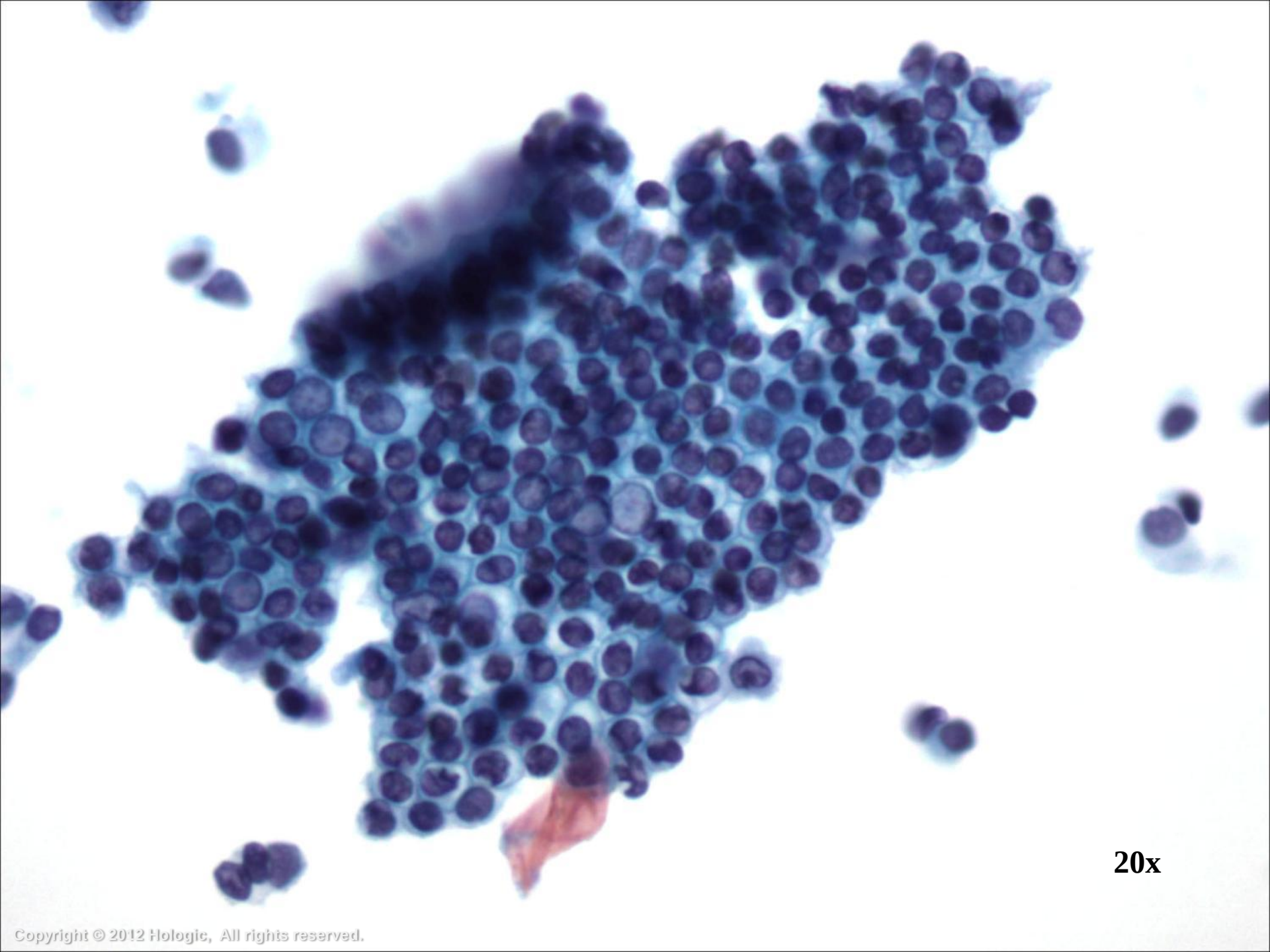
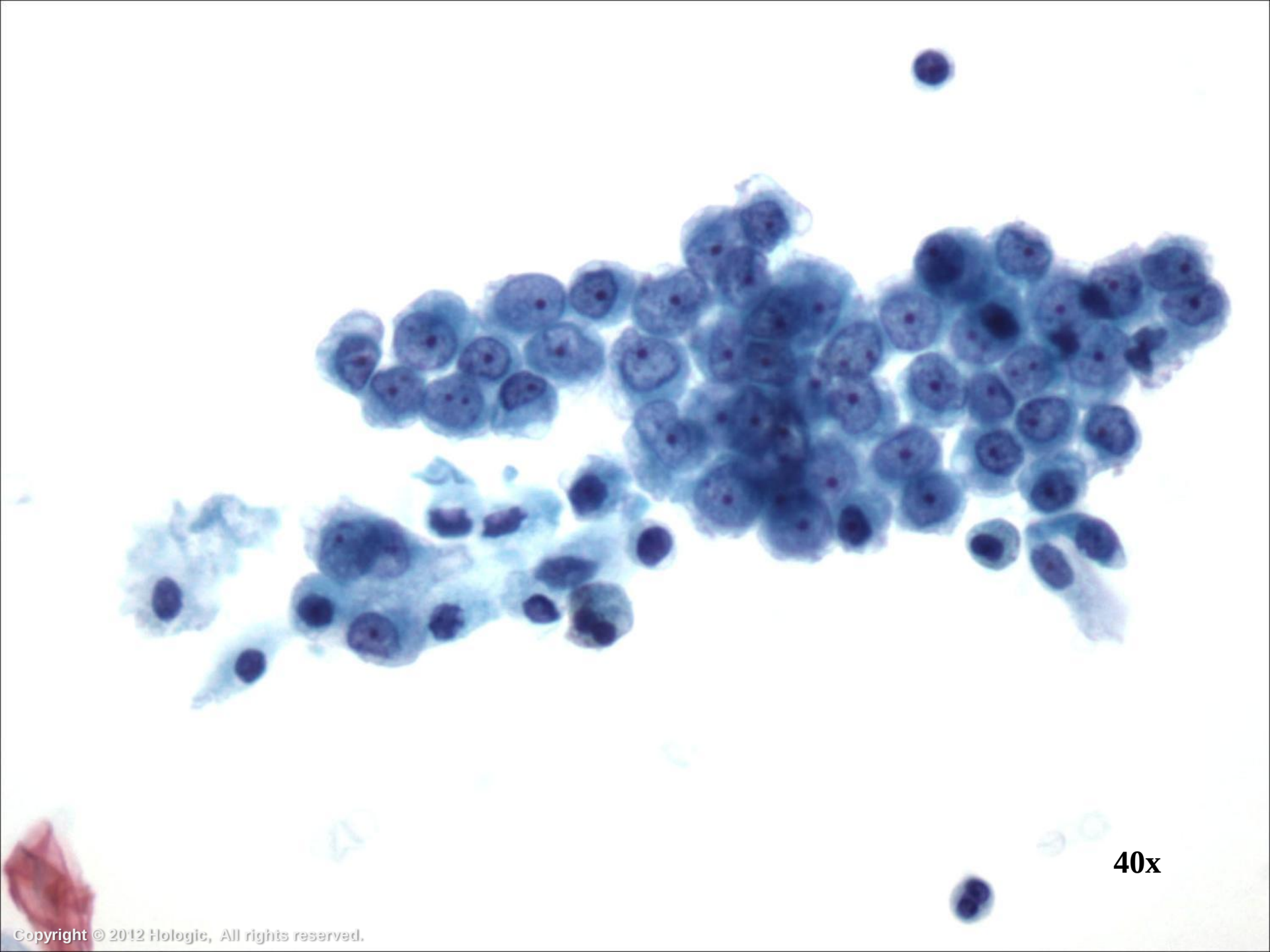
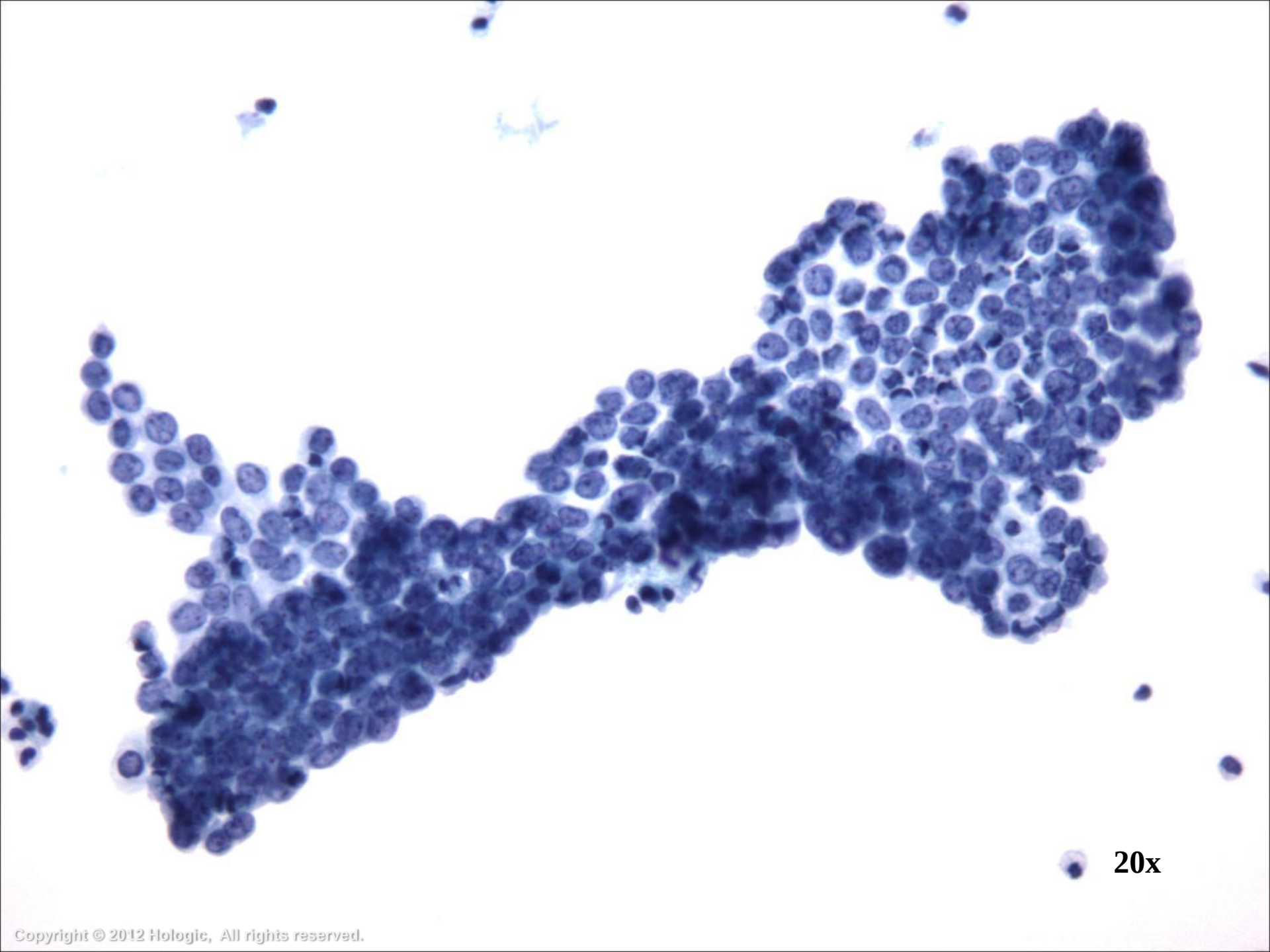
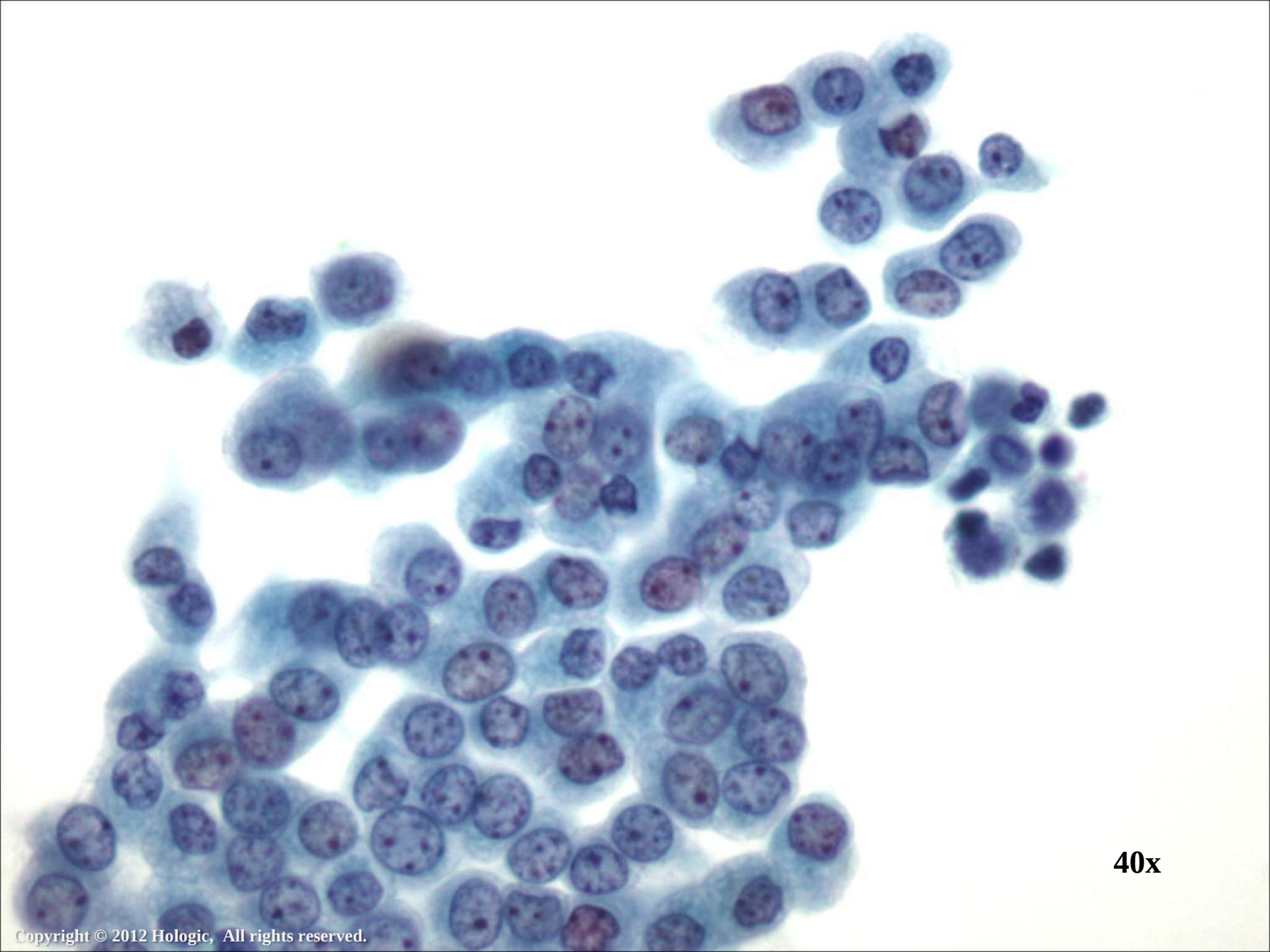




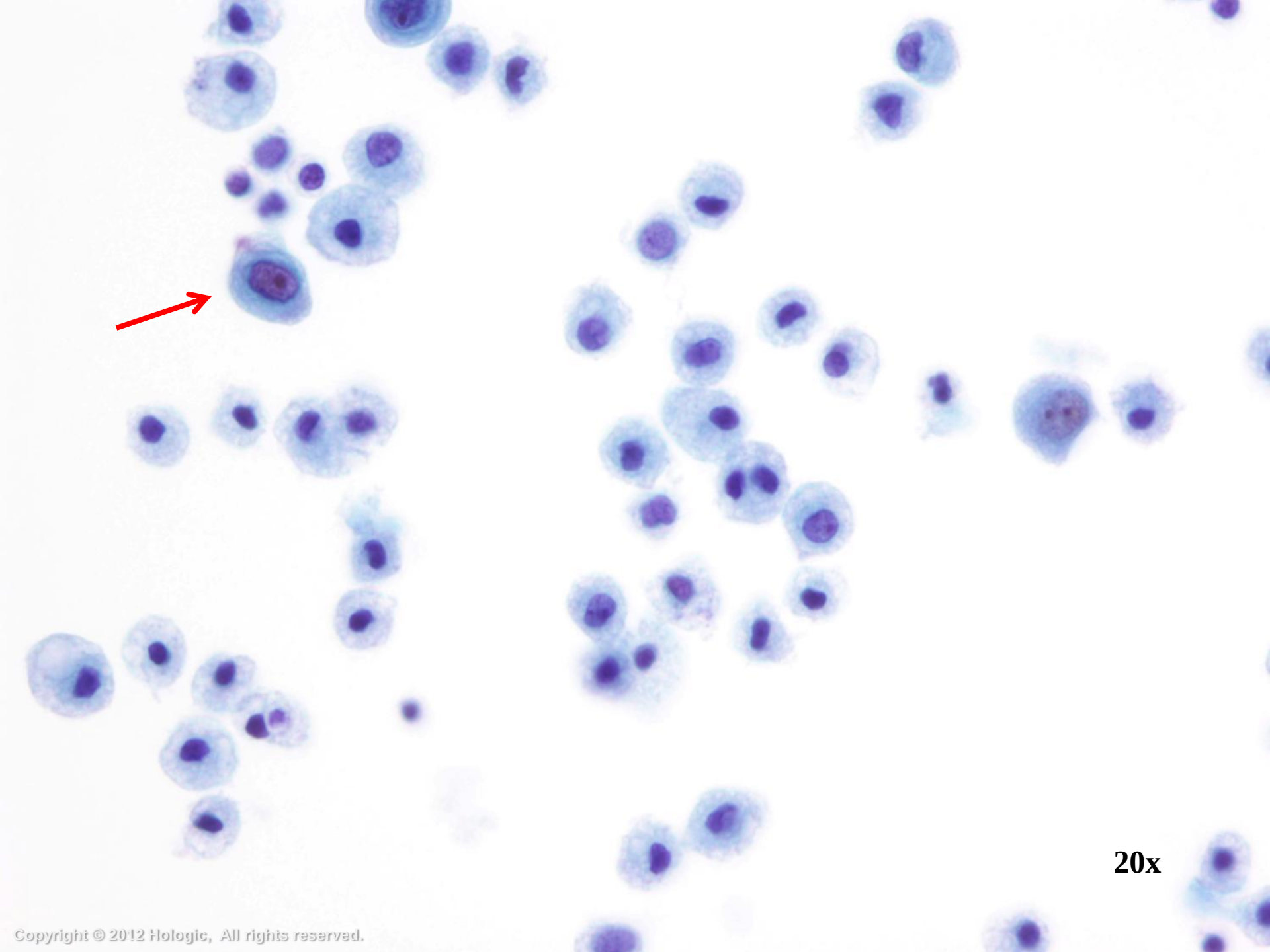


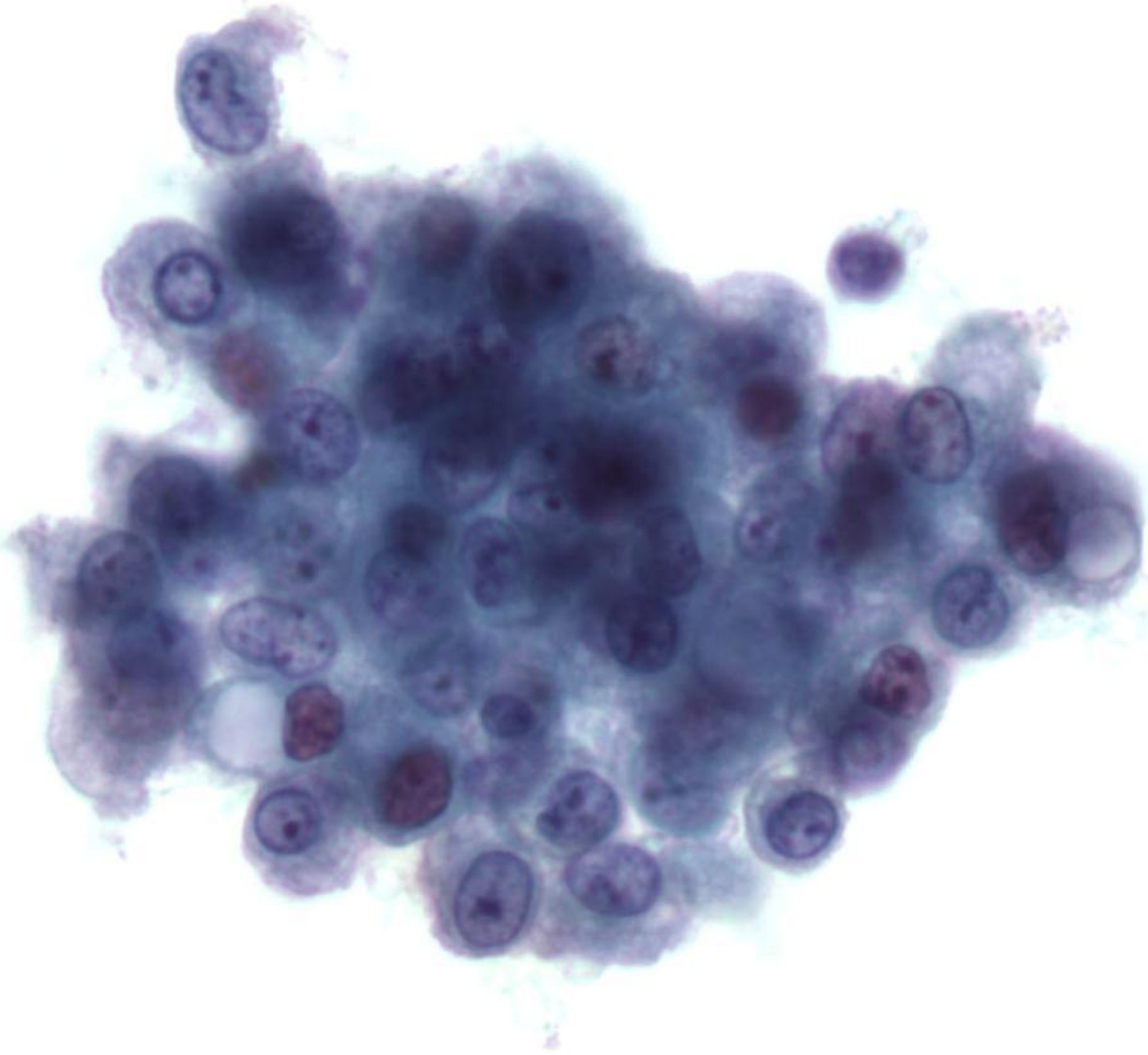




Normal Components

- Histiocytes
  - Usually present in effusion. Prominent in cancer, TB and embolism
- Lymphocytes
  - Some amount usually present. Can be numerous in conditions ranging from congestive heart failure and infections to carcinoma and lymphoma
- Eosinophils
  - Usually a good prognosis. Most eosinophilic effusions are pleural and due to allergic reactions to dust. Other causes include pulmonary infarct, pneumonia, trauma, hydatid disease
- Neutrophils
  - Presence can have many causes, particularly infection. Malignant effusions are seldom associated with acute inflammation





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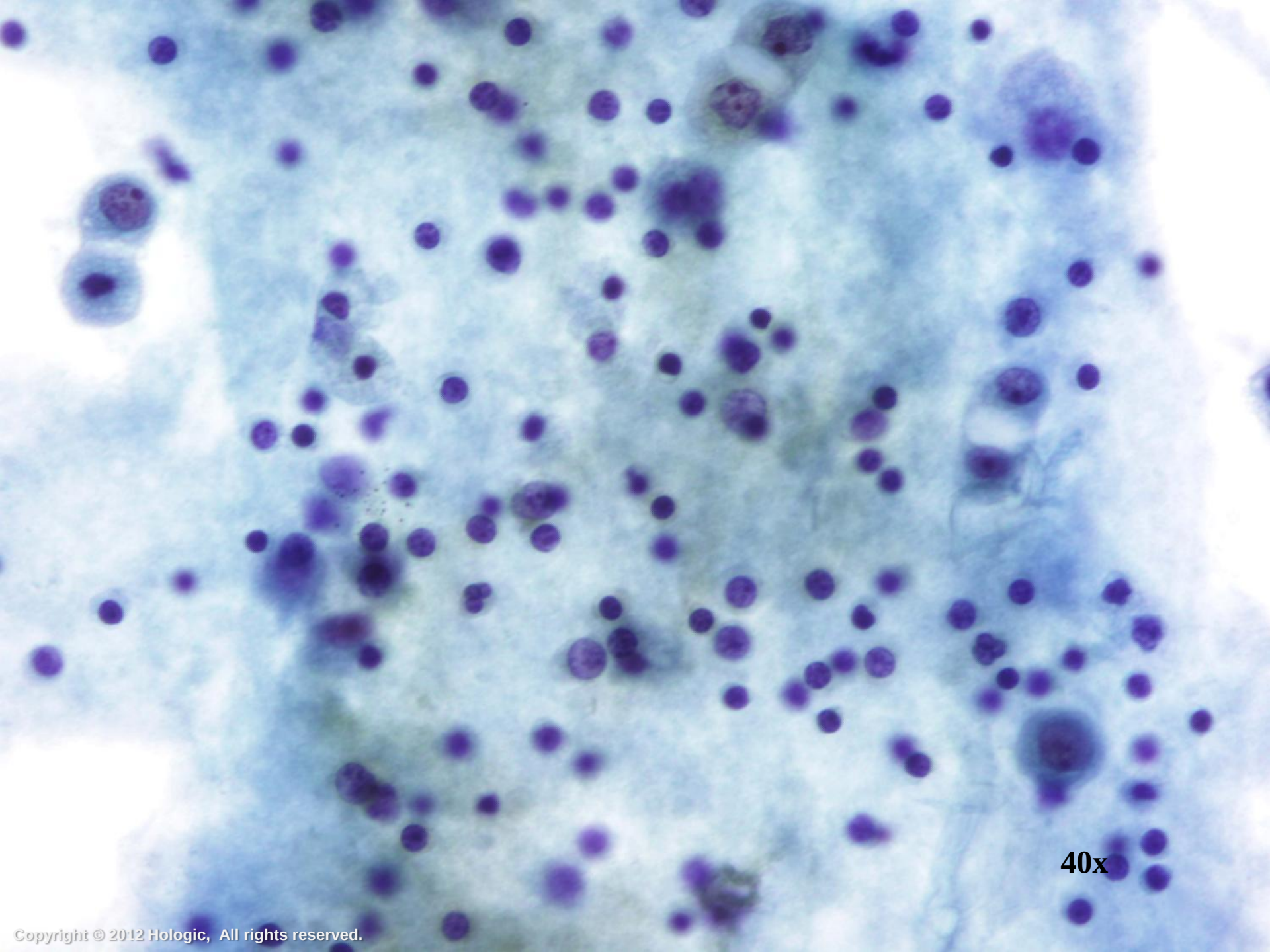

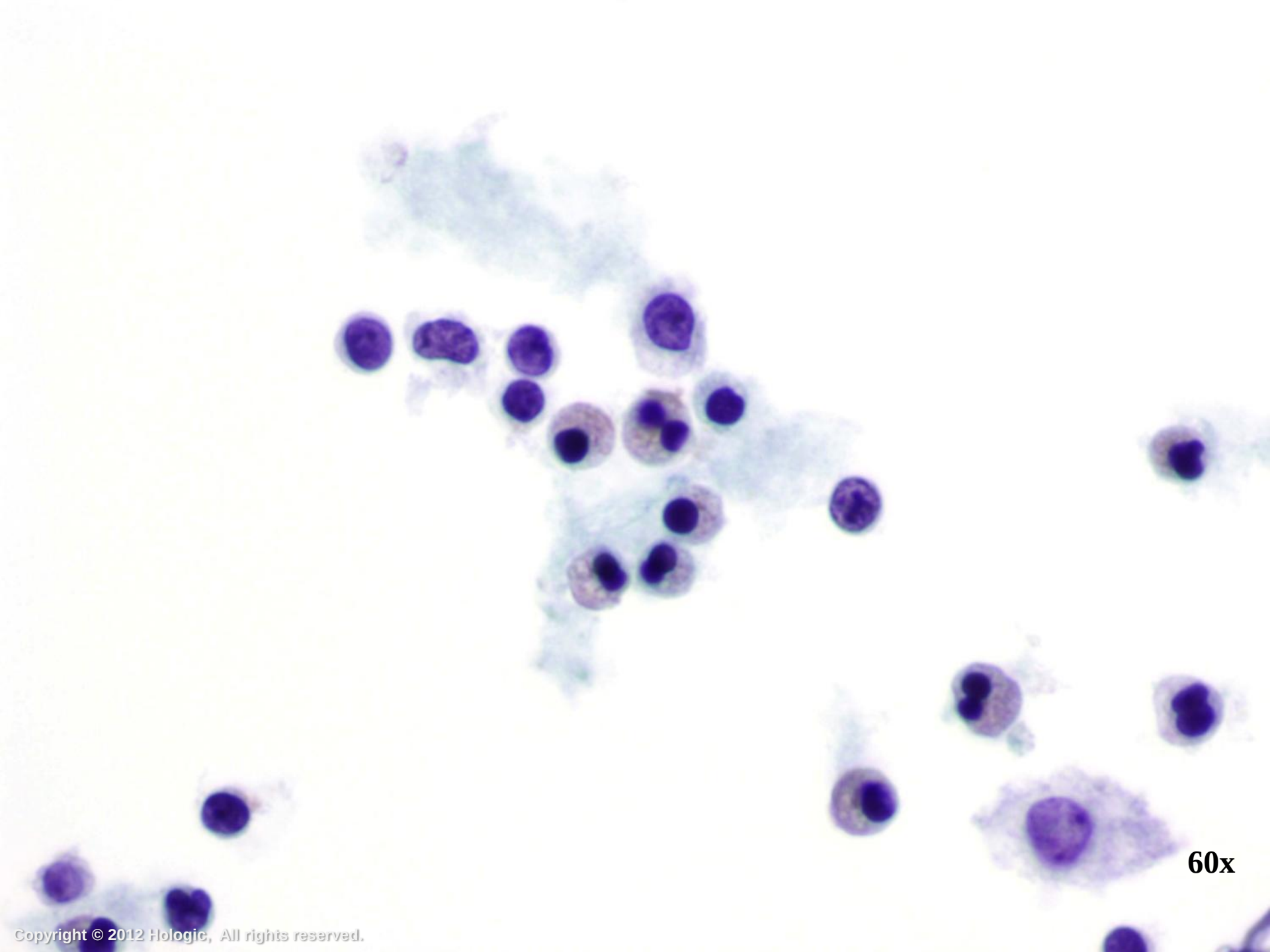

Normal Components

- Microorganisms
- LE cells
- Sickle cells
- Charcot-Leyden crystals
- Psammoma bodies
- Collagen balls

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LE Cells

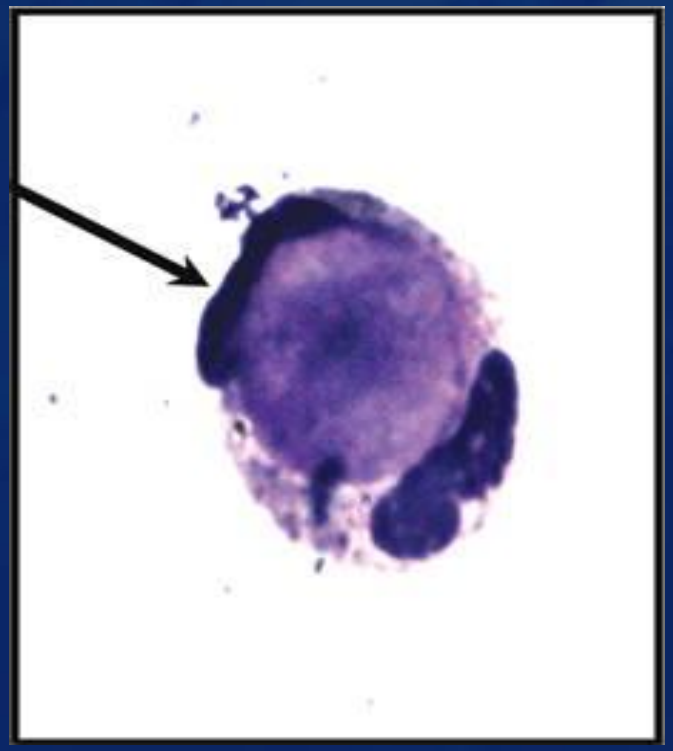

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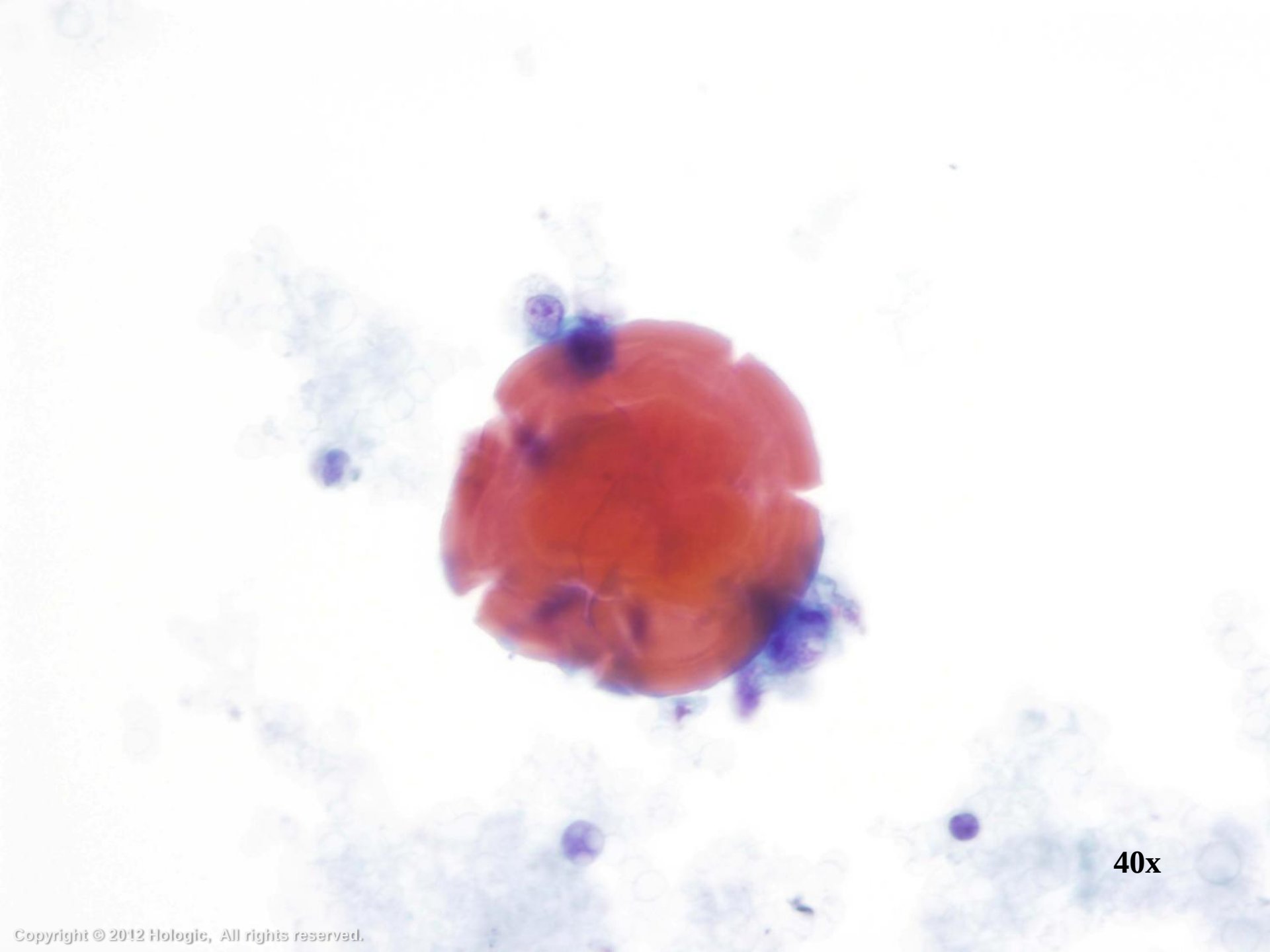
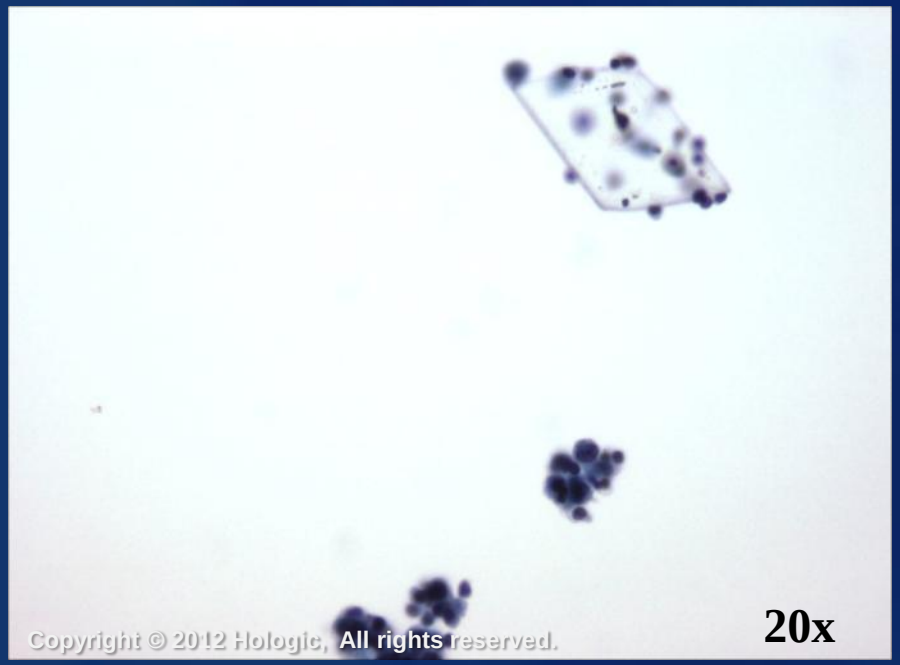
Image reprinted with permission from MLO-Medical Laboratory Observer, November 2008.



Non-cellular Components

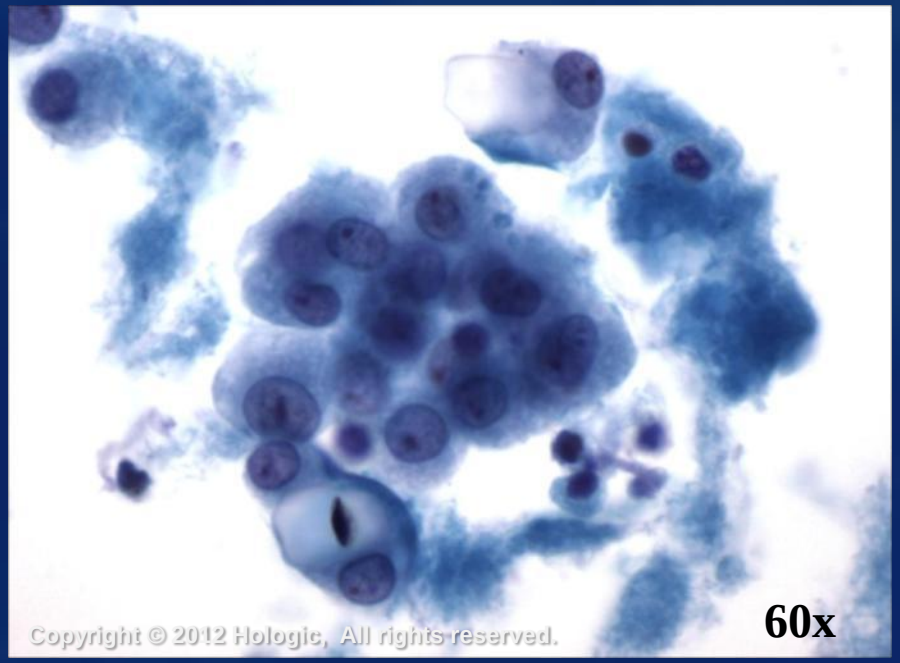
Cholesterol crystals

Peritoneal wash



Starch granules

Peritoneal wash



Reactive changes

- Reactive changes are common and may be due to:
  - Pulmonary embolism or infarct
  - Active cirrhosis or hepatitis
  - Uremia
  - Pancreatitis
  - Long-term dialysis
  - Radiation and chemotherapy

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Reactive changes

- Features of reactive mesothelial cells may include:
  - Varying cell sizes
  - Increased numbers of mesothelial cells
  - Enlarged central or paracentral nuclei
  - Binucleation or multinucleation
  - Coarsening of chromatin that remains evenly distributed
  - Chromocenters and nucleoli may become prominent
  - “Sibling images” are a clue

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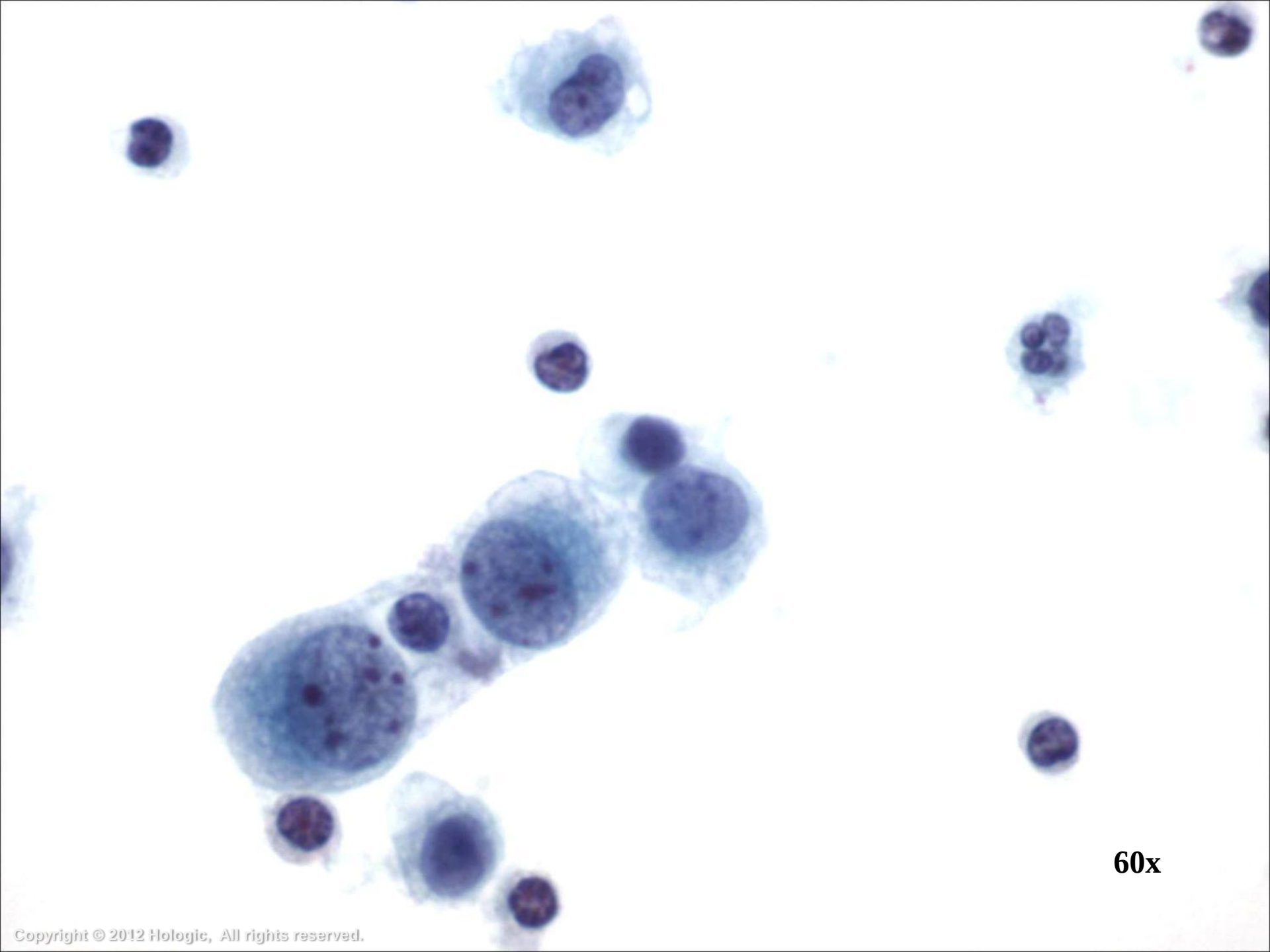

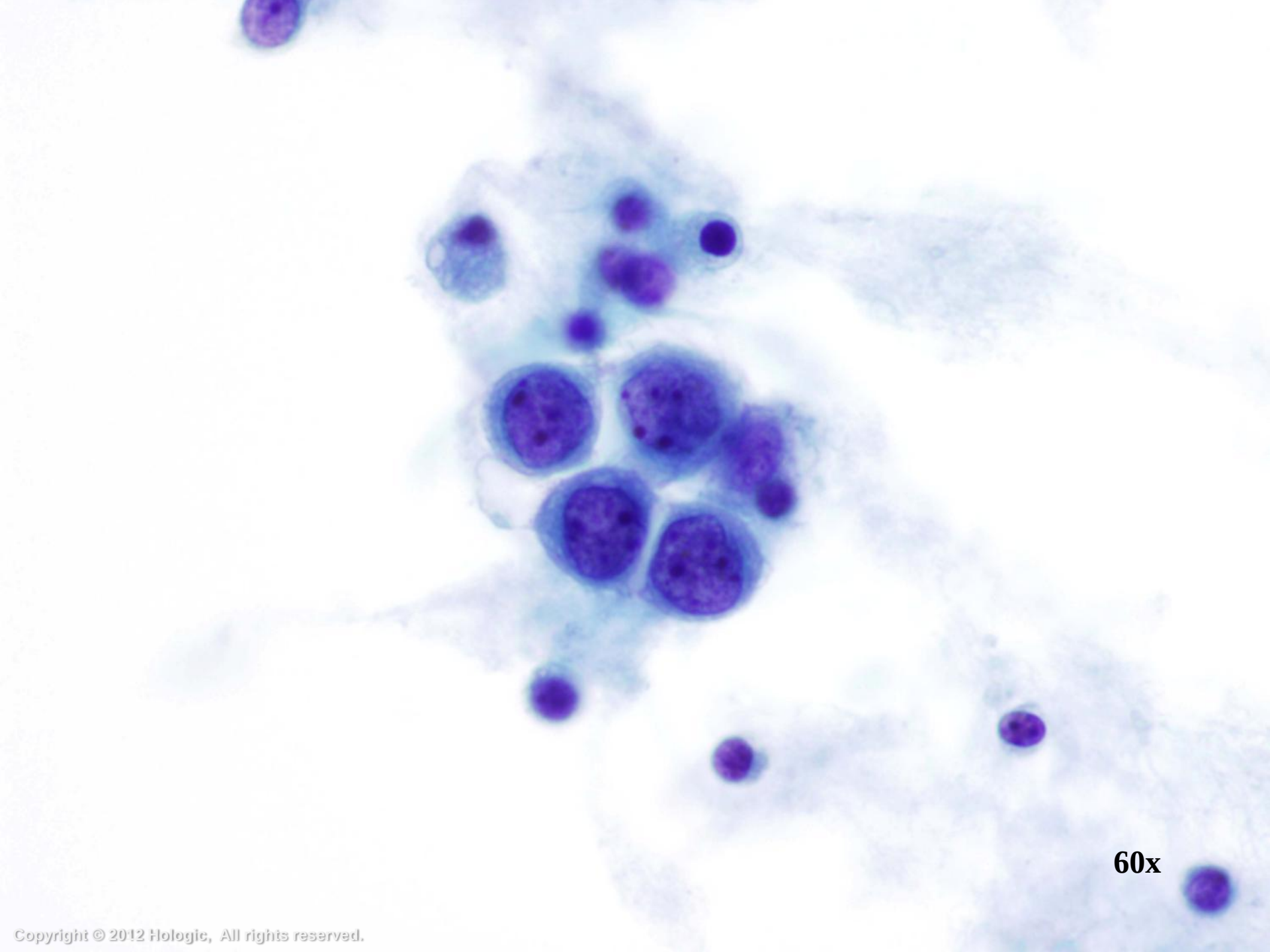

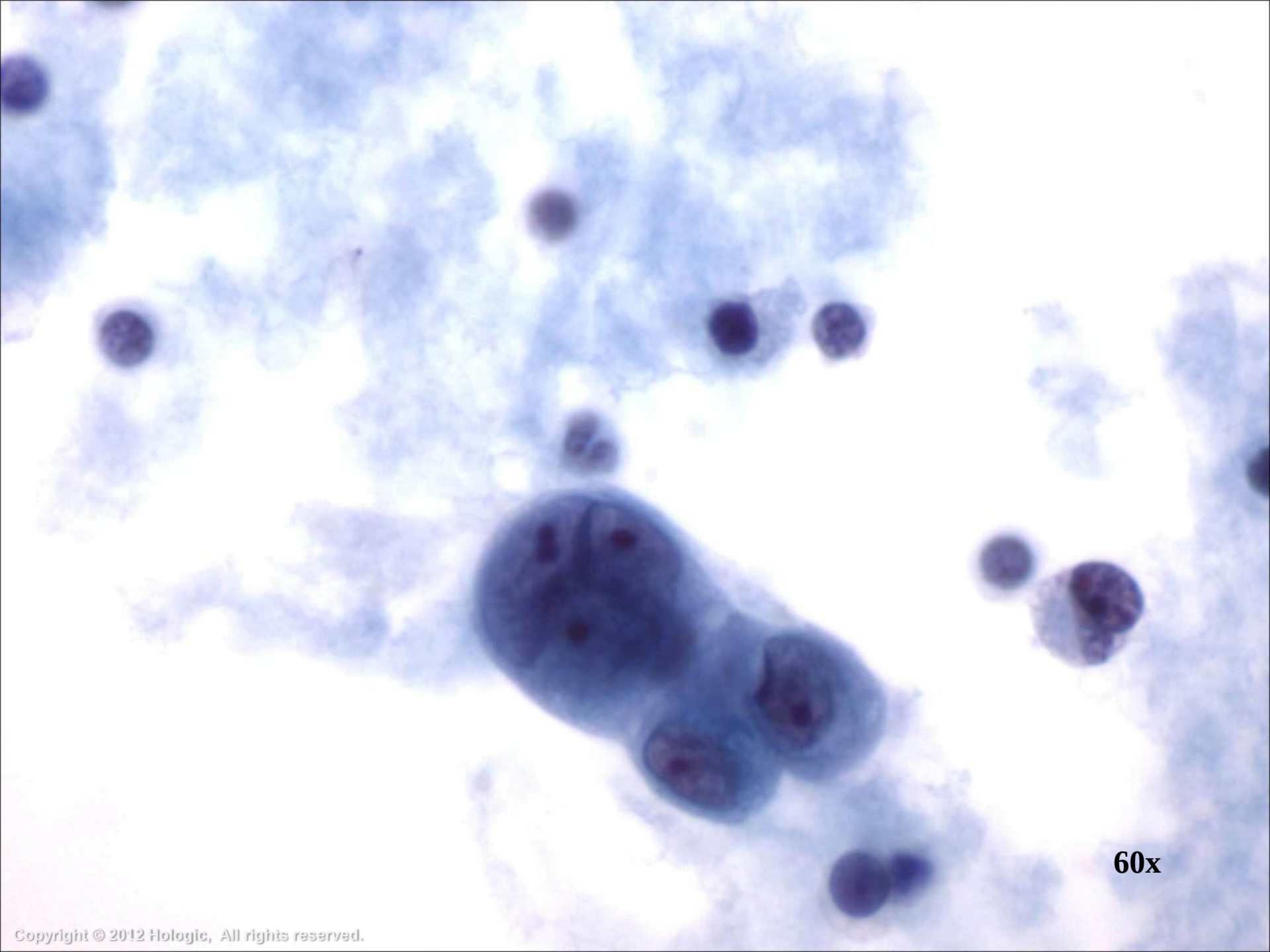

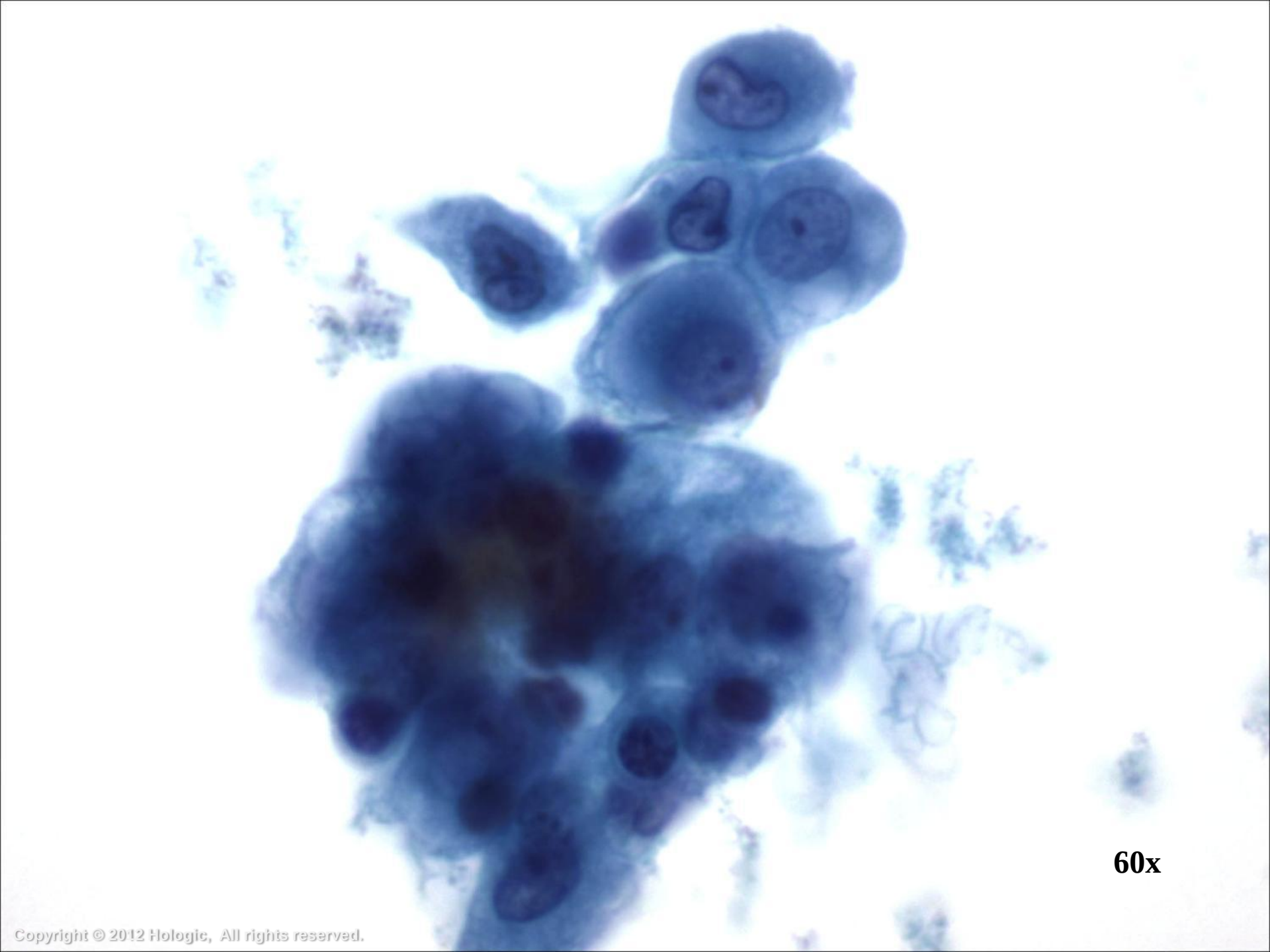

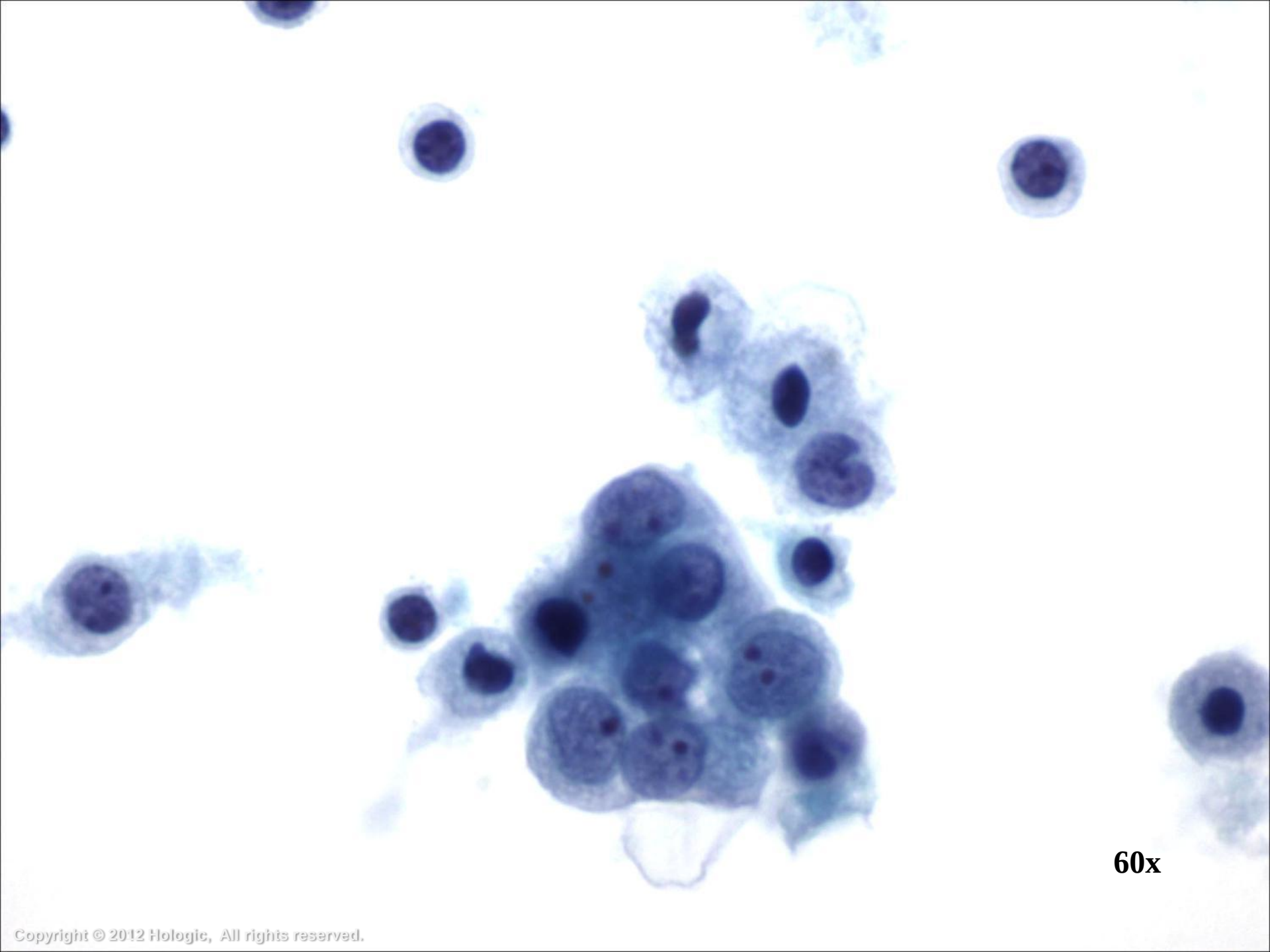

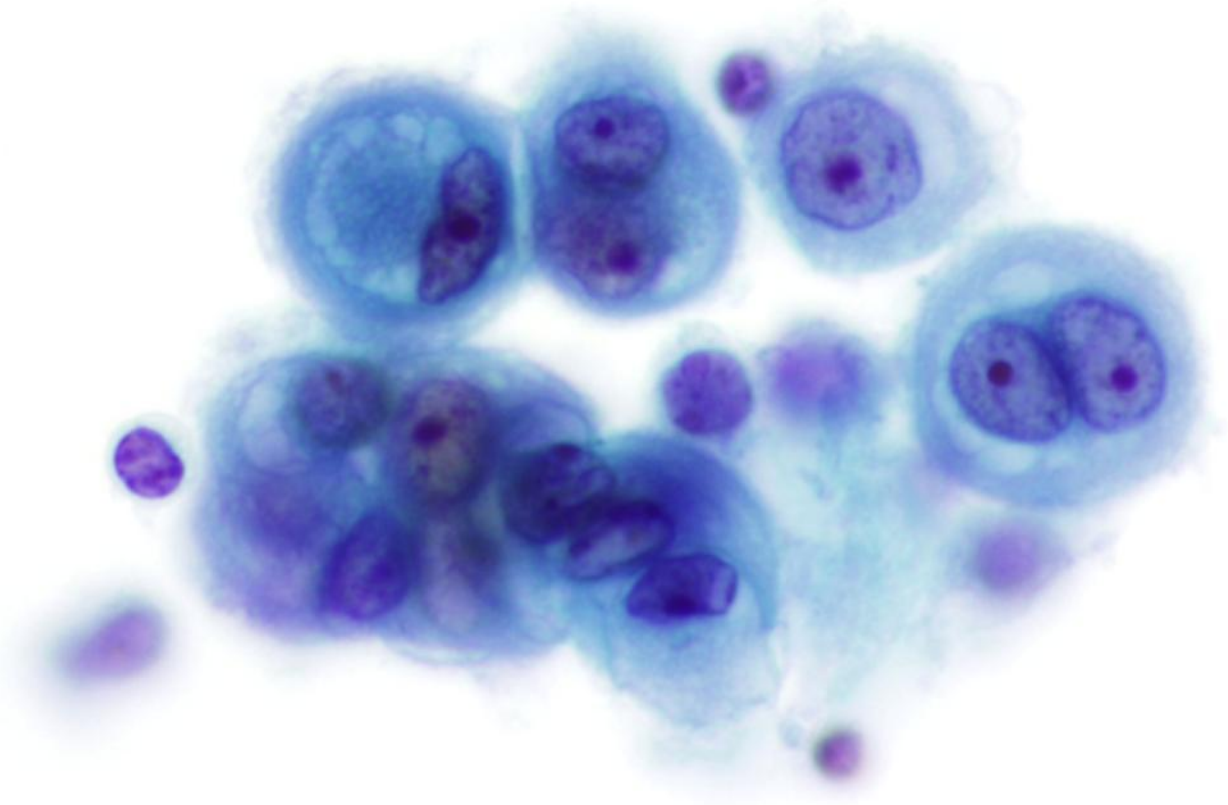

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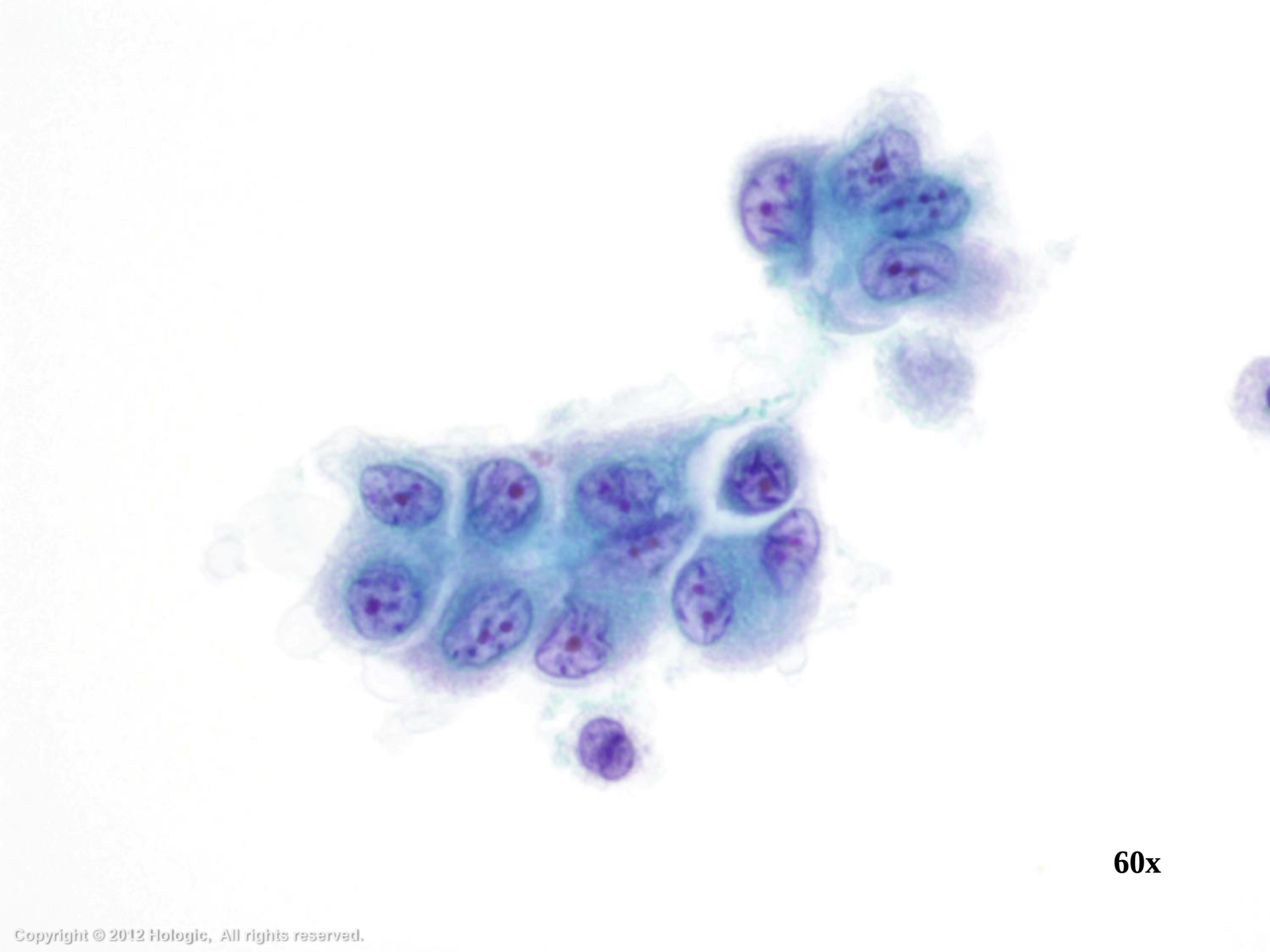

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Suggested Immunochemistry Markers

Reactive mesothelial cells

- **Calretinin** +
- CK 5/6 +
- p53 -
- Desmin +
- EMA -

Mesothelioma

- **Calretinin** +
- CK 5/6 +
- p53 +
- Desmin -
- EMA +

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Note – Expected staining results; observed in most but not all cases.

Abnormal Findings

Tumors causing malignant effusions:

- Primary:
  - Malignant Mesothelioma
  - Effusion Lymphoma
- Metastatic:
  - Adenocarcinoma
  - Squamous Cell Carcinoma
  - Neuroendocrine Tumors
  - Lymphoma/leukemia
  - Melanoma
  - Sarcoma
  - Other Neoplasms

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Malignant Mesothelioma

- Very rare, usually associated with asbestos exposure
- Most cases occur in men aged 50 to 70 years, particularly with a background in auto mechanics or construction
- Commonly found in the pleural cavity but can occur in the peritoneal cavity as well

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Malignant Mesothelioma

- Cytologic features:
  - One cell population of malignant mesothelial cells
  - Larger clusters with irregular, knobby, flower-like borders
  - Dense endoplasm with delicate, lacy ectoplasm
  - “Windows”, “skirts” or blebs of cytoplasm
  - Central enlarged nucleus with macronucleoli and irregular borders
  - Abnormal mitotic figures may be seen but are not diagnostic of mesothelioma

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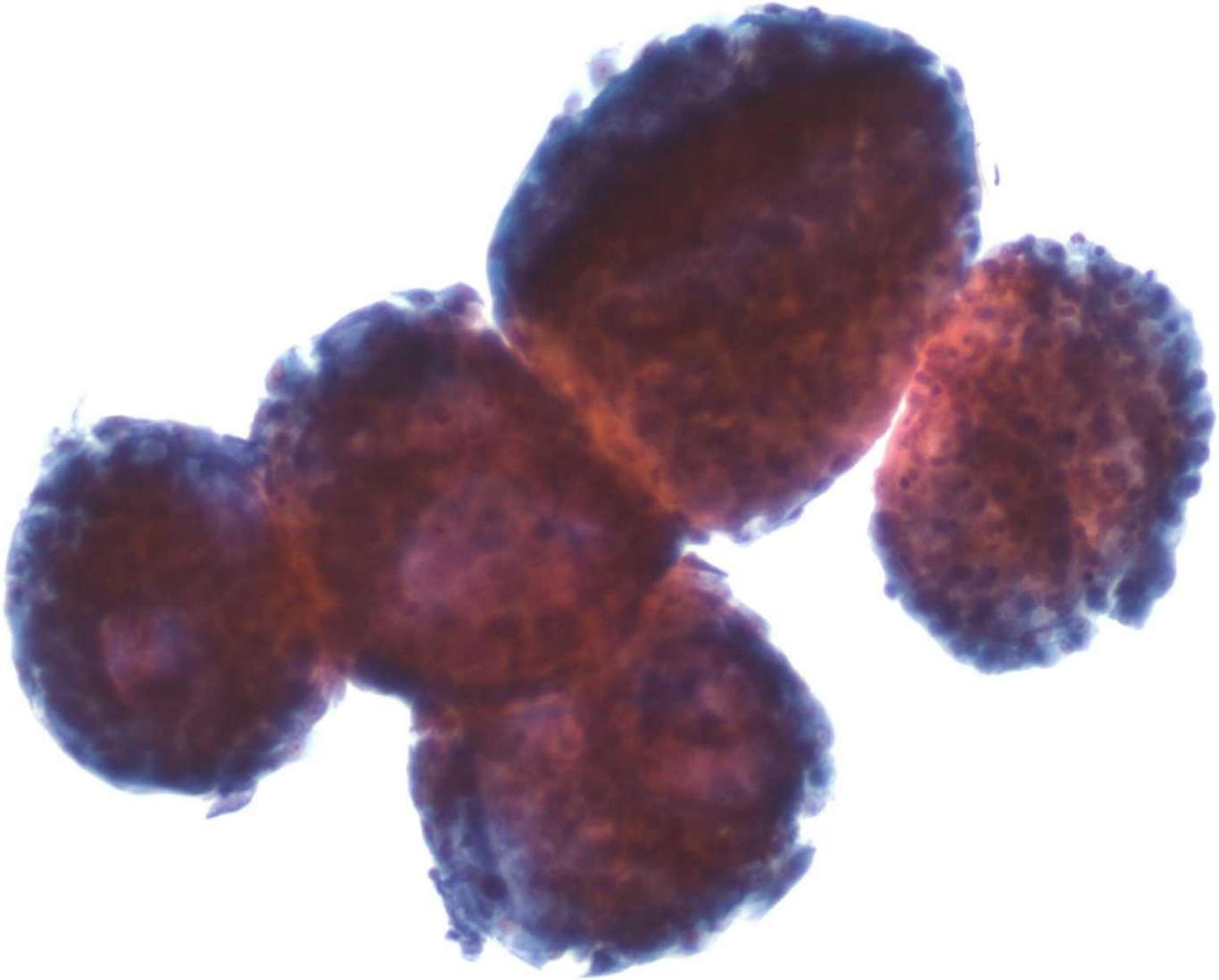

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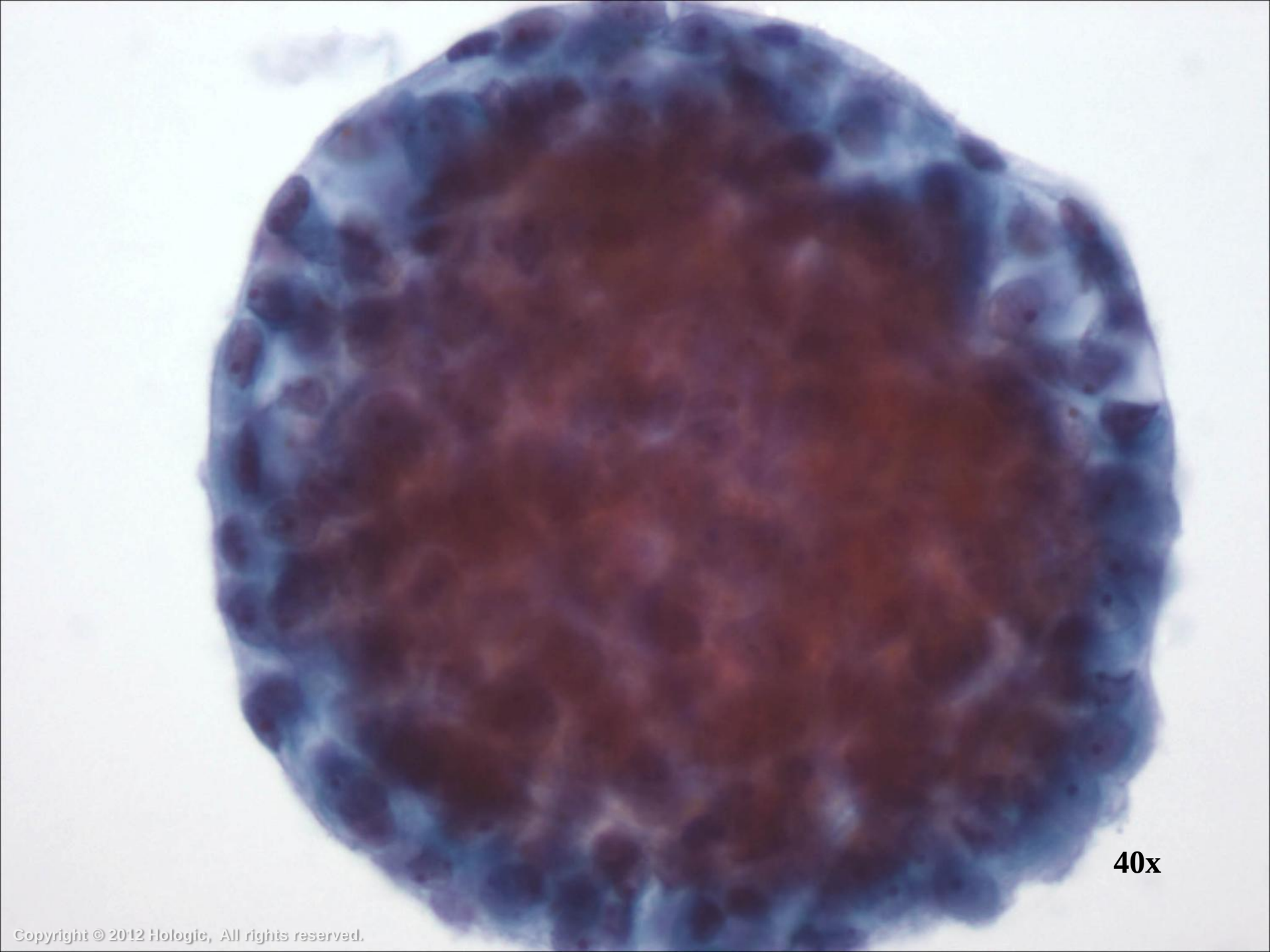

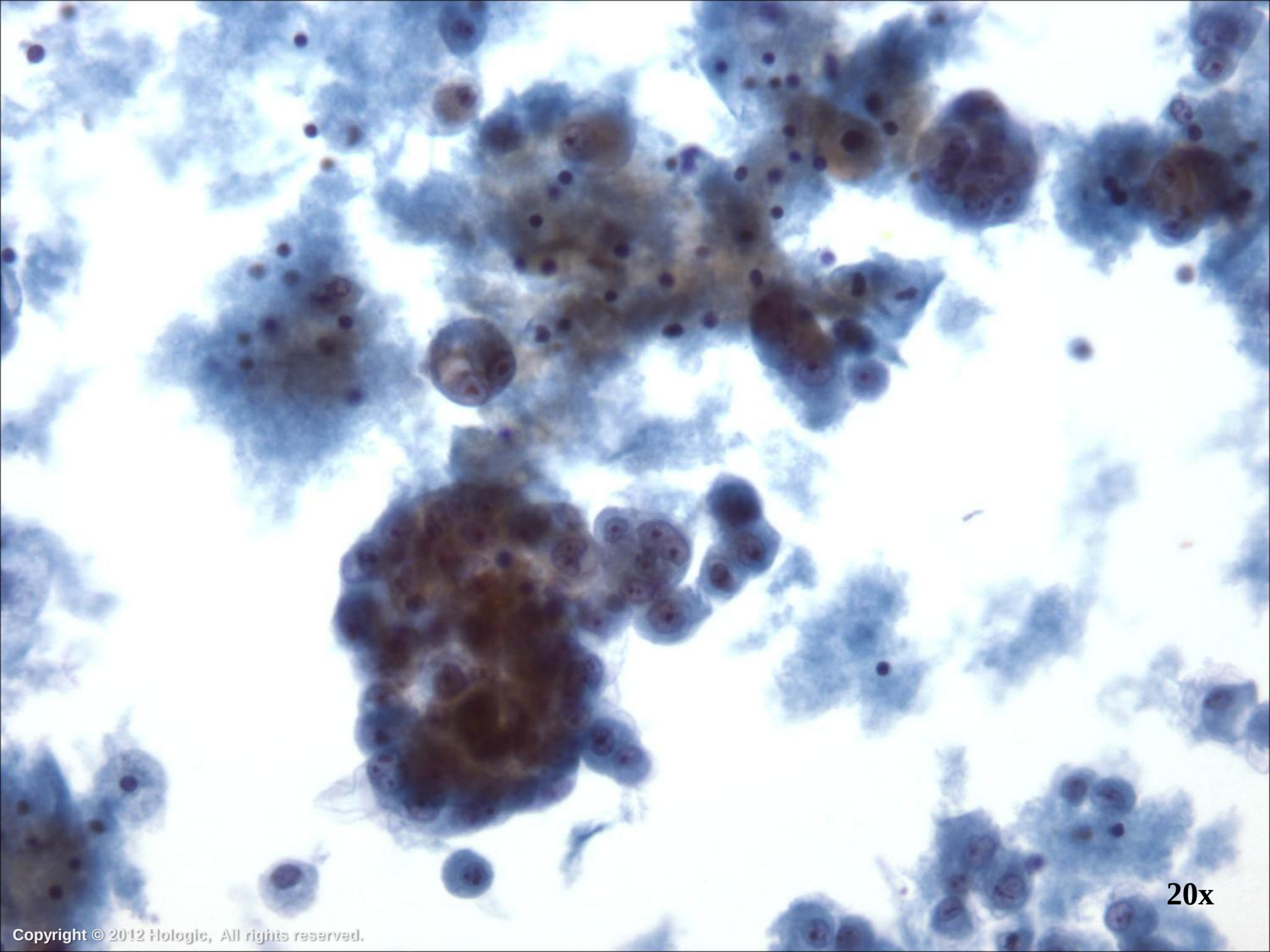

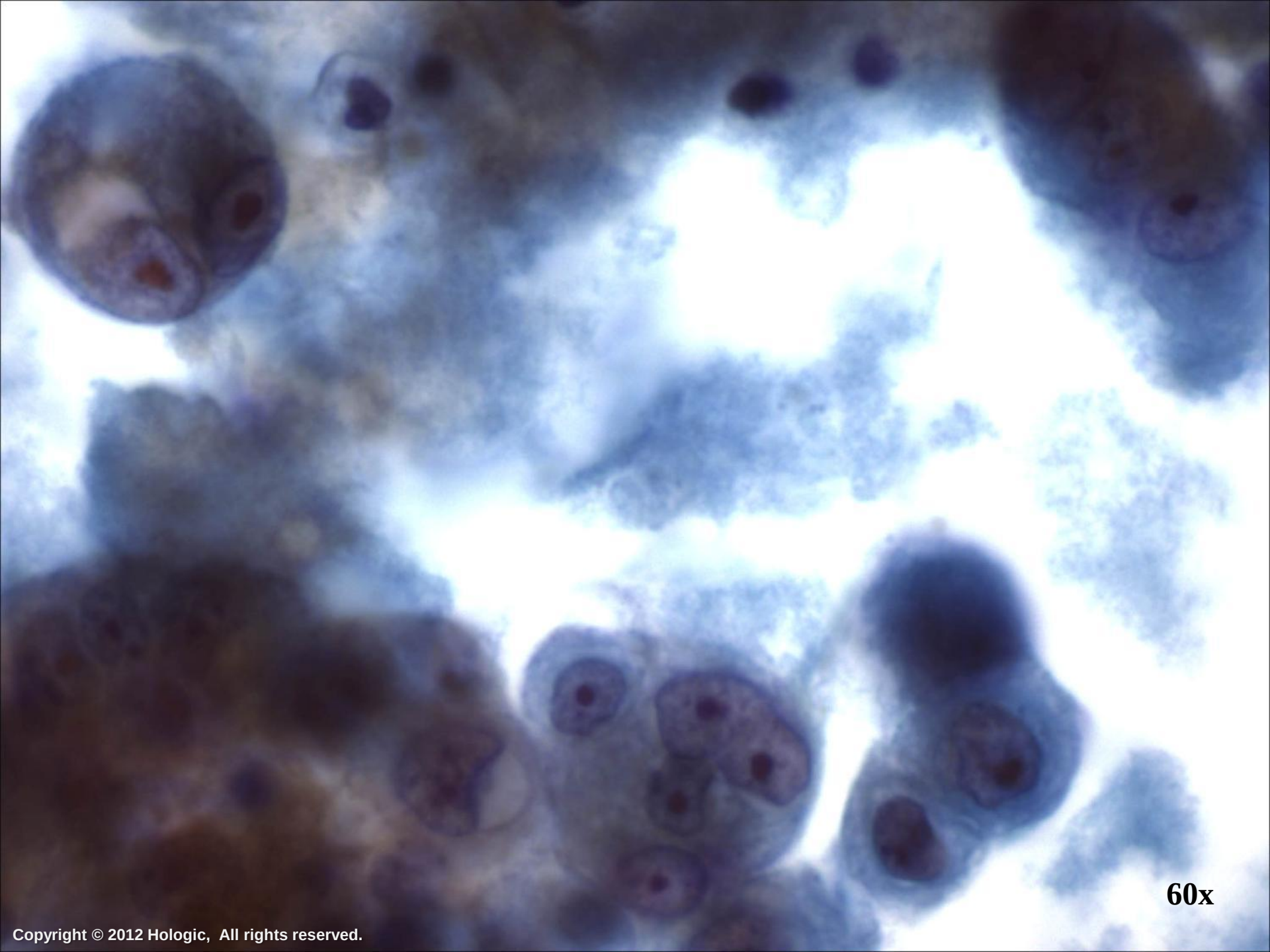

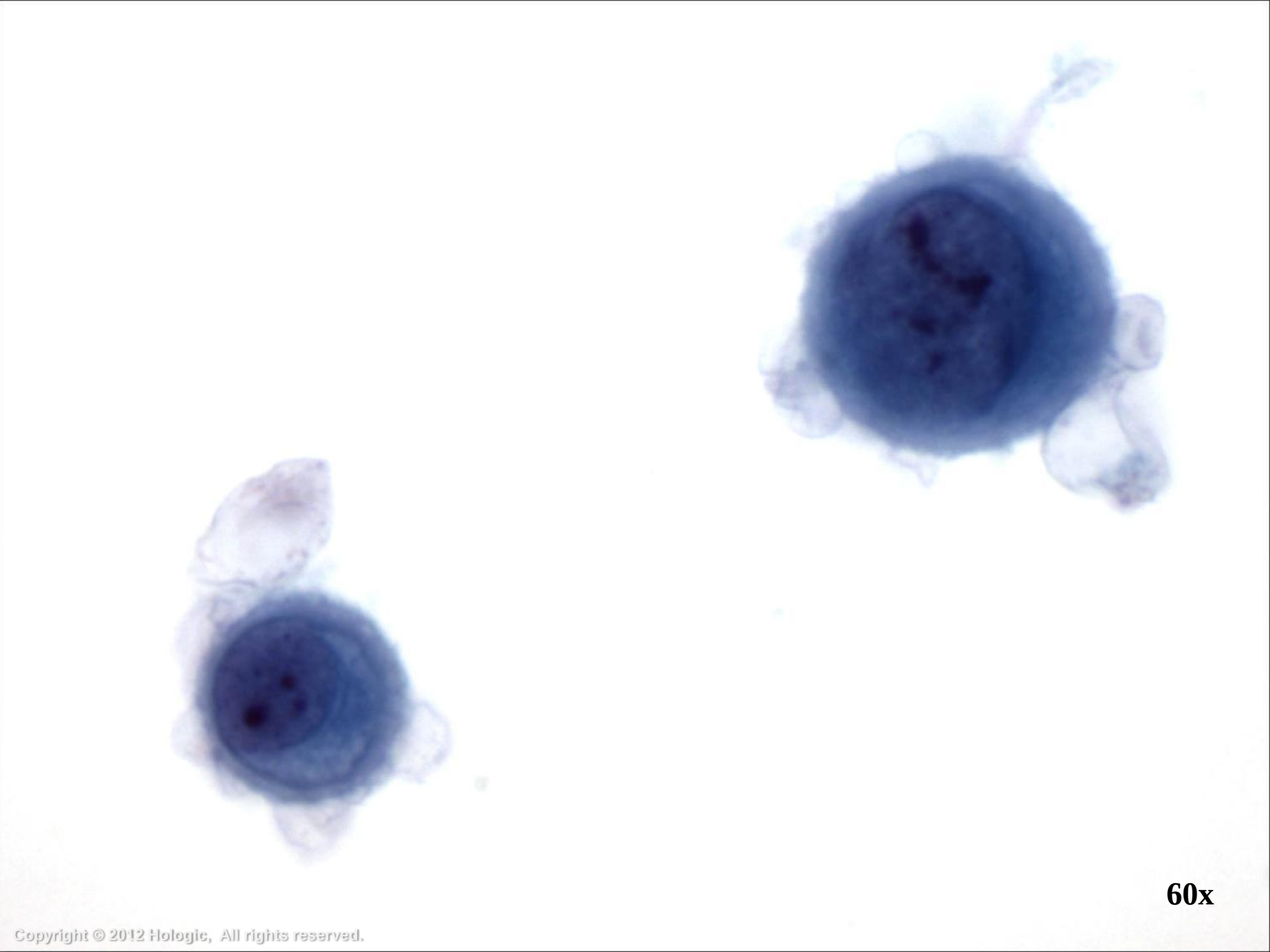

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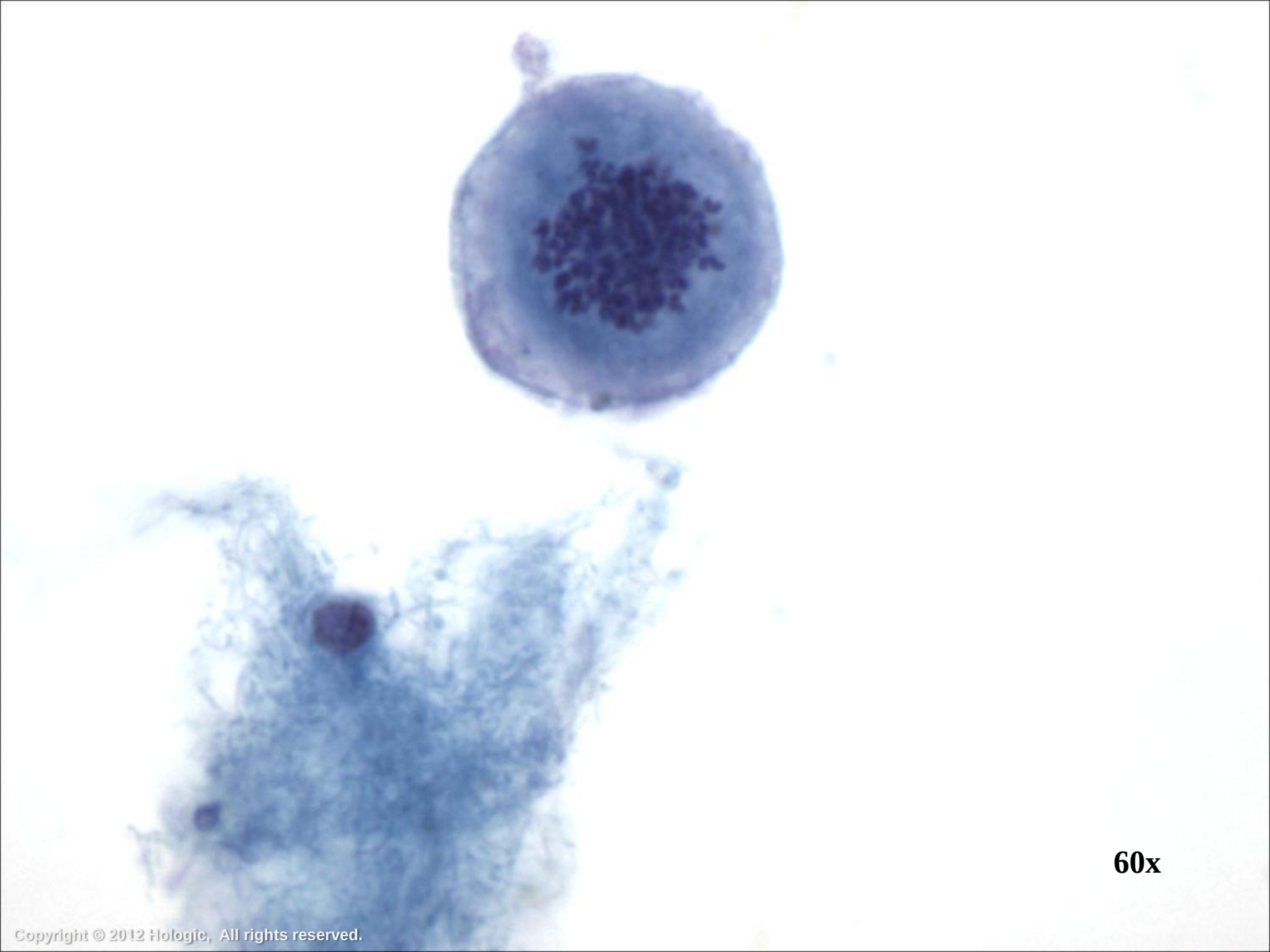

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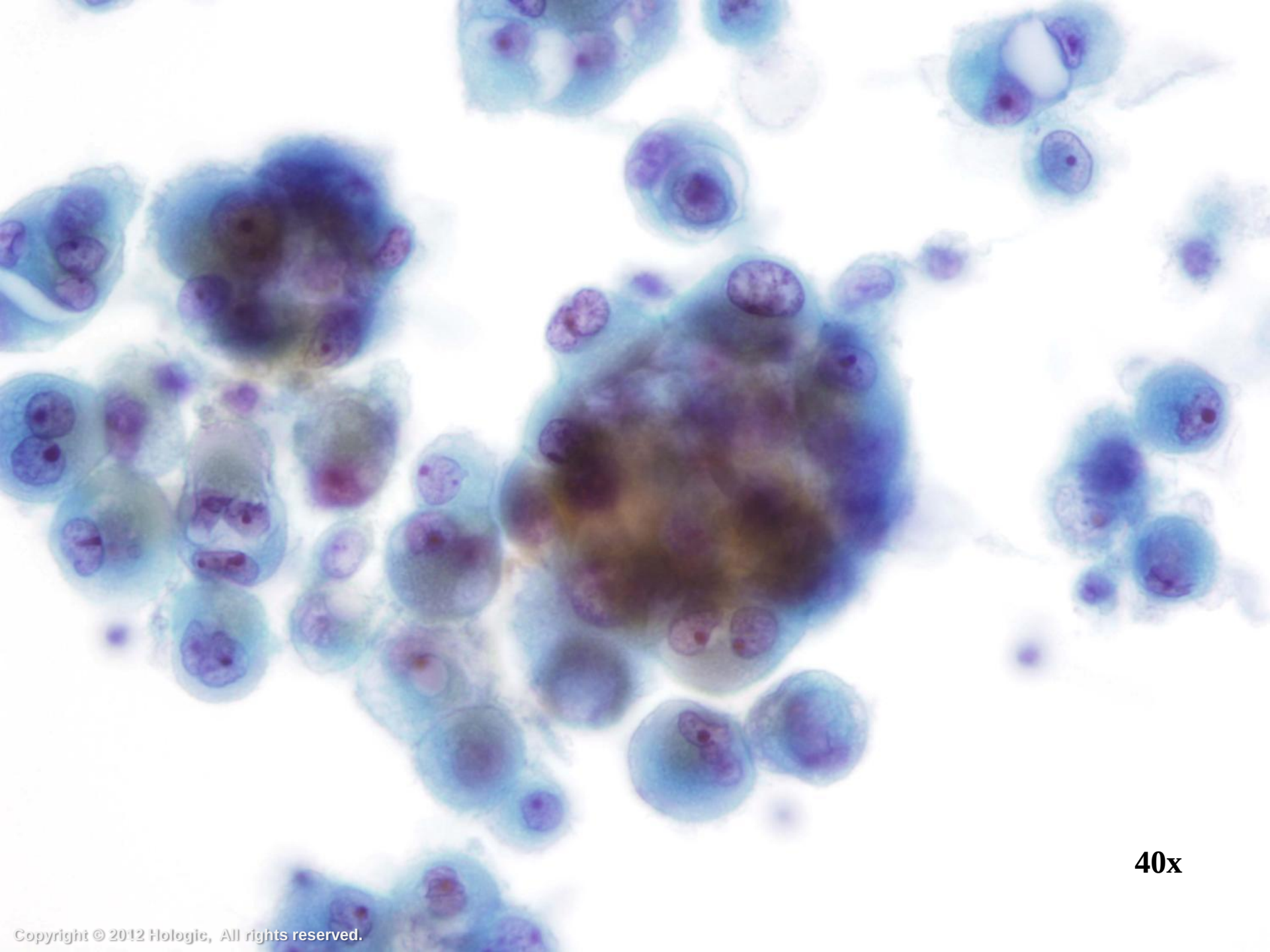

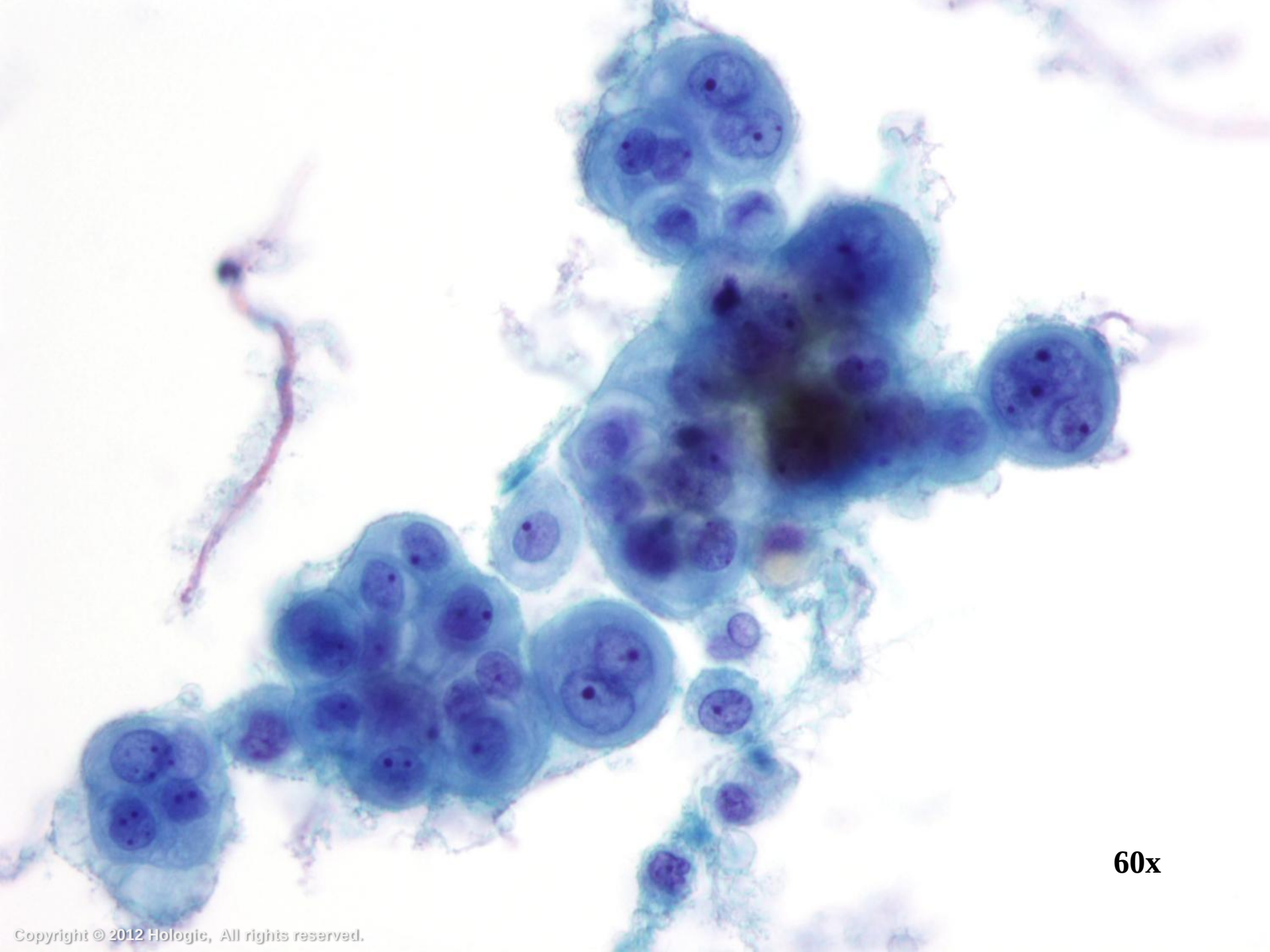

Suggested Immunochemistry Markers

Mesothelioma		Adenocarcinoma	
Calretinin	+	Calretinin	-
WT-1	+	WT-1	-
CEA	-	CEA	+
TTF-1	-	TTF-1*	+
MOC-31	-	MOC-31	+
Ber-EP4	-	Ber-EP4	+
B72.3	-	B72.3	+

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*Lung and thyroid only

Note – Expected staining results; observed in most but not all cases.

Primary Effusion Lymphoma

- Very rare subtype of diffuse large B-cell lymphoma
- Associated with human herpes virus 8 (HHV-8)
- Pleural, pericardial or peritoneal effusion
- Most cases occur in HIV positive patients with rare cases in immunocompromised patients
- Absence of mass lesion and HHV-8 positivity is essential for diagnosis

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Primary Effusion Lymphoma

- Population of single, large cells
- Nuclei range from round to irregular and lobulated
- Coarse chromatin
- Prominent, irregular nucleoli
- Cytoplasm is scant to abundant
- Morphologically similar to diffuse large B-cell lymphoma

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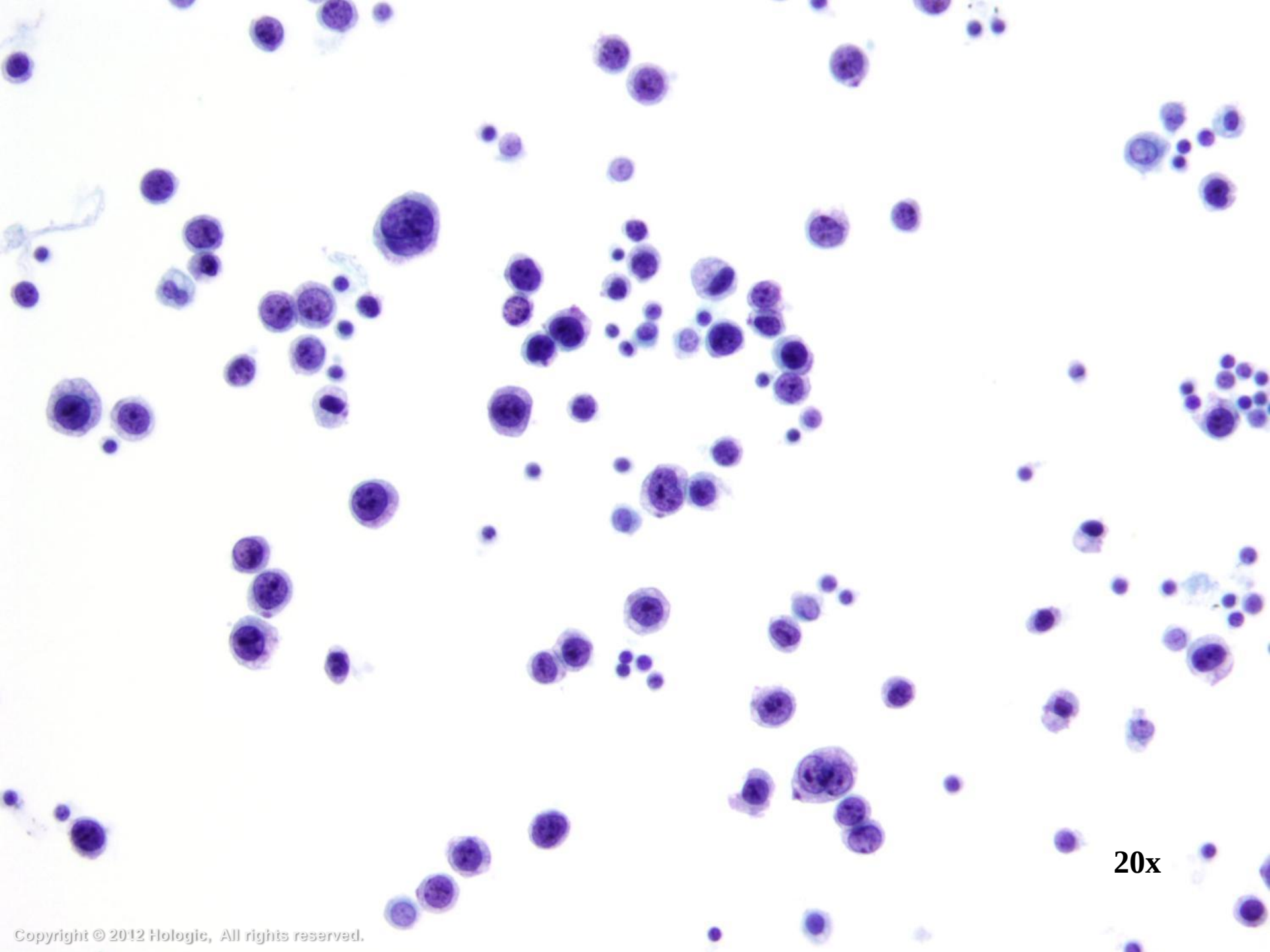

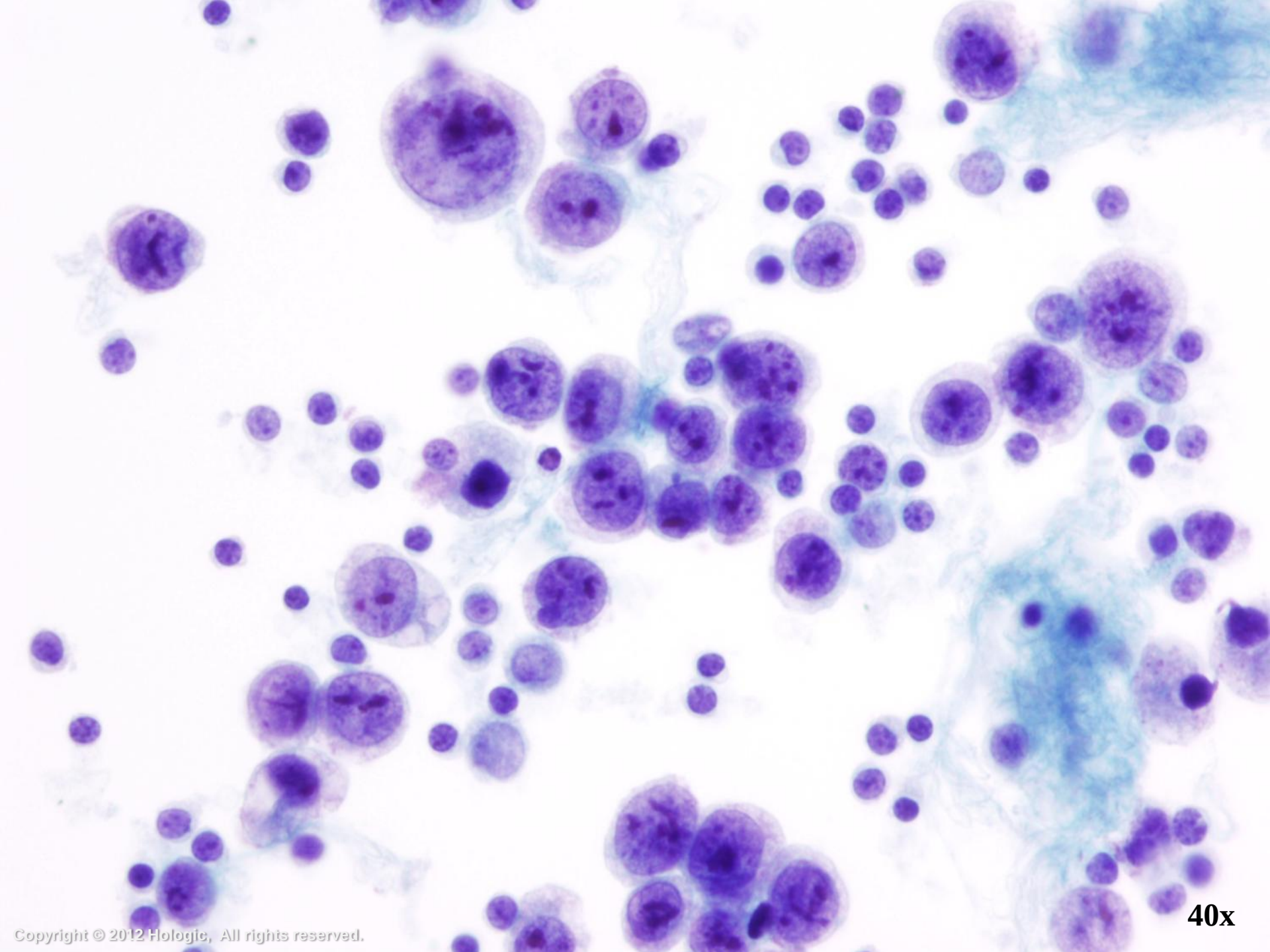

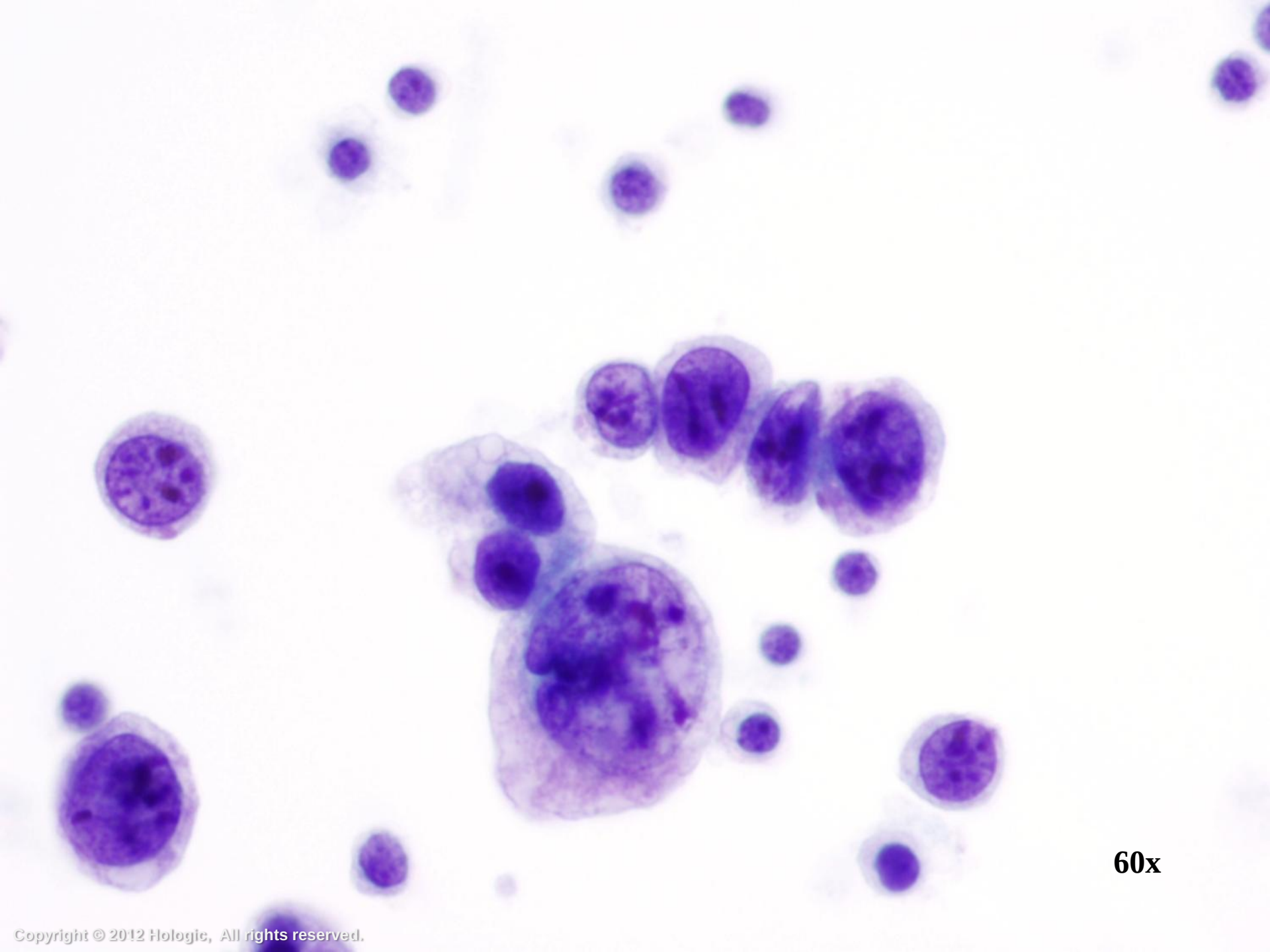

Suggested Immunochemistry Markers – Primary Effusion Lymphoma

- CD30 +
- CD45 +
- CD5 -
- CD20 -
- S100 -
- CEA -

Note – Expected staining results; observed in most but not all cases.

Adenocarcinoma

- Cytologic features:
  - Glandular acini, papillae or cell balls
  - Increased N/C ratios
  - Irregular nuclear membranes
  - Abnormal chromatin
  - Large or irregular nucleoli
  - Secretory vacuoles with mucin

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Specific Patterns of Adenocarcinoma

- Breast
  - Cells are usually fairly bland and uniform
  - Aggregates or predominantly single
  - Diagnostic clues are cannonballs, intracytoplasmic lumens and single file chains of cells
  - Positive for mucicarmine stain

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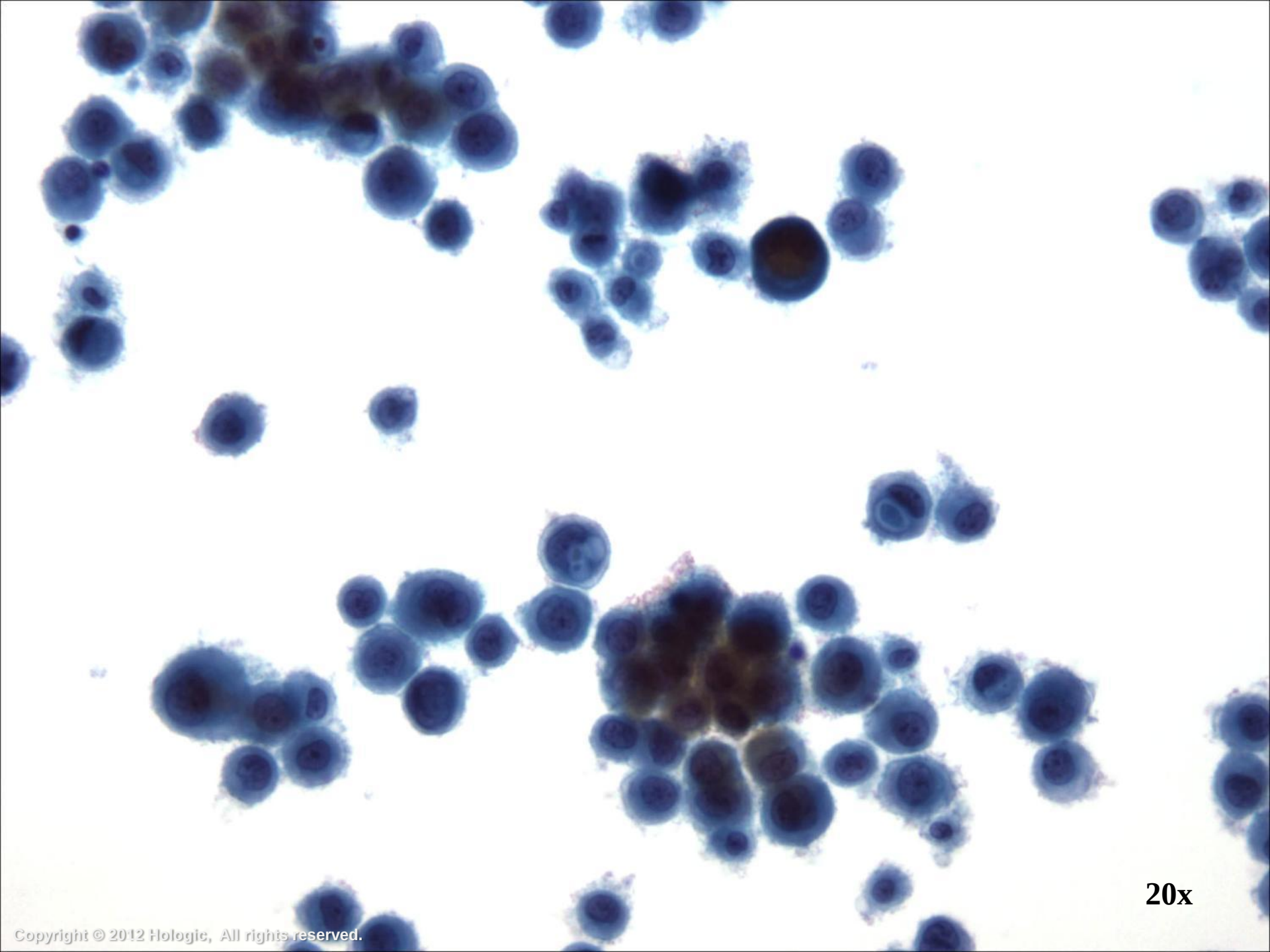

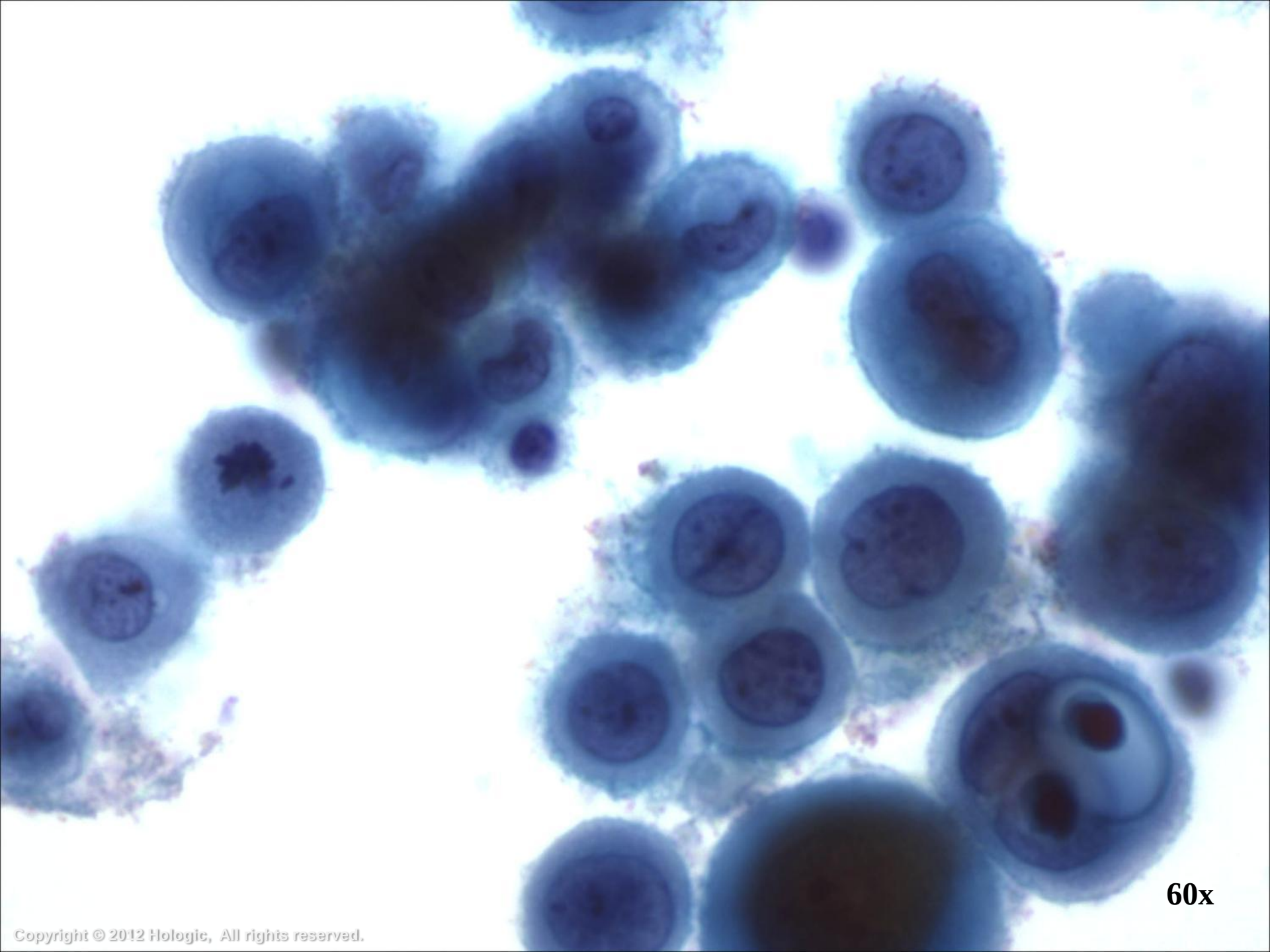

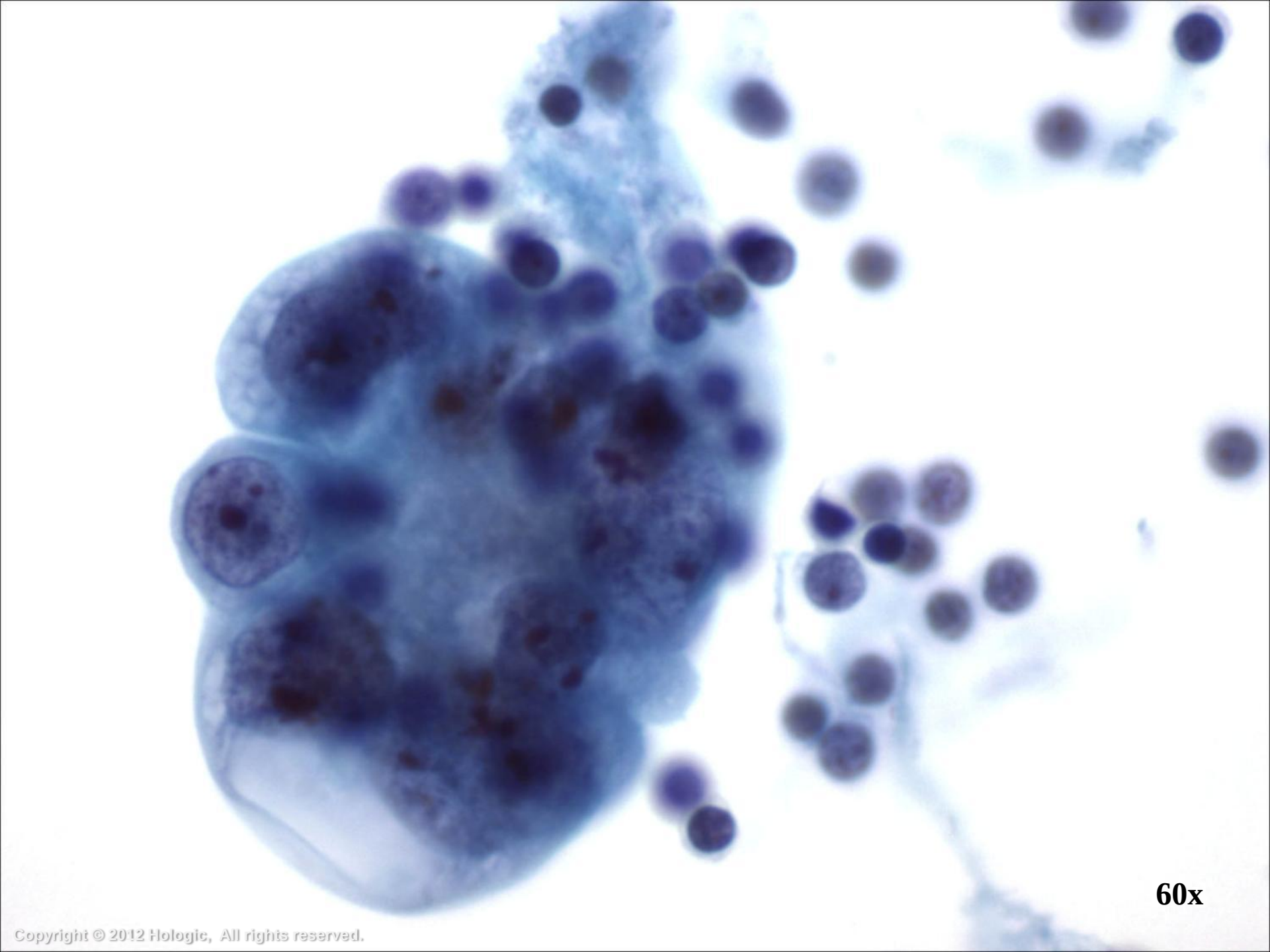

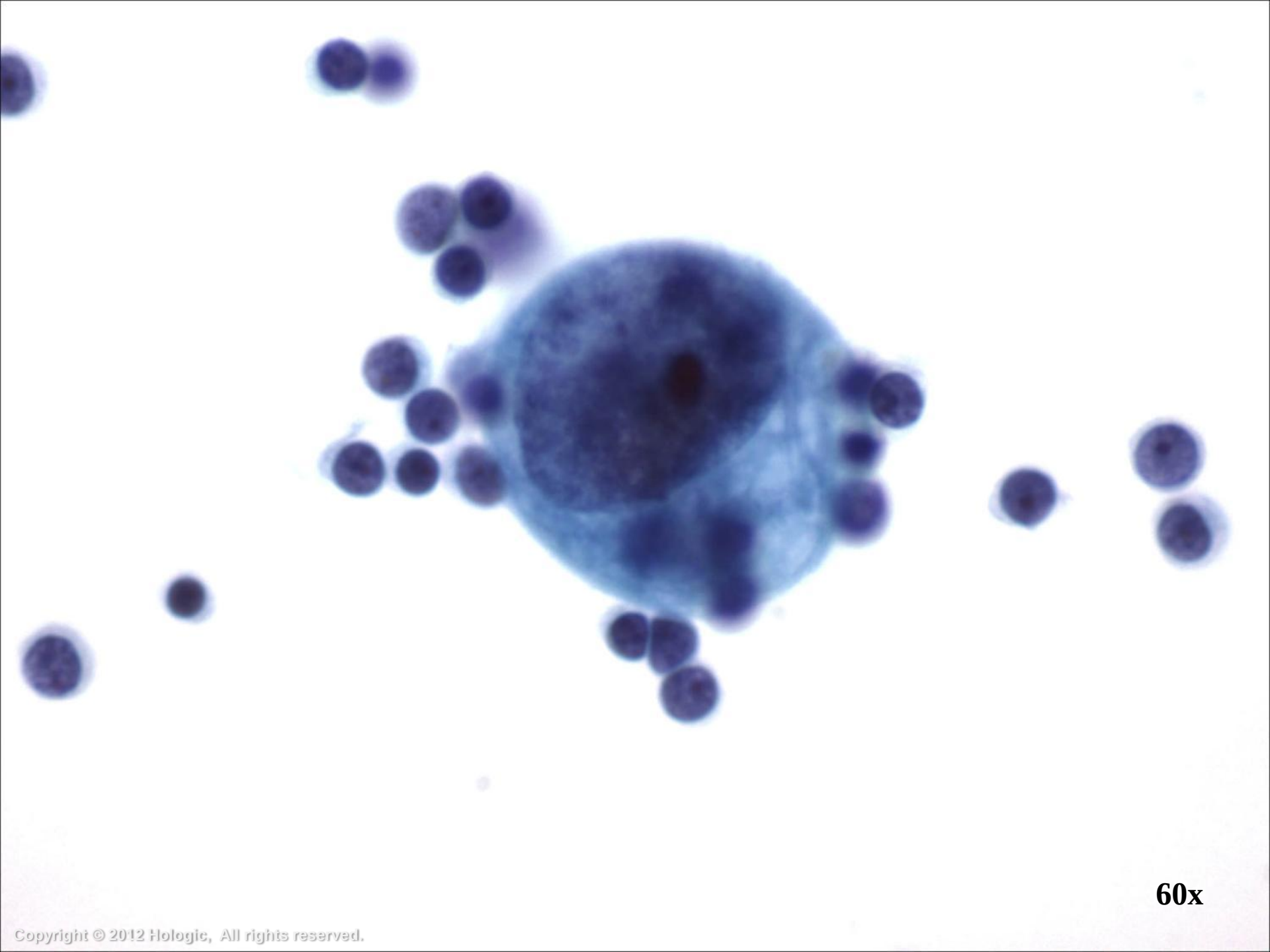

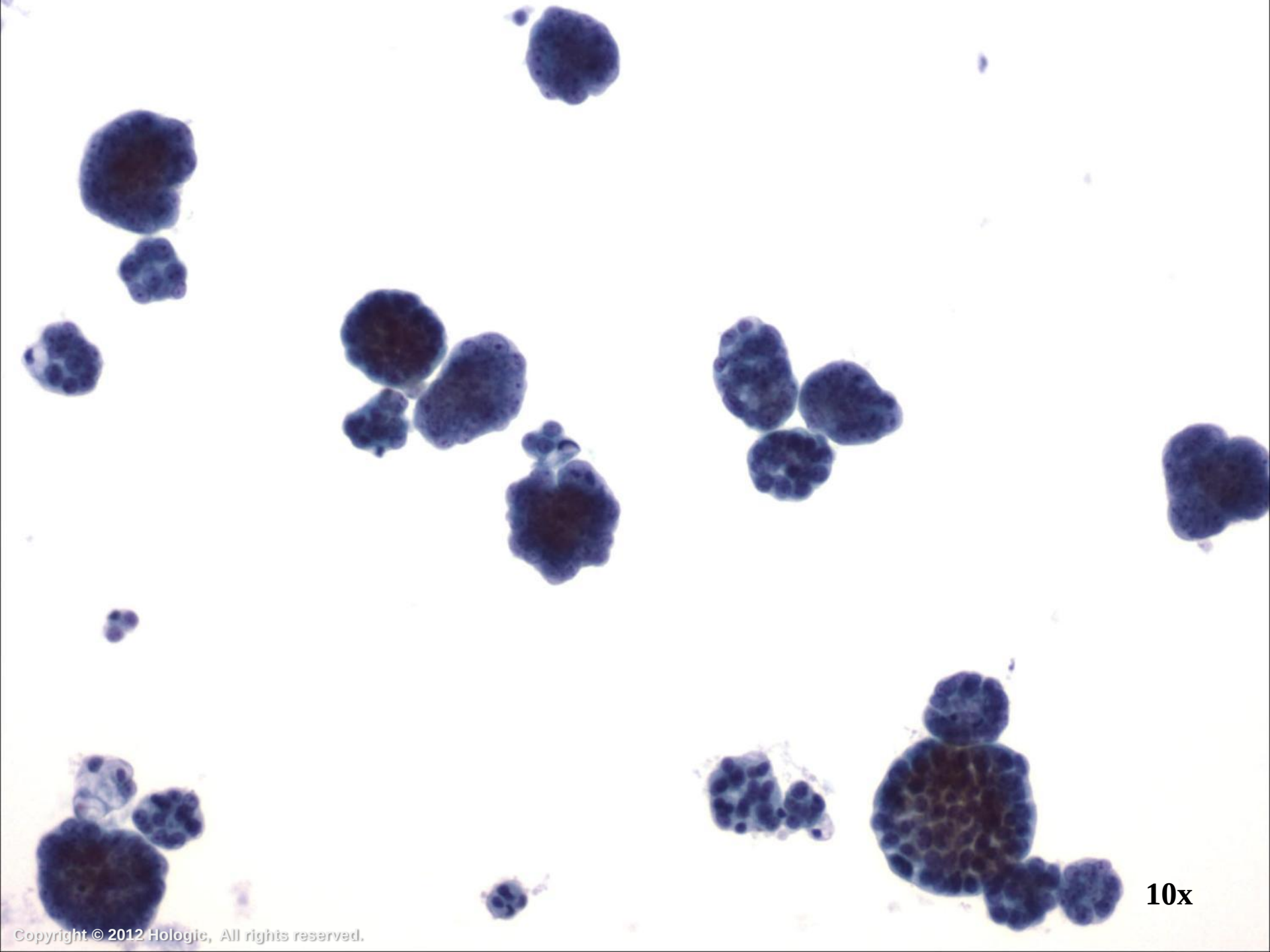

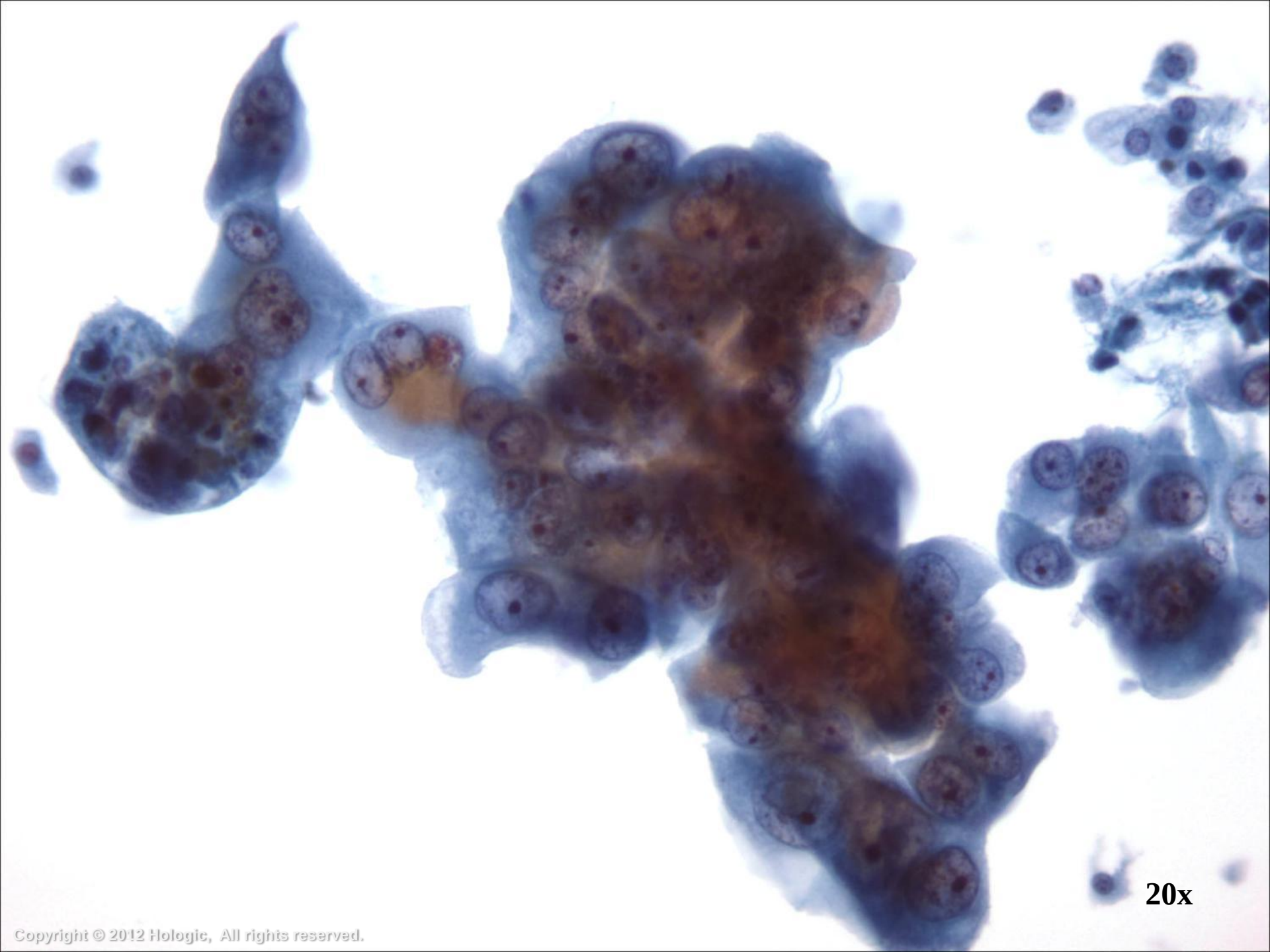

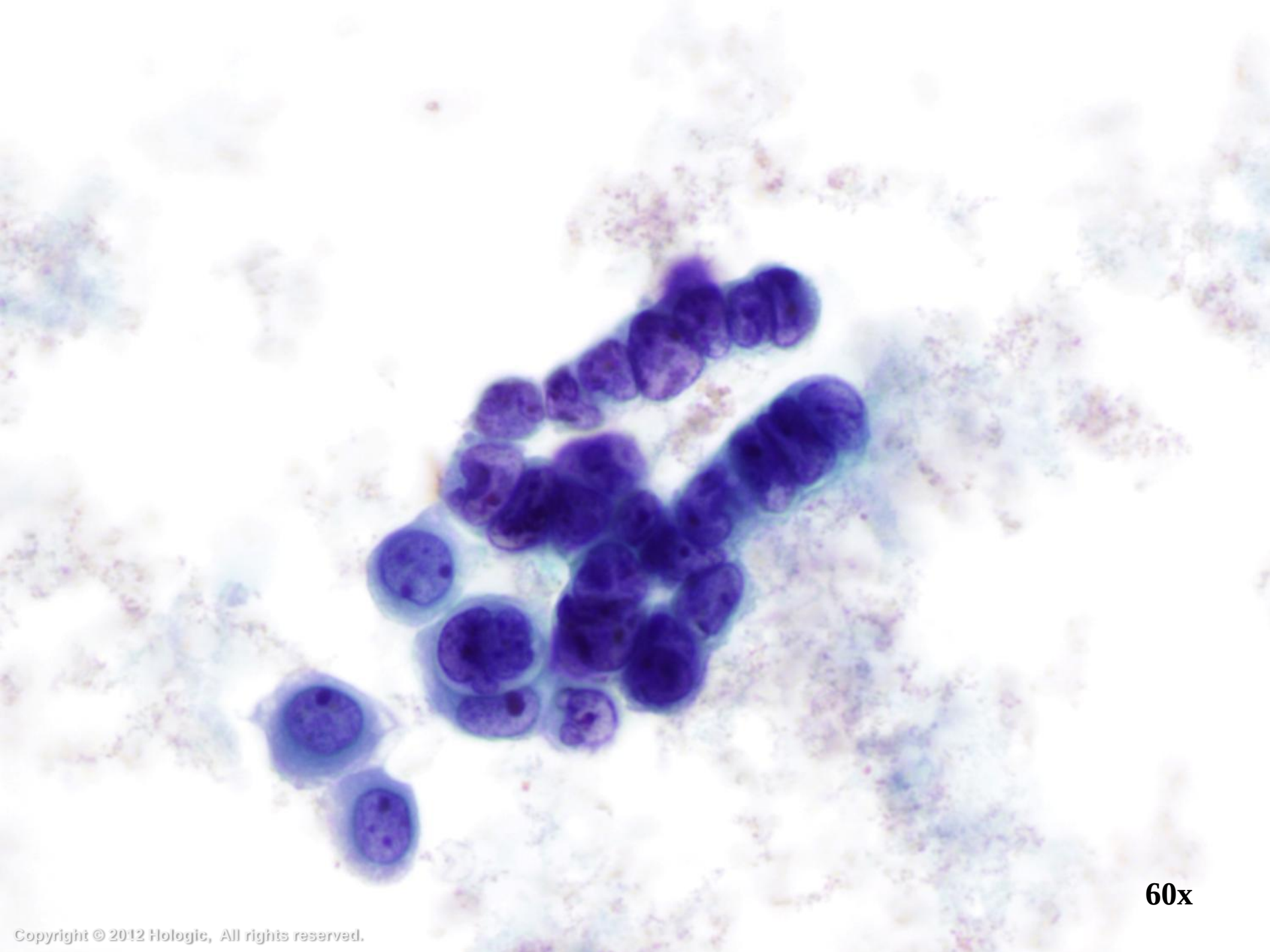

60x

Suggested Immunochemistry Markers – Adenocarcinoma Breast

- CK 7 +
- CK 20 -
- BerEP4 +
- ER/PR +
- Mammaglobin +
- TTF-1 -

Note – Expected staining results; observed in most but not all cases.

Specific Patterns of Adenocarcinoma

- Lung
  - Large pleomorphic cells that can range from bland to bizarre
  - Nuclei are often hyperchromatic with fine to coarsely granular chromatin and prominent nucleoli
  - Cytoplasm is often vacuolated and may contain mucin

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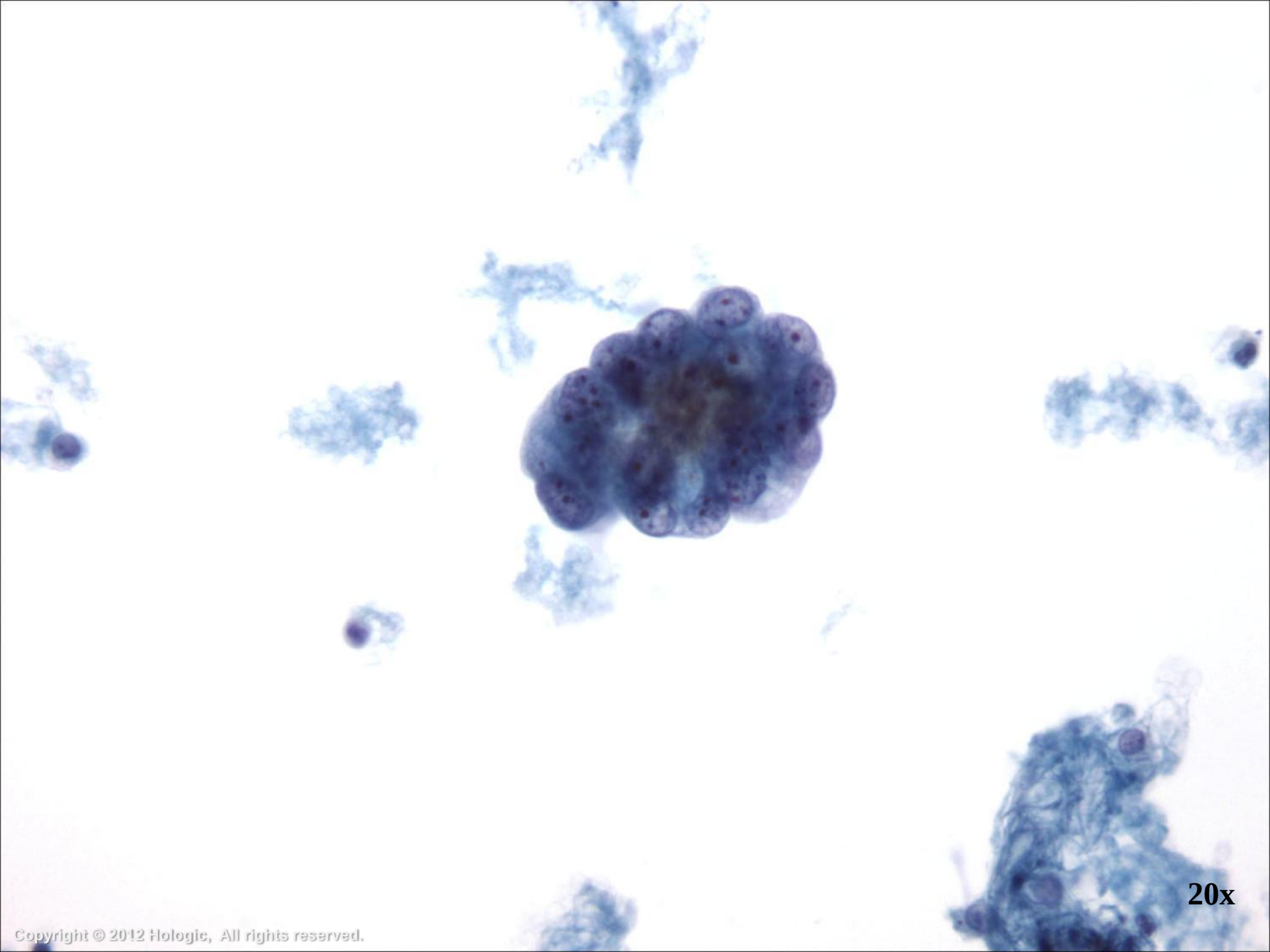

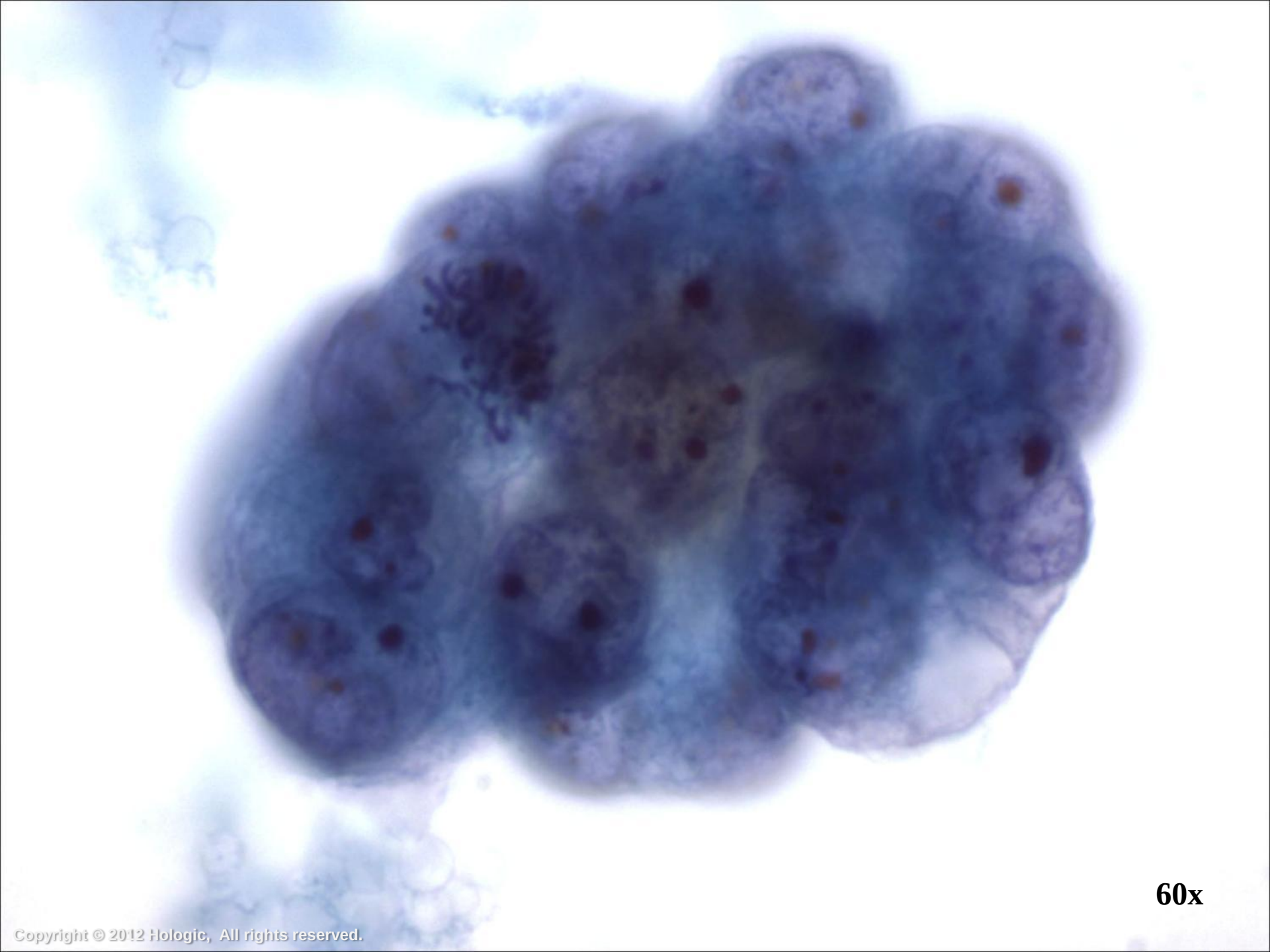

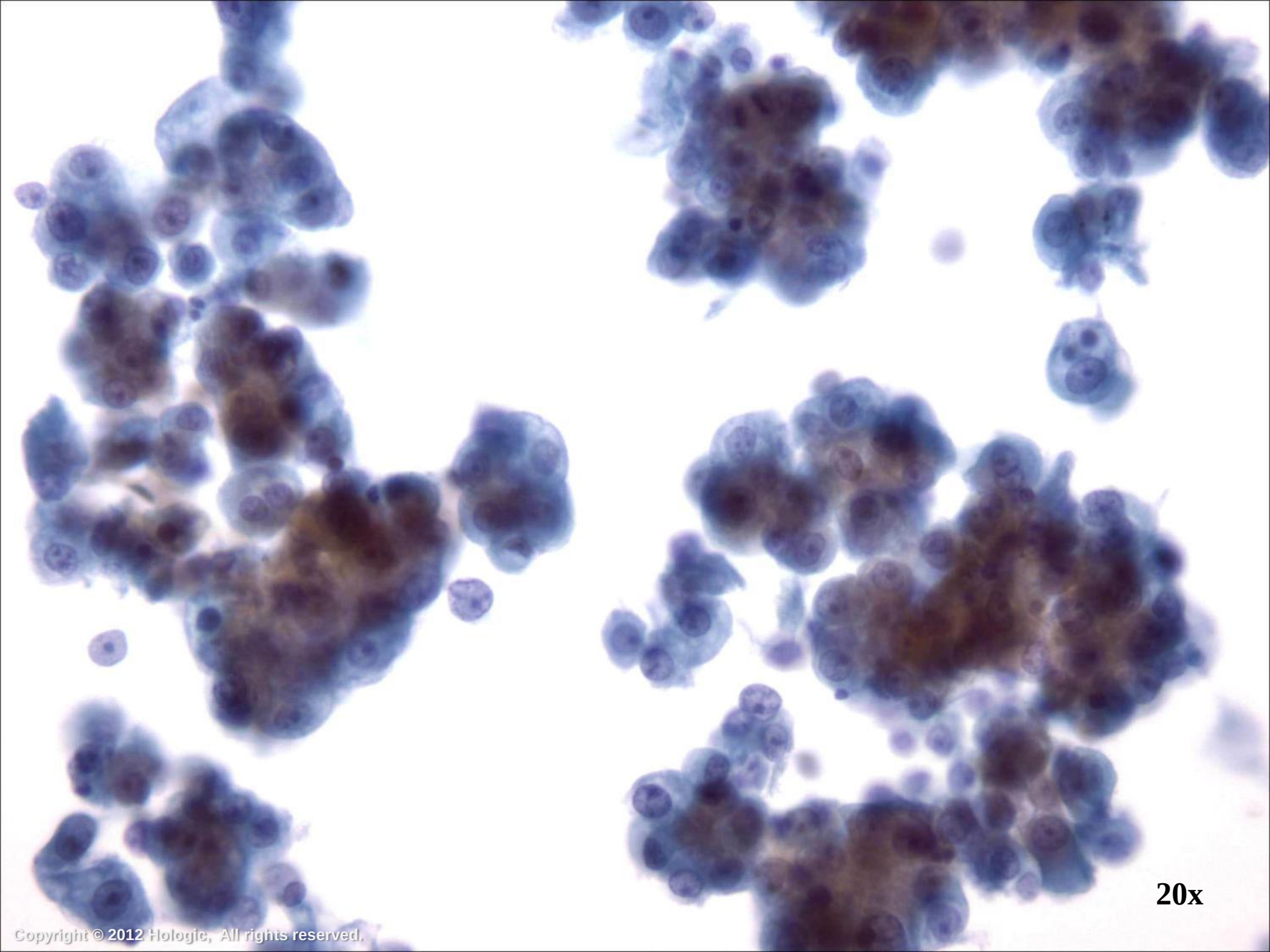

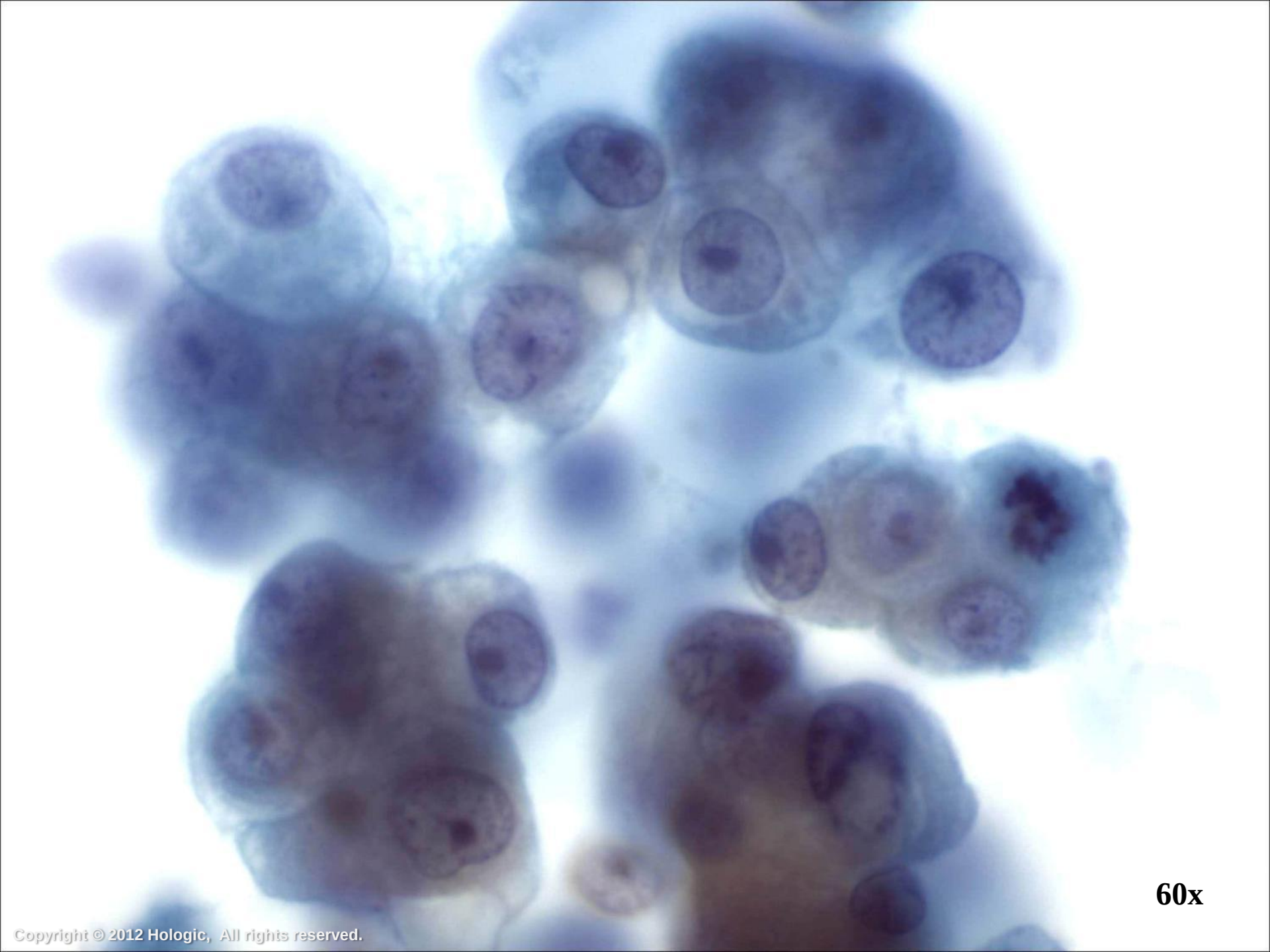

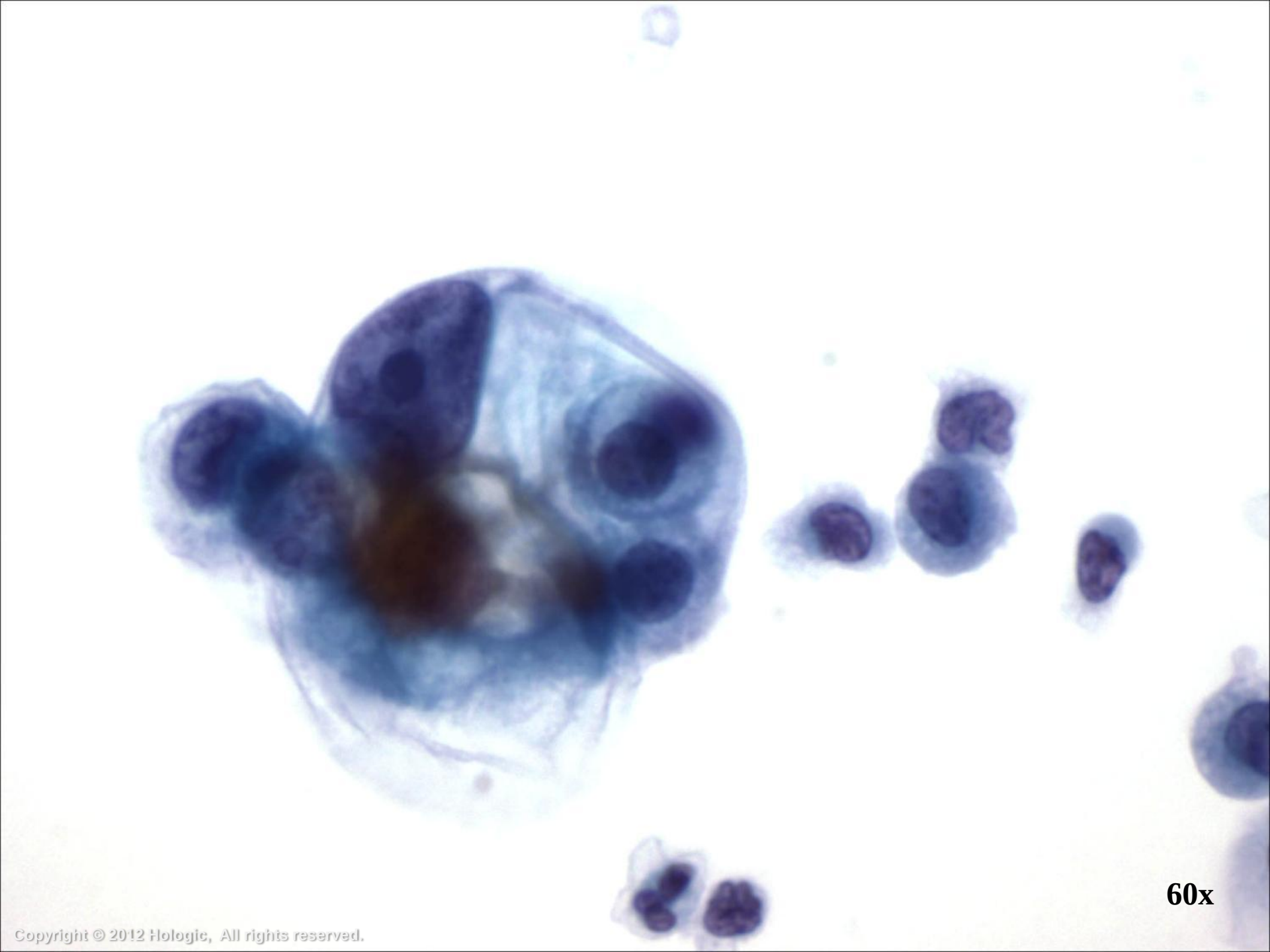

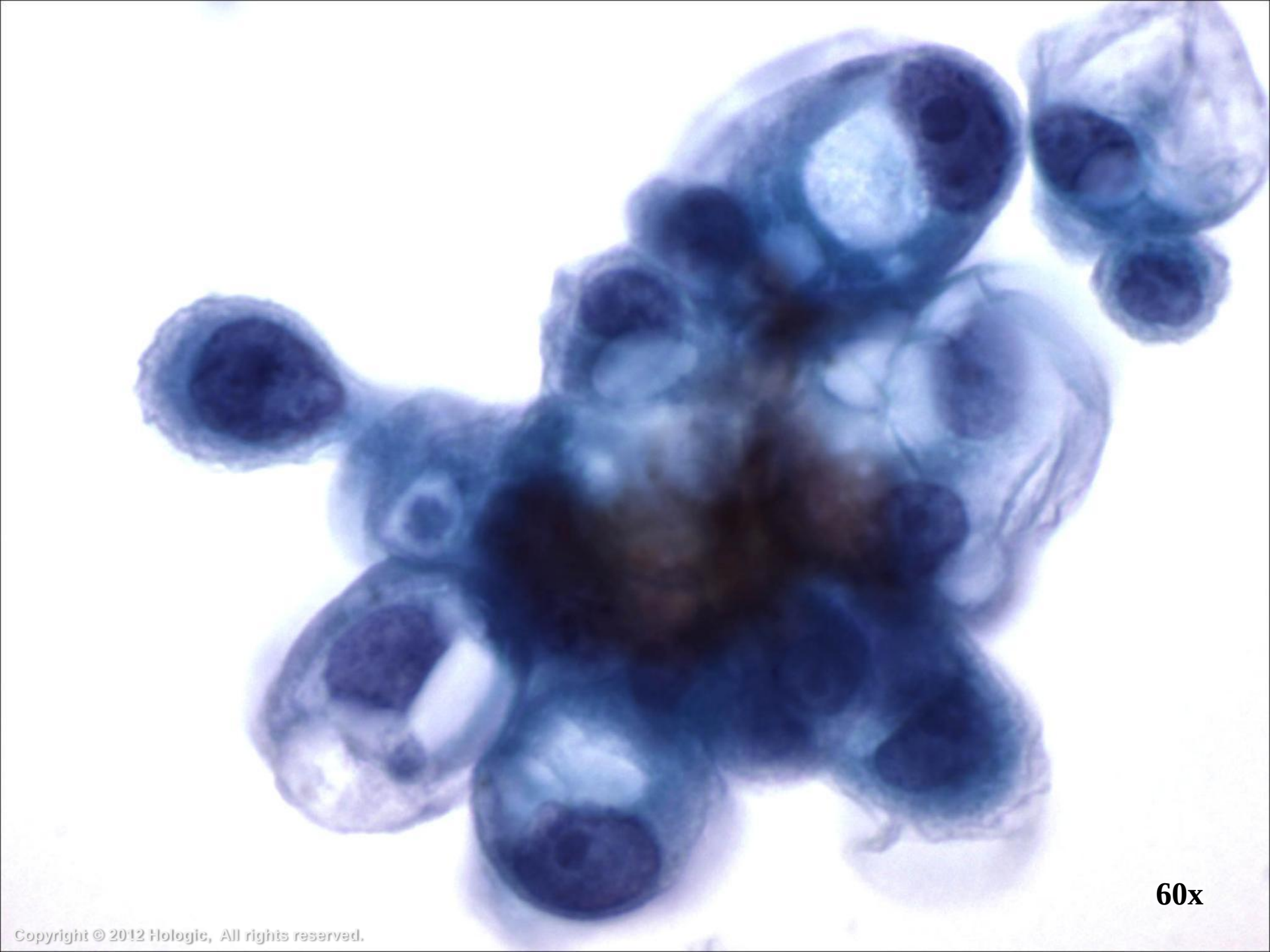

Suggested Immunochemistry Markers – Adenocarcinoma Lung

- CK 7 +
- CK 20 -
- TTF-1 +
- BerEP4 +

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Note – Expected staining results; observed in most but not all cases.

Specific Patterns of Adenocarcinoma

- Renal
  - Papillary or acinar groups of cells
  - Granular or clear cytoplasm (dense in effusions)
- Gastric
  - Intestinal type sheds clusters of large, highly atypical cells
  - Gastric type sheds single signet ring cells
- Colorectal
  - Papillary or acinar aggregates of tall columnar cells
  - Palisading nuclei with highly irregular nuclear borders
  - Signet rings can also be seen

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Renal Cell Carcinoma

- The following slides contain examples of renal cell carcinoma

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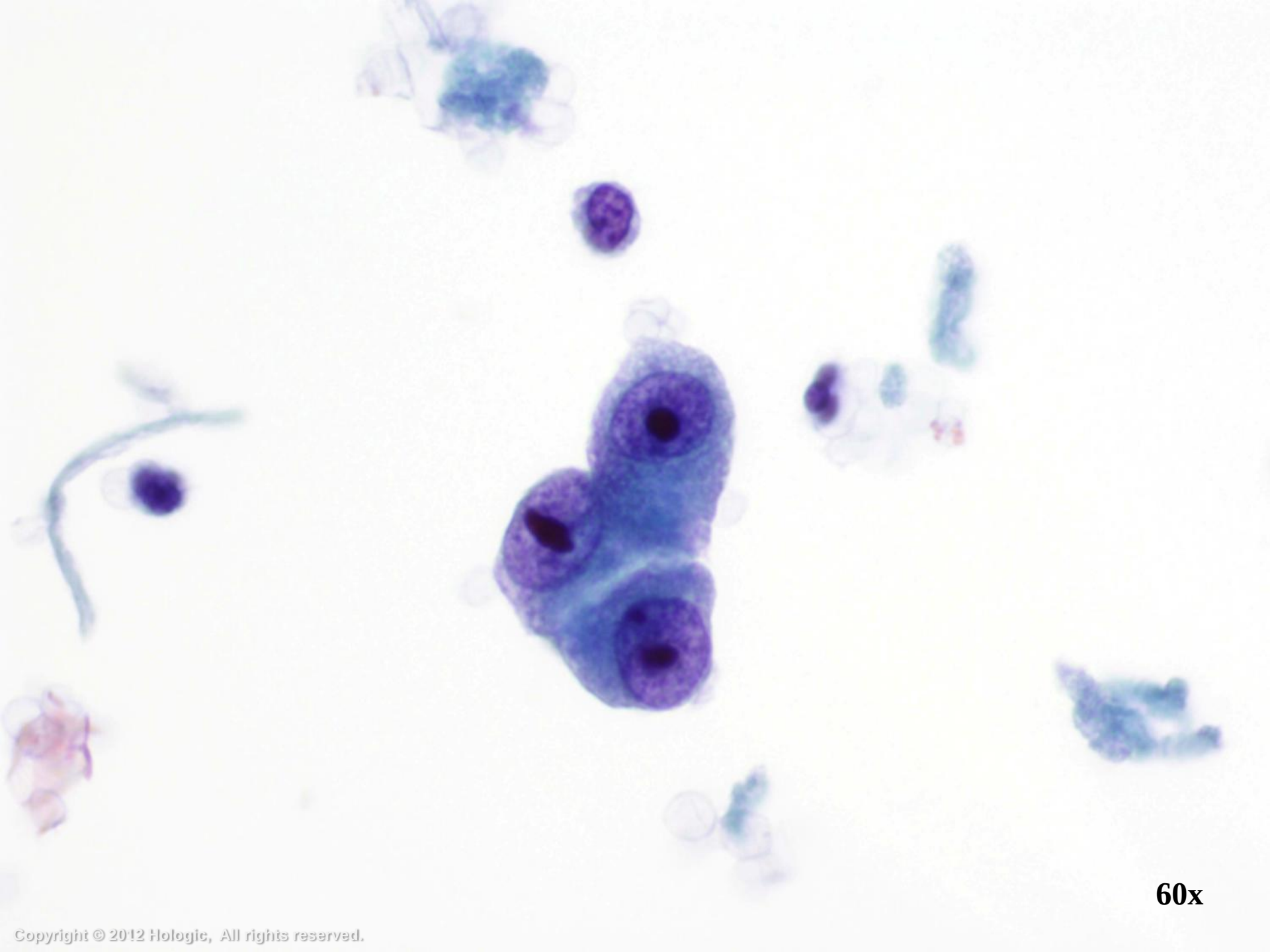

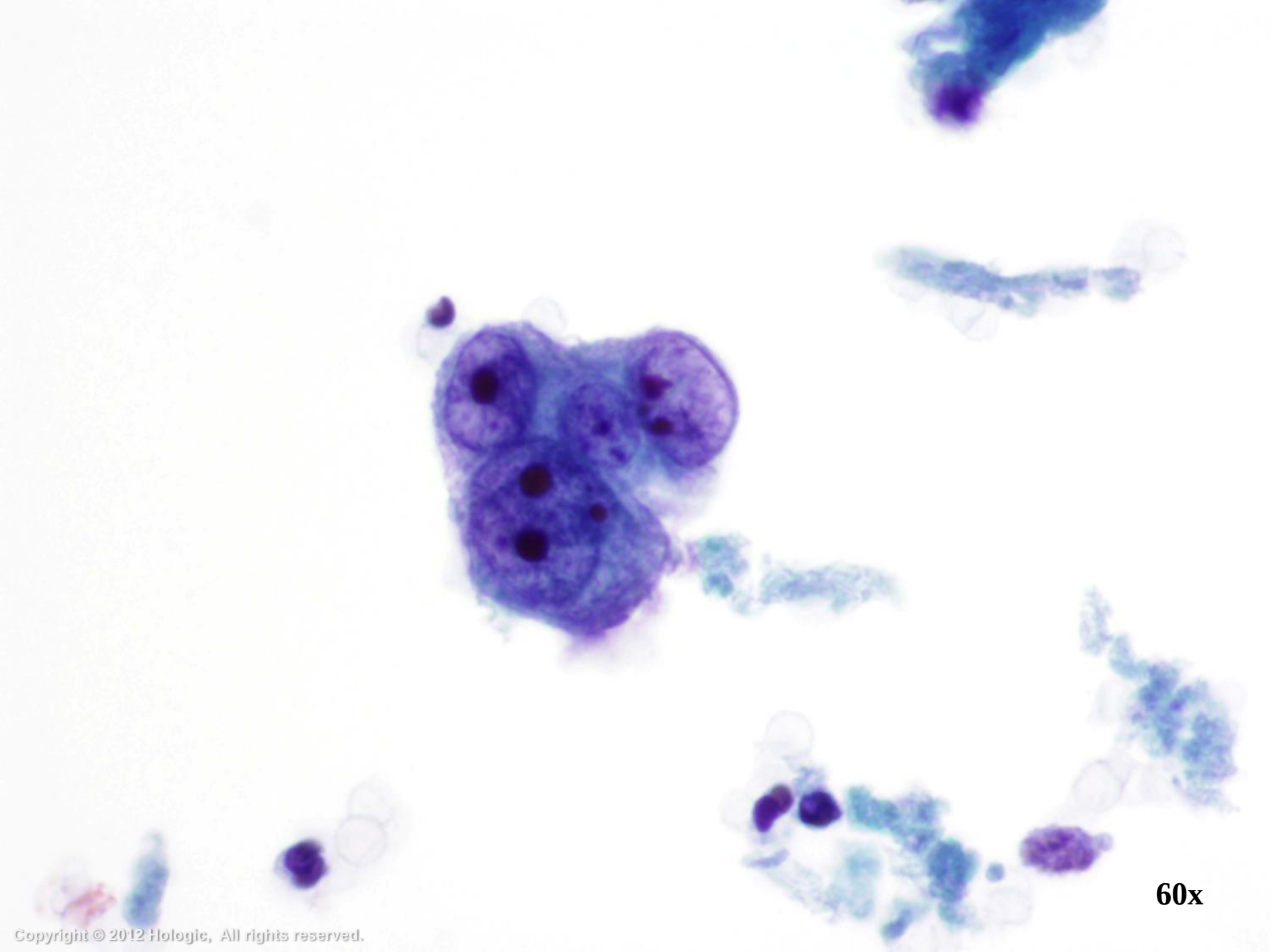

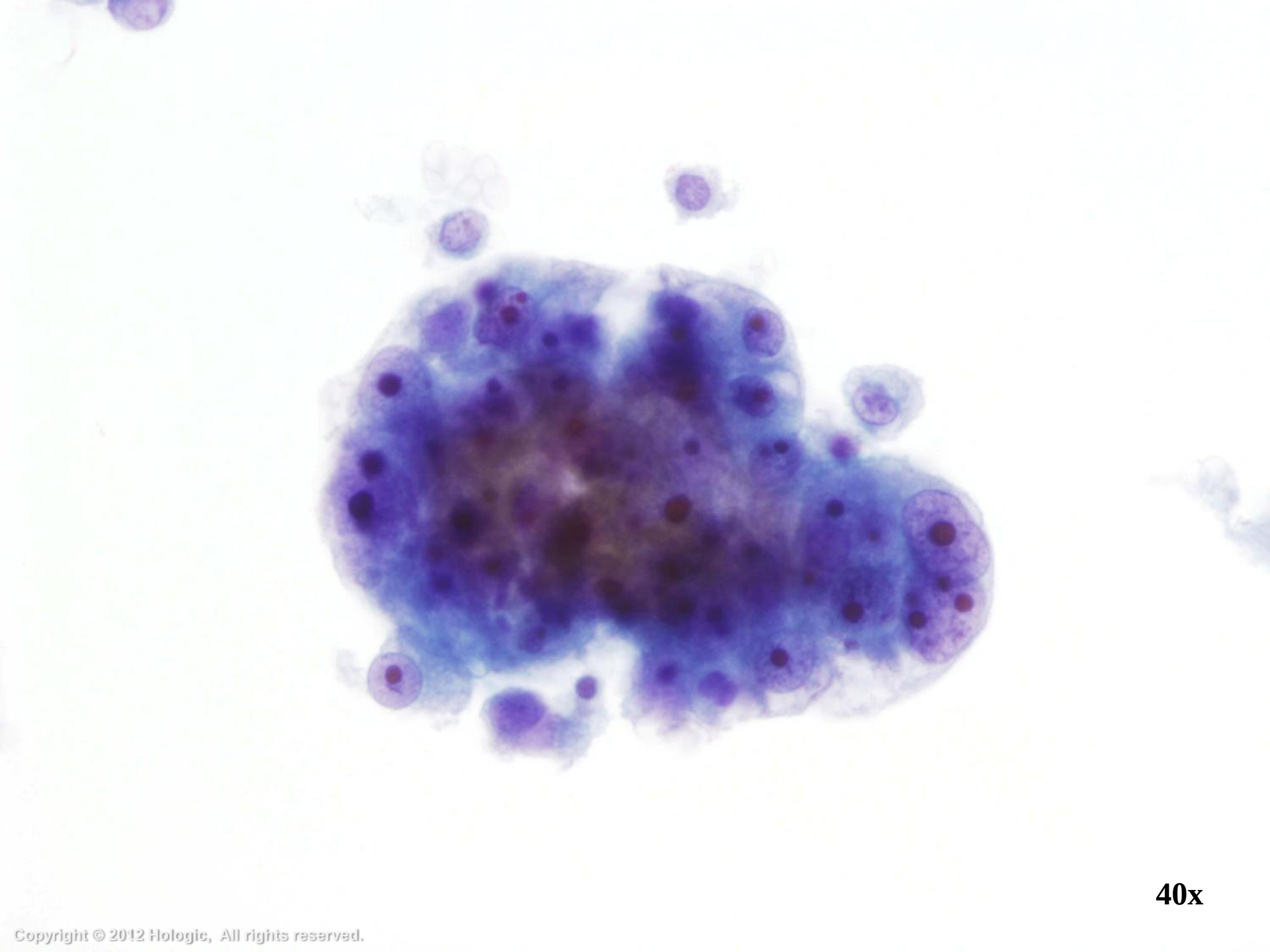

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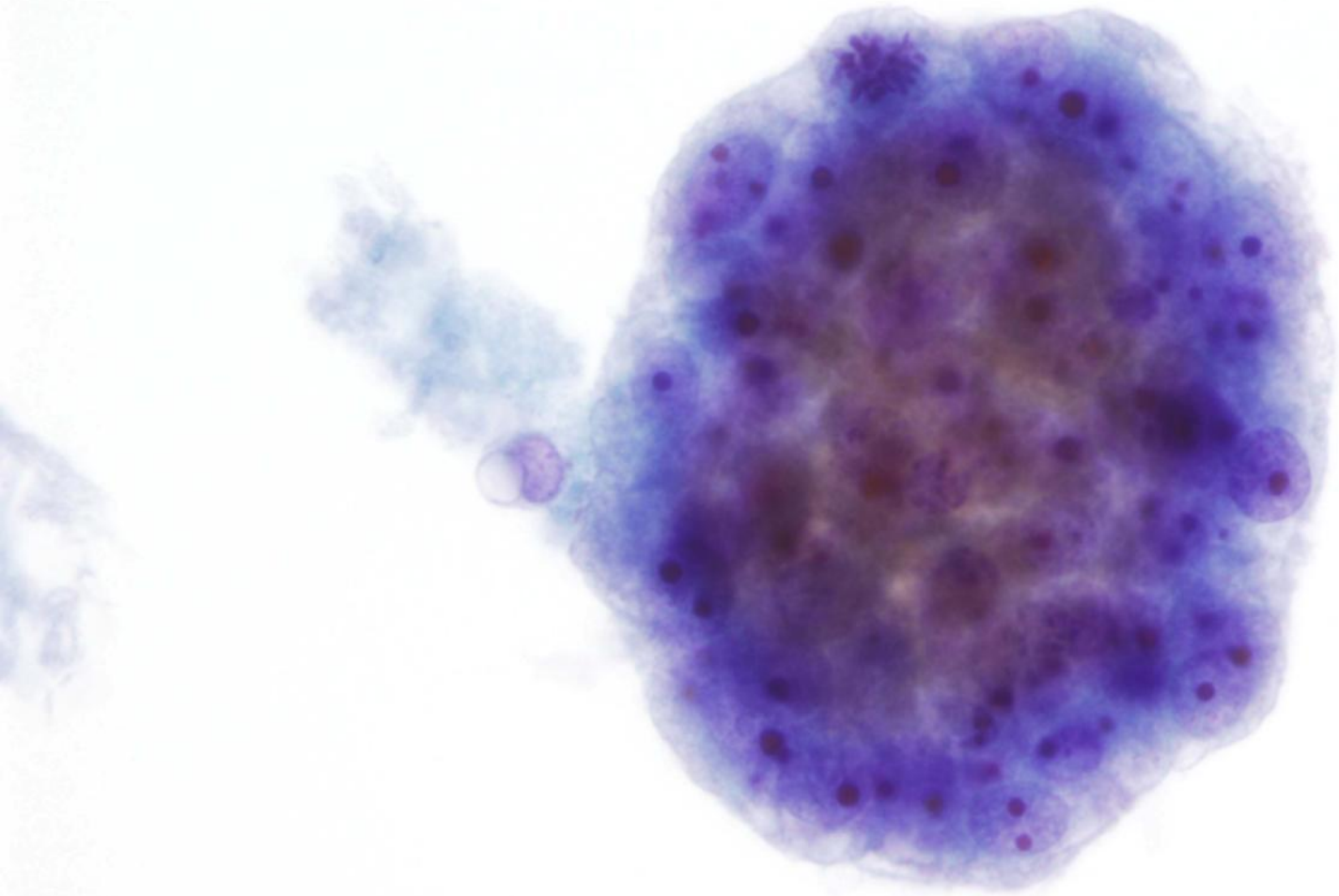

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Suggested Immunochemistry Markers – Adenocarcinoma Renal

- CK 7 -
- CK 20 -
- Vimentin +
- CD 10 +
- RCC-m +

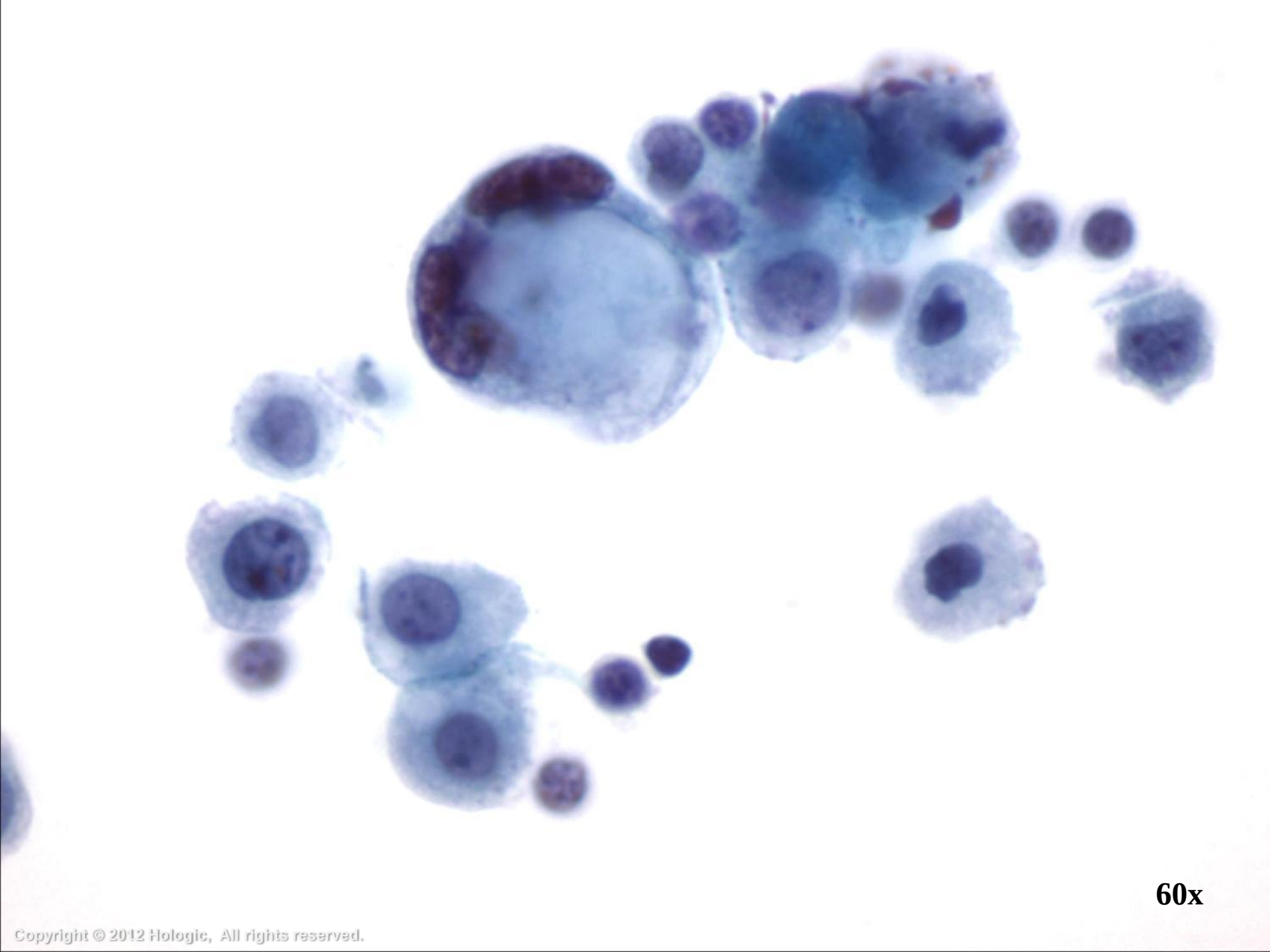
ThinPrep Non-Gyn

Note – Expected staining results; observed in most but not all cases.

Gastric Carcinoma

- The following slides contain examples of gastric carcinoma

ThinPrep Non-Gyn



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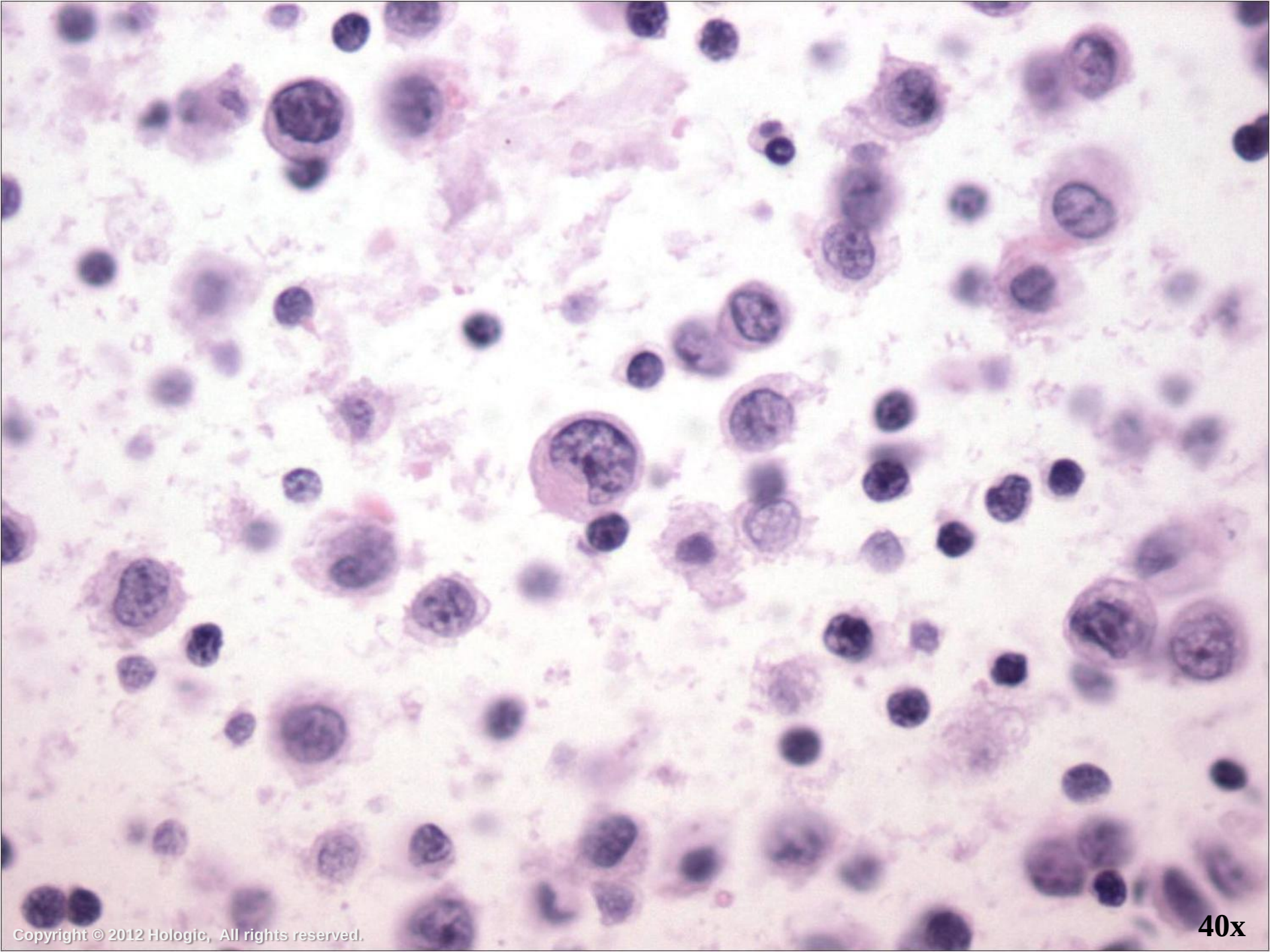

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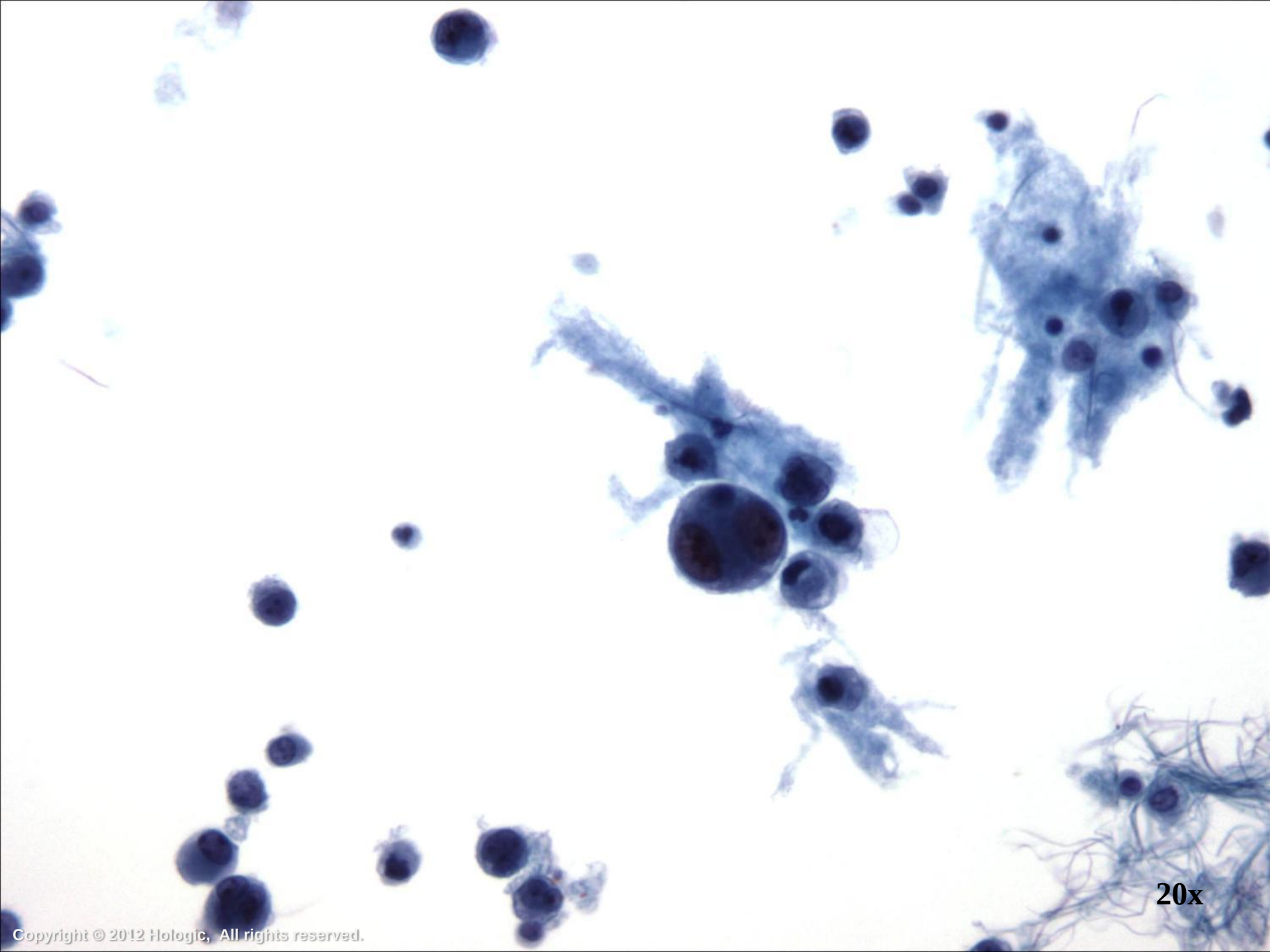

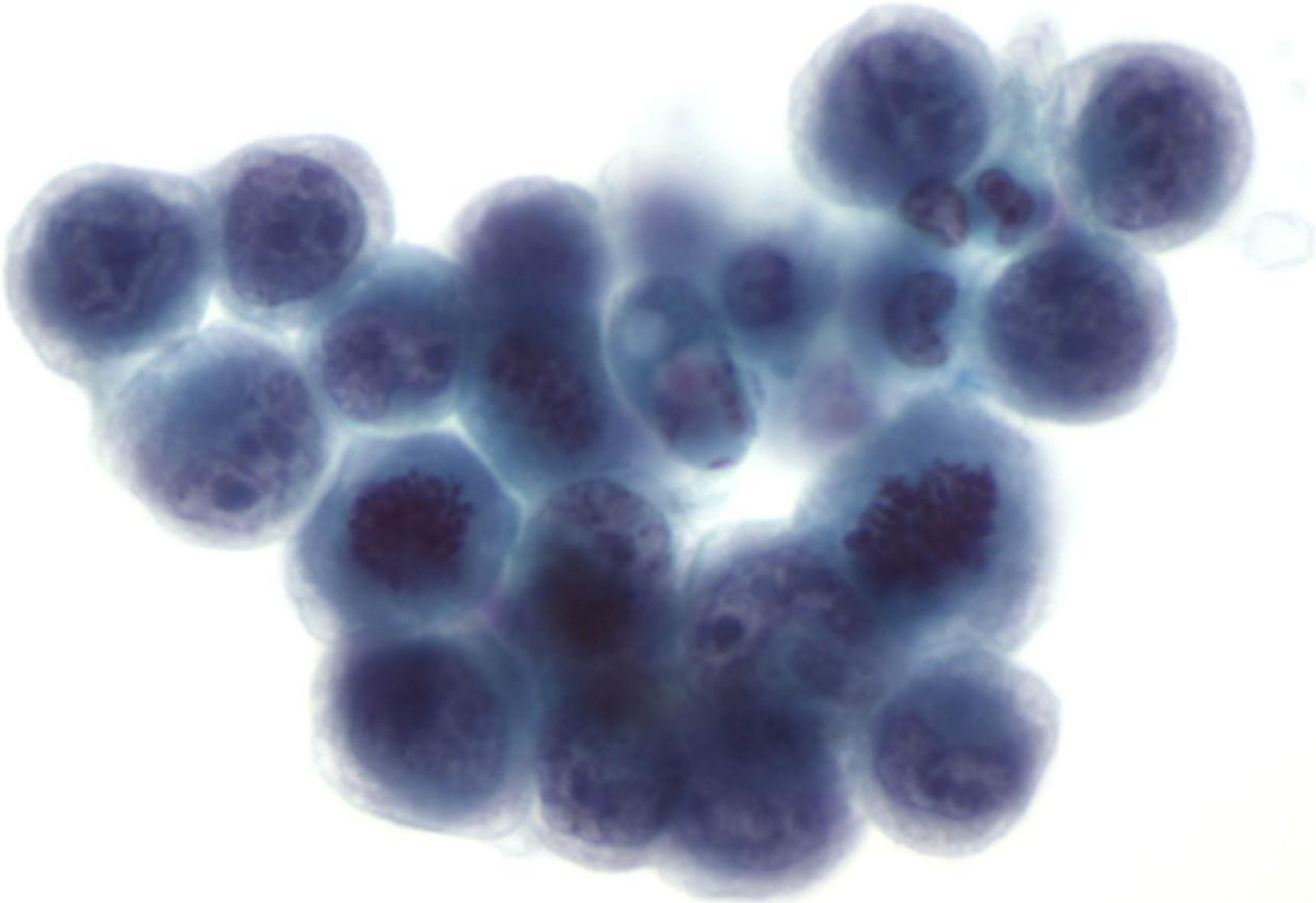

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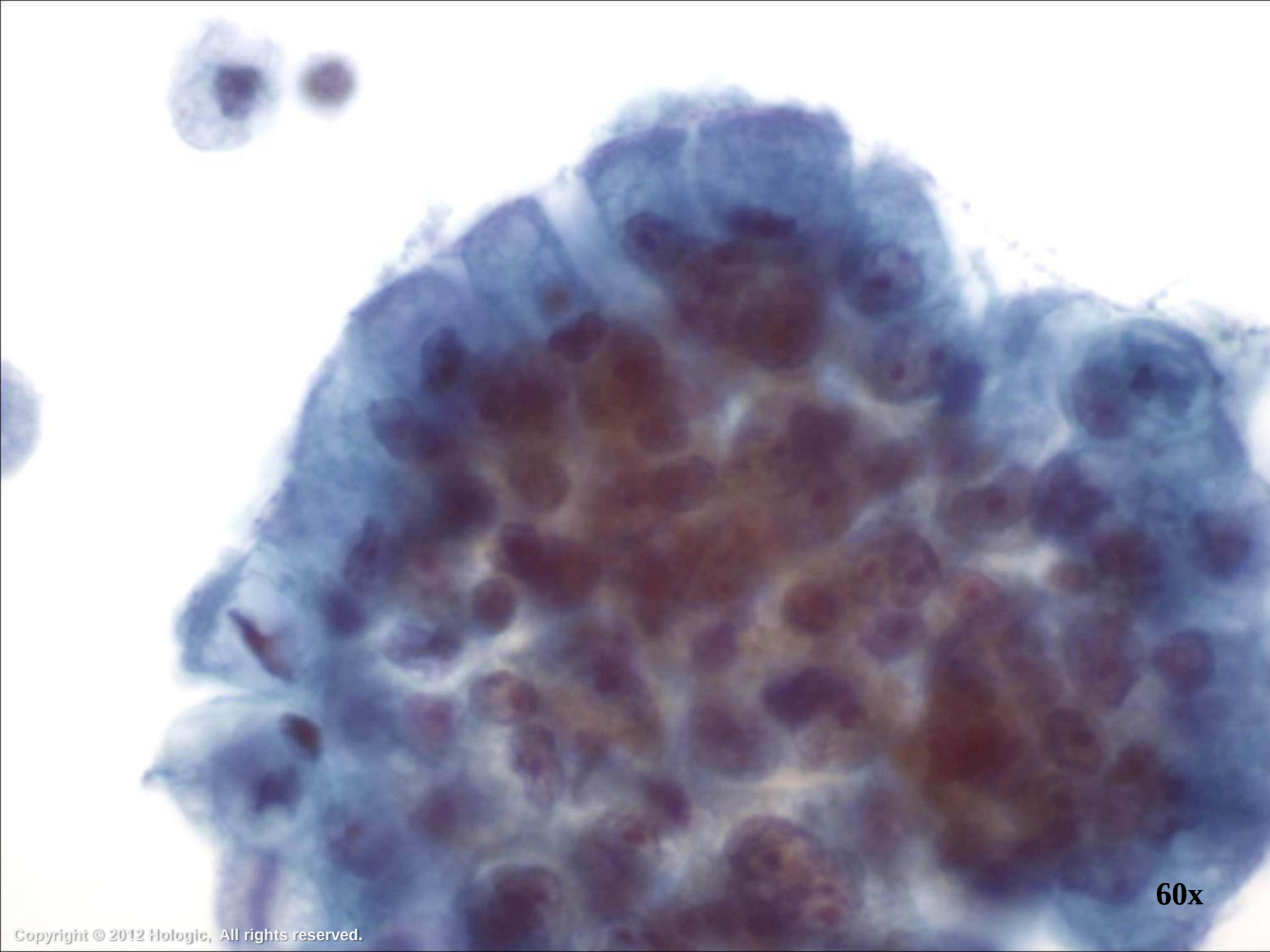


Suggested Immunochemistry Markers – Adenocarcinoma Gastric

- CK 7 -
- CK 20 +
- MUC5AC +
- BerEP4 +

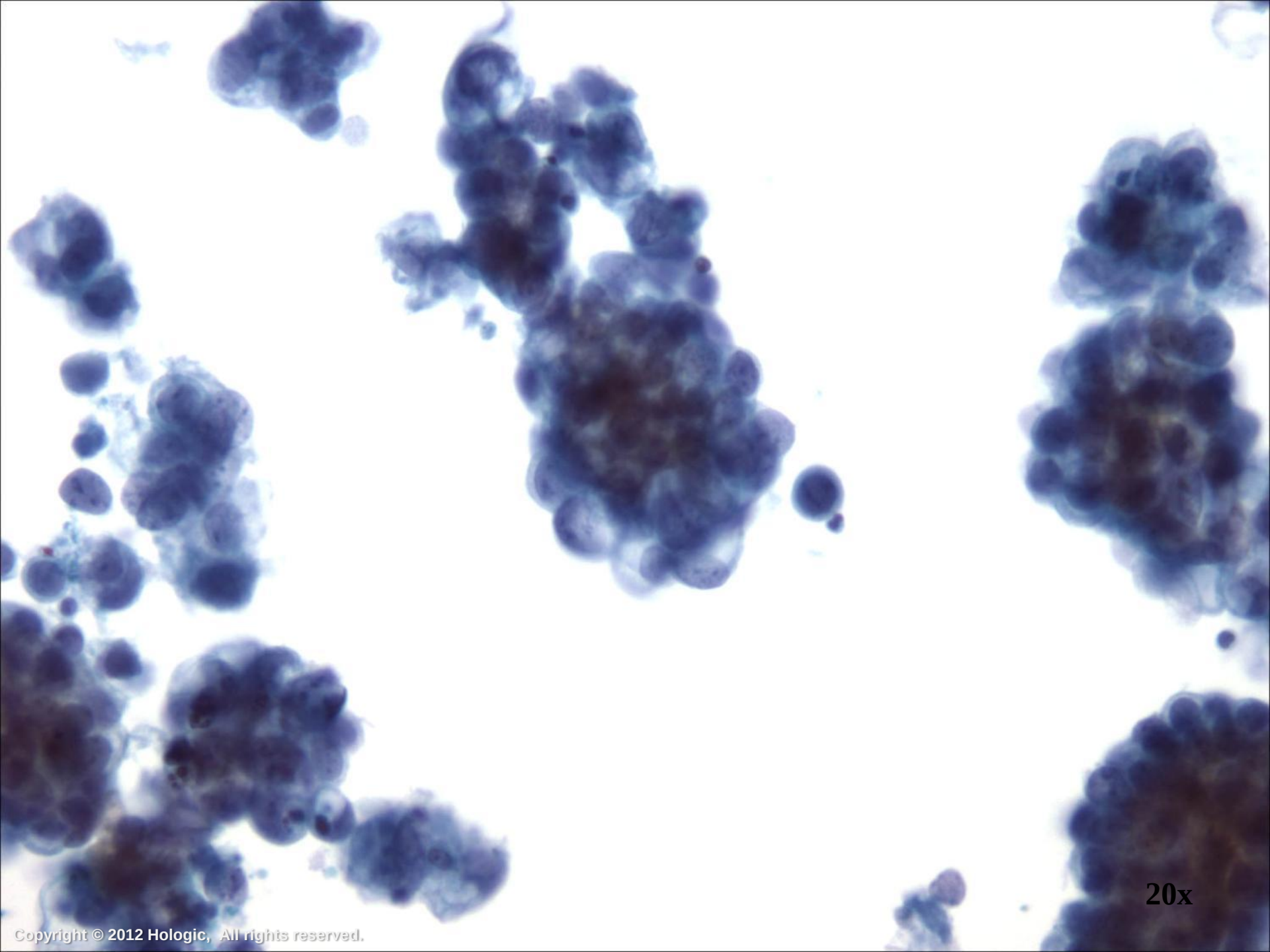
ThinPrep Non-Gyn

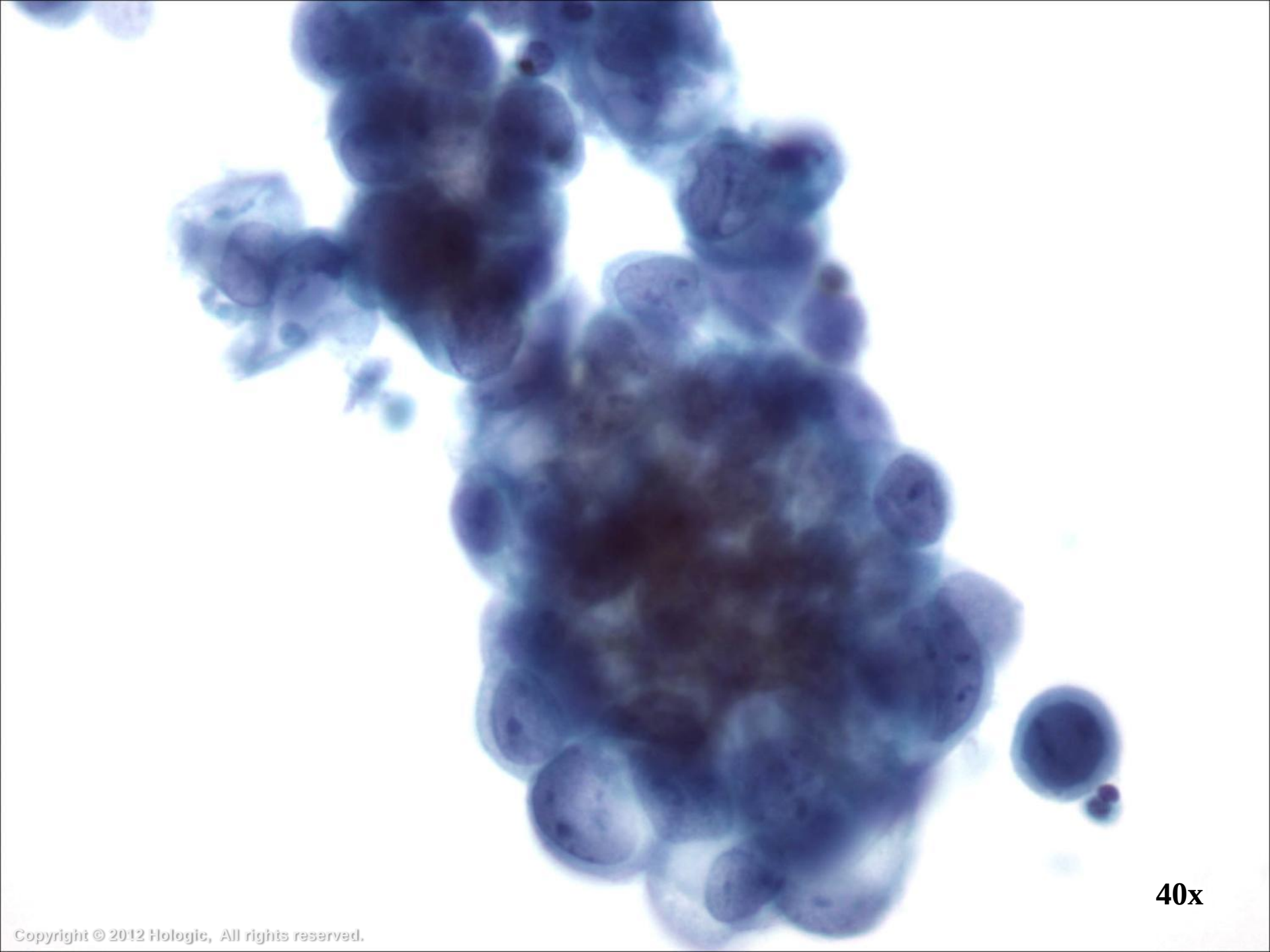
Note – Expected staining results; observed in most but not all cases.

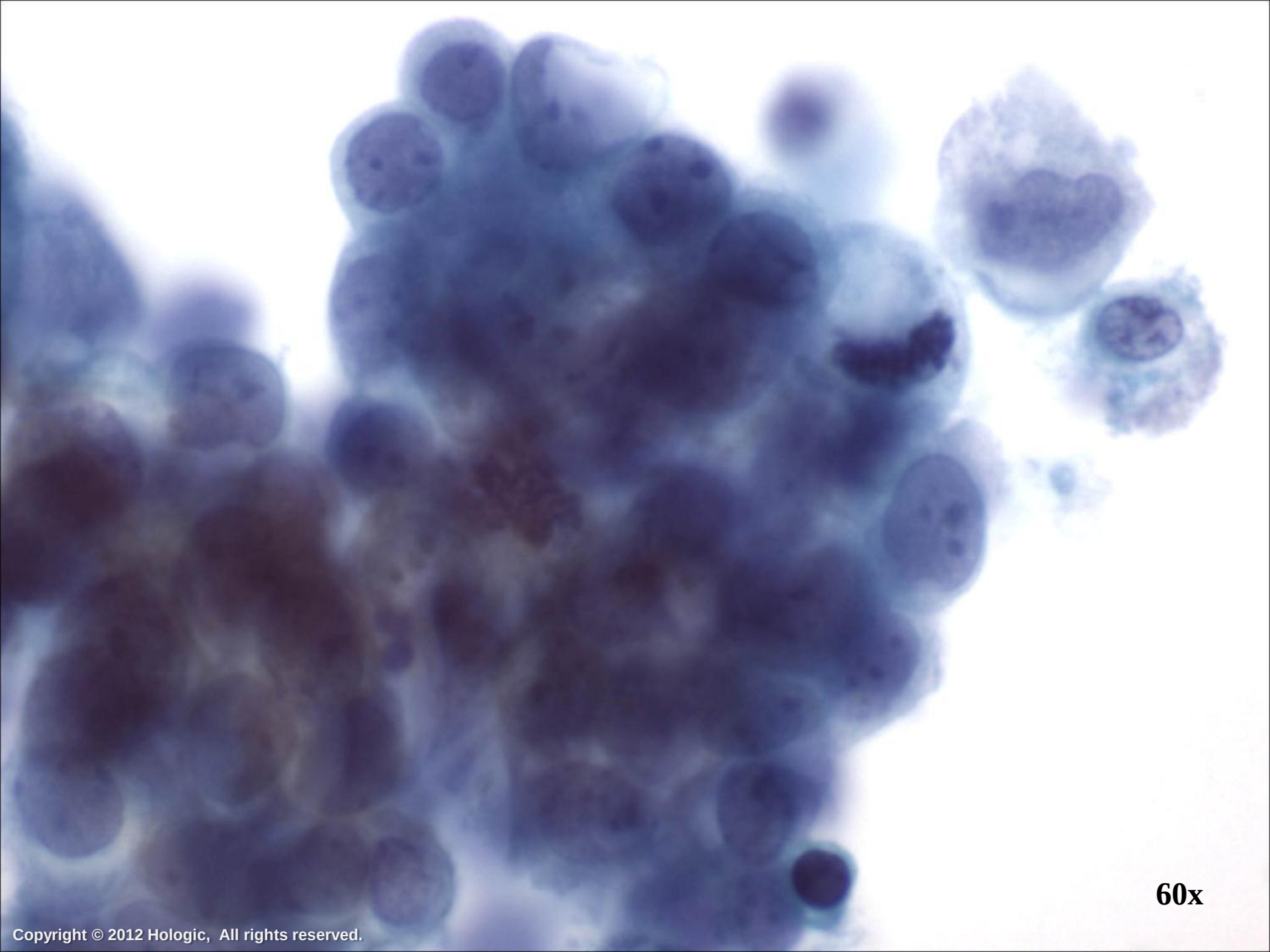
Colorectal Carcinoma

- The following slides contain examples of colorectal carcinoma

ThinPrep Non-Gyn







Suggested Immunochemistry Markers – Adenocarcinoma Colorectal

- CK 7 -
- CK 20 +
- CDX2 +

ThinPrep Non-Gyn

Note – Expected staining results; observed in most but not all cases.

Specific Patterns of Adenocarcinoma

- Pancreatobiliary
  - Cancers of the pancreas and bile ducts are morphologically indistinguishable
  - Cells may have dense cytoplasm similar to non-small cell lung cancer or vacuolated similar to ovarian cancer
  - Single file chains may be seen

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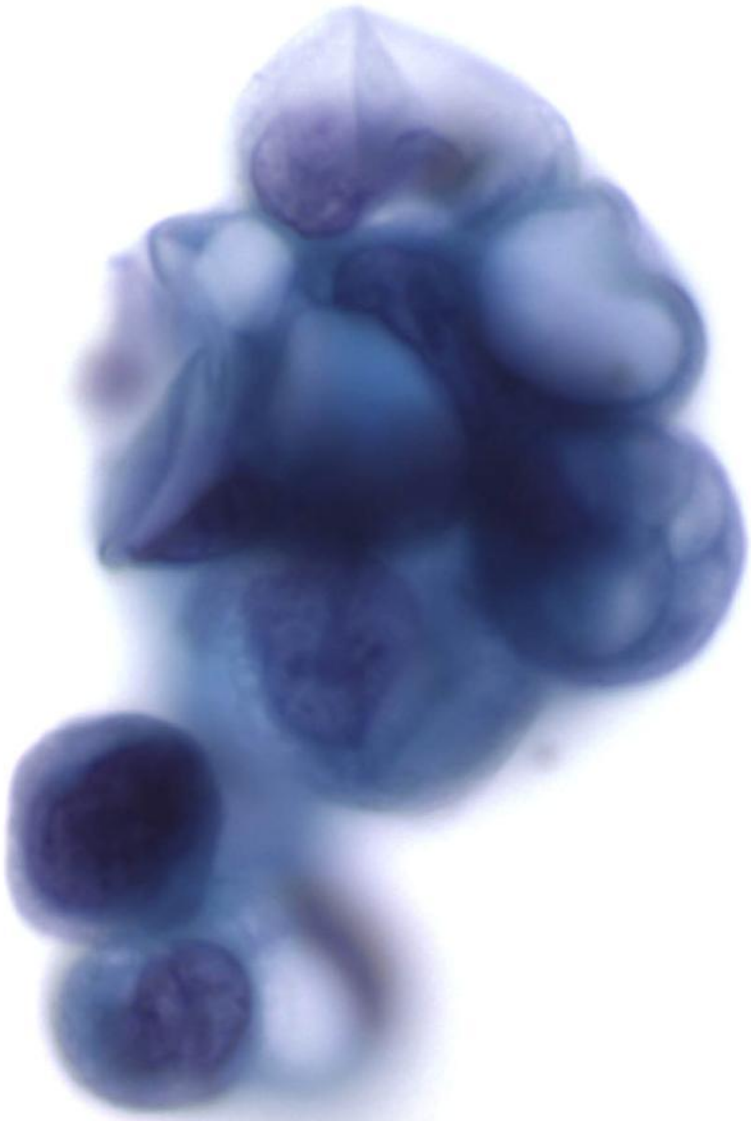
Pancreatobiliary Adenocarcinoma

- The following slides contain images of pancreatobiliary adenocarcinoma

ThinPrep Non-Gyn



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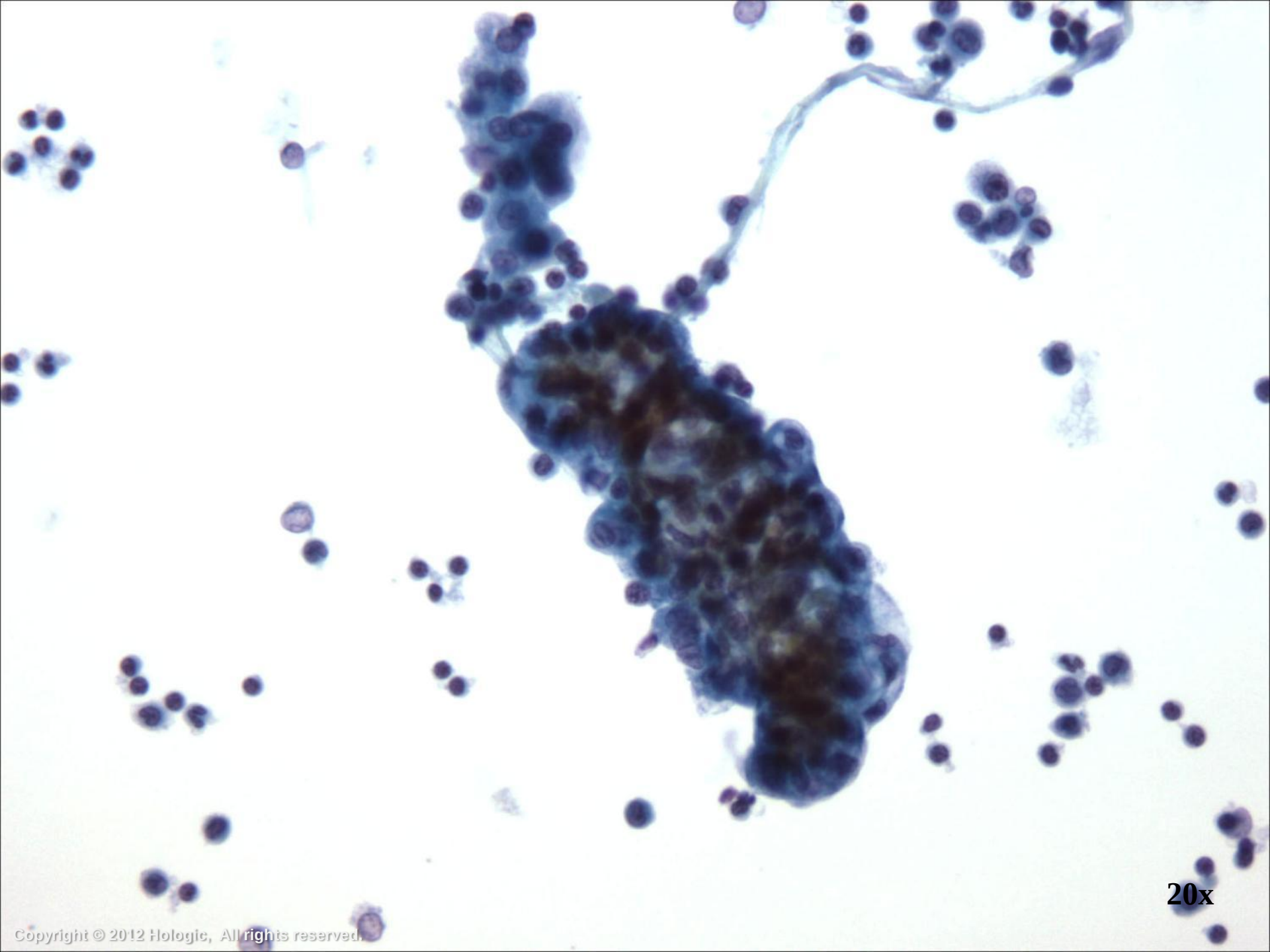

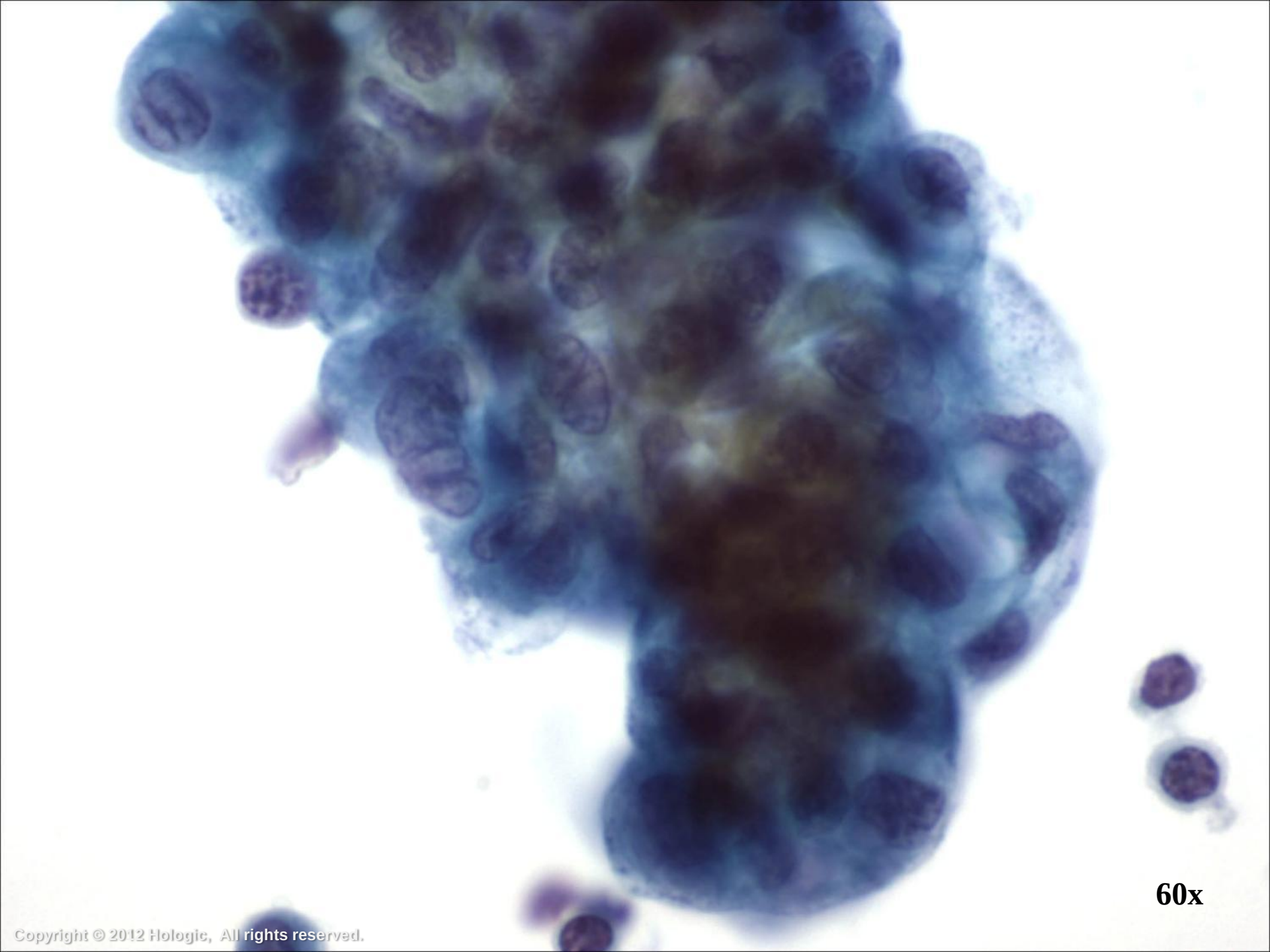

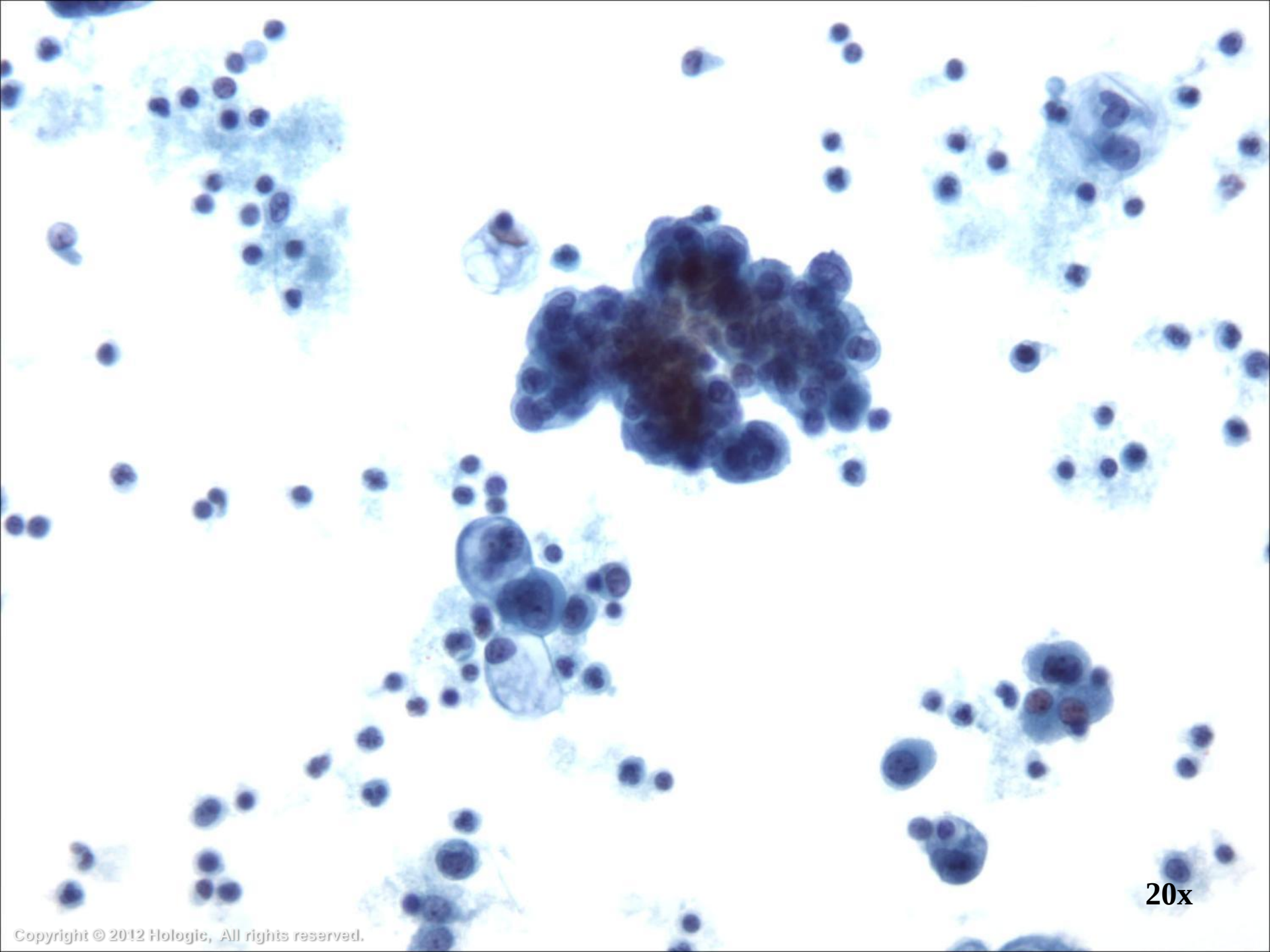

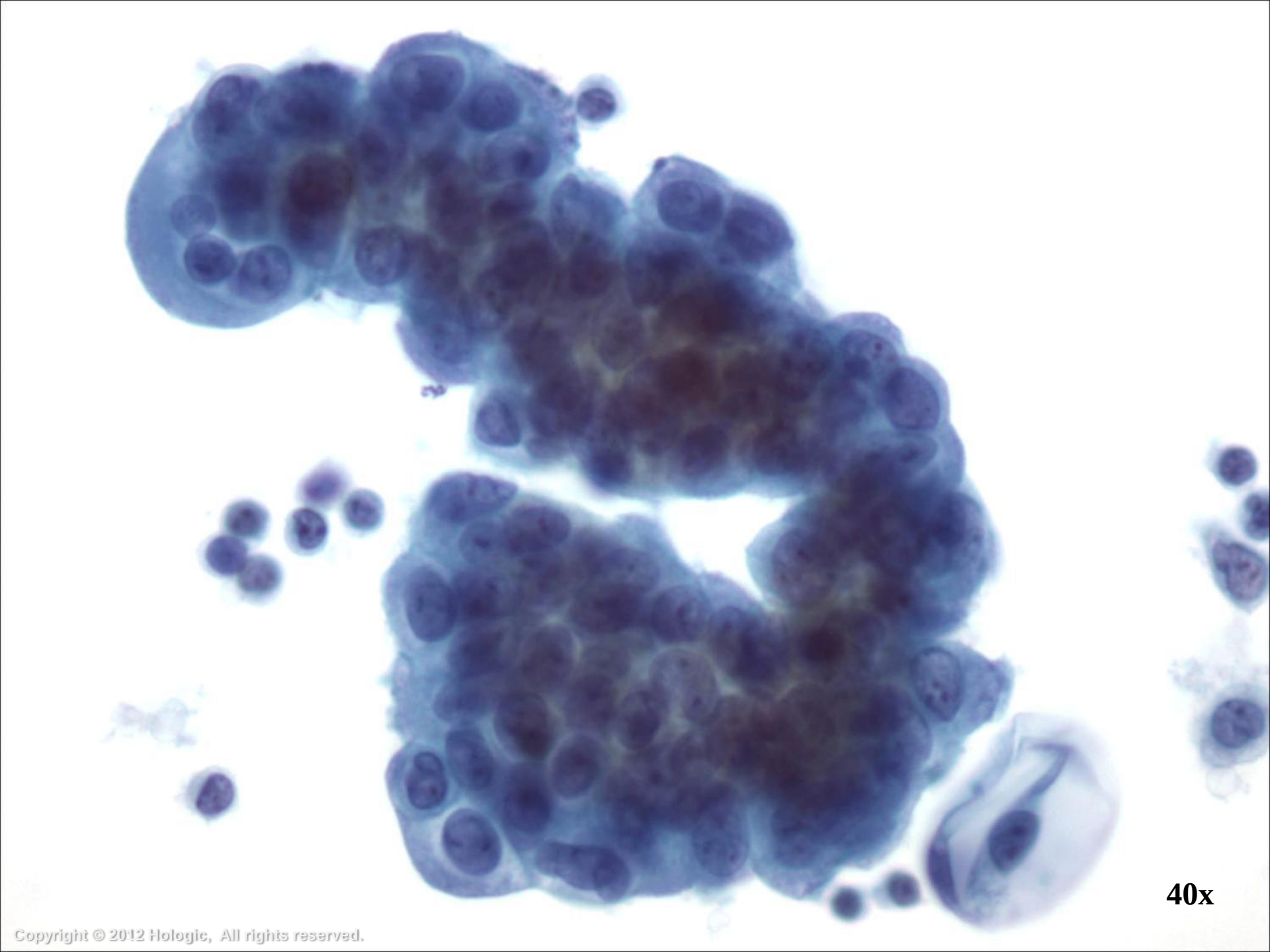

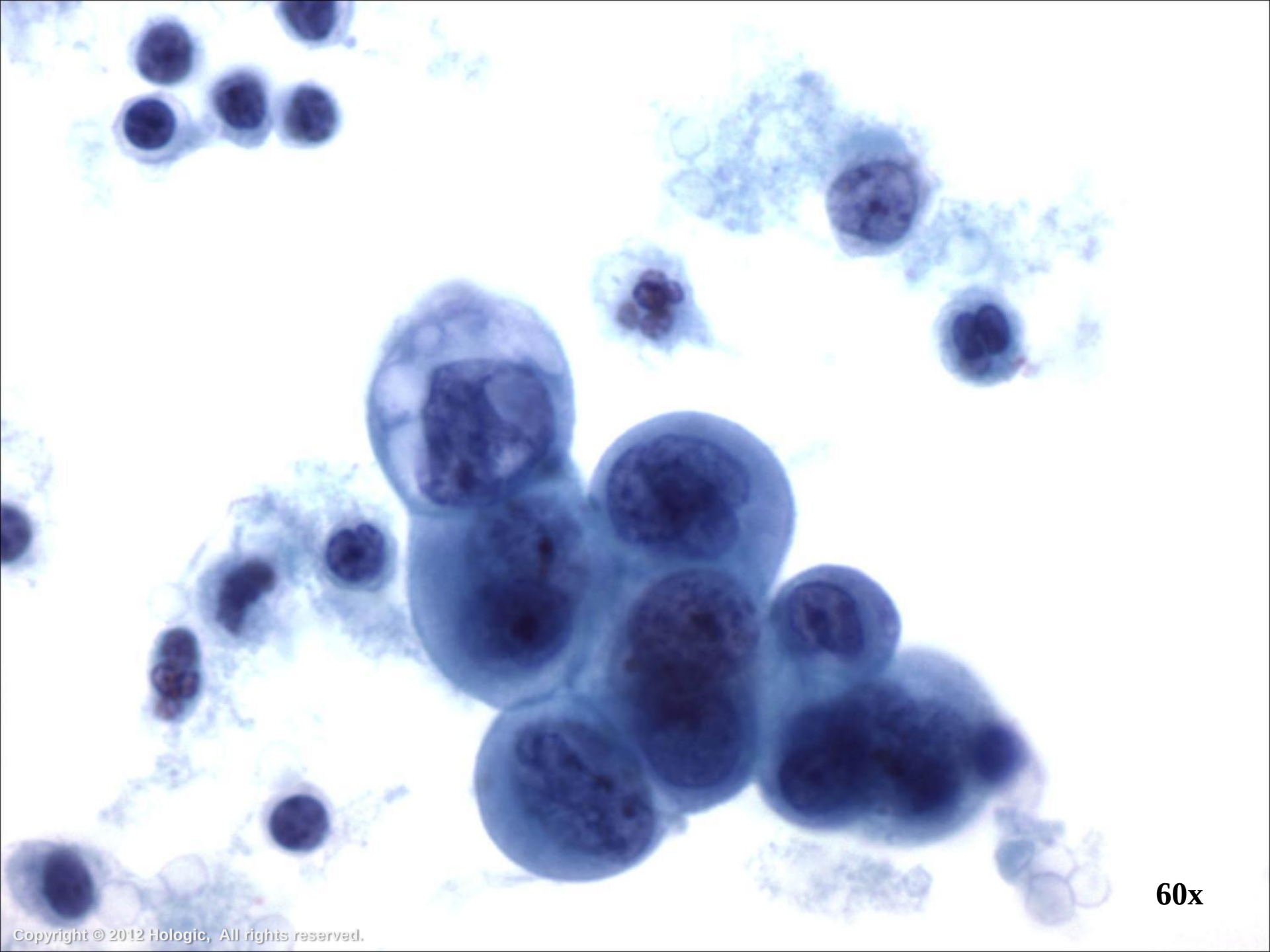

Suggested Immunochemistry Markers – Adenocarcinoma Pancreatobiliary

Pancreatic Adenocarcinoma

- CK 7 -
- CK 20 +
- CK17 +
- MUC1 +

Cholangiocarcinoma

- CK 7 +
- CK 20 +/-
- MUC5AC +

ThinPrep Non-Gyn

Note – Expected staining results; observed in most but not all cases.

Specific Patterns of Adenocarcinoma

- Ovarian
  - 3 types
    - Mucinous
    - Serous
    - Endometrioid
  - Most often characterized by cells with large, transparent vacuoles
  - Presence of psammoma bodies may suggest ovarian origin but not diagnostic of malignancy

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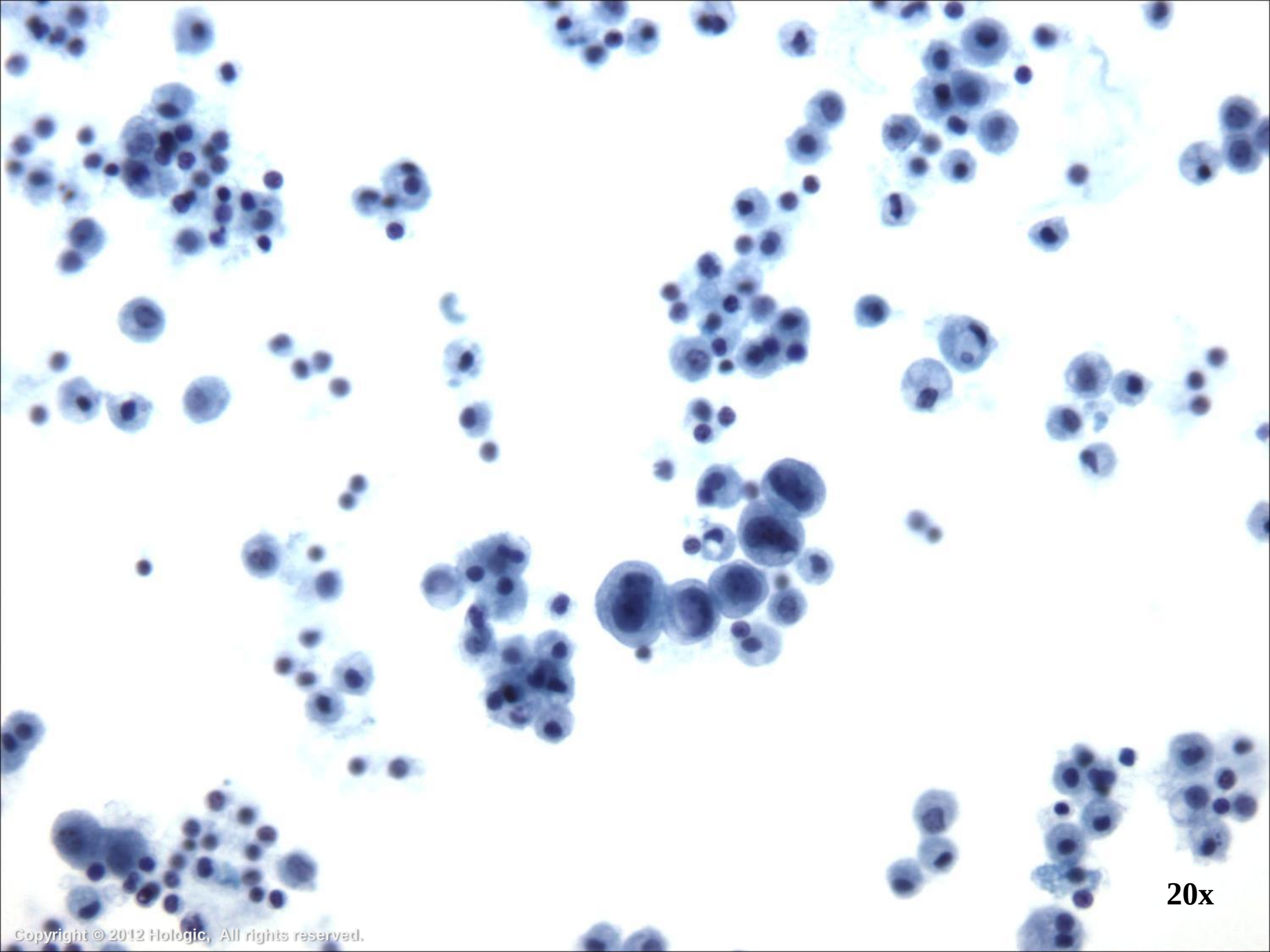

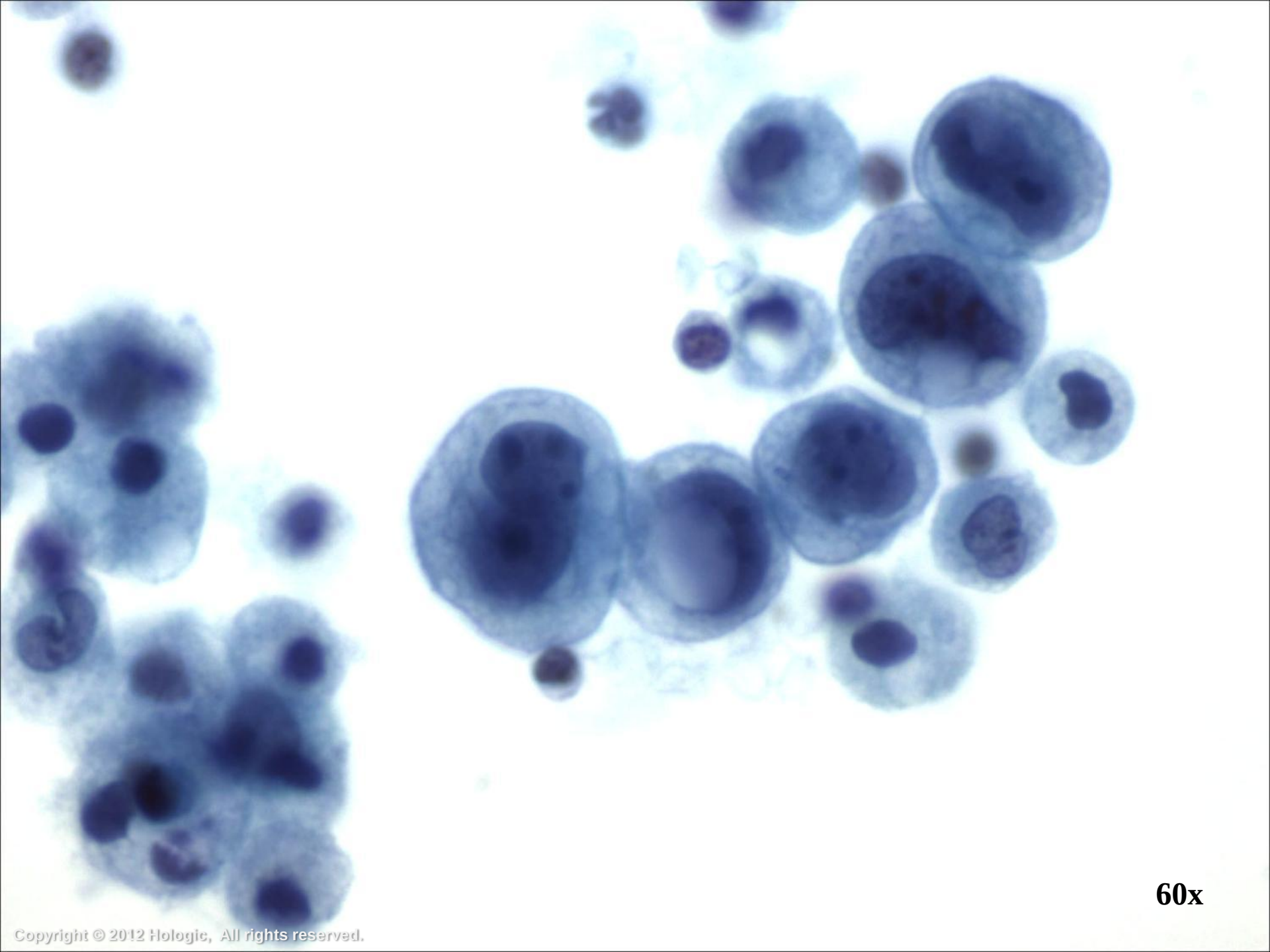

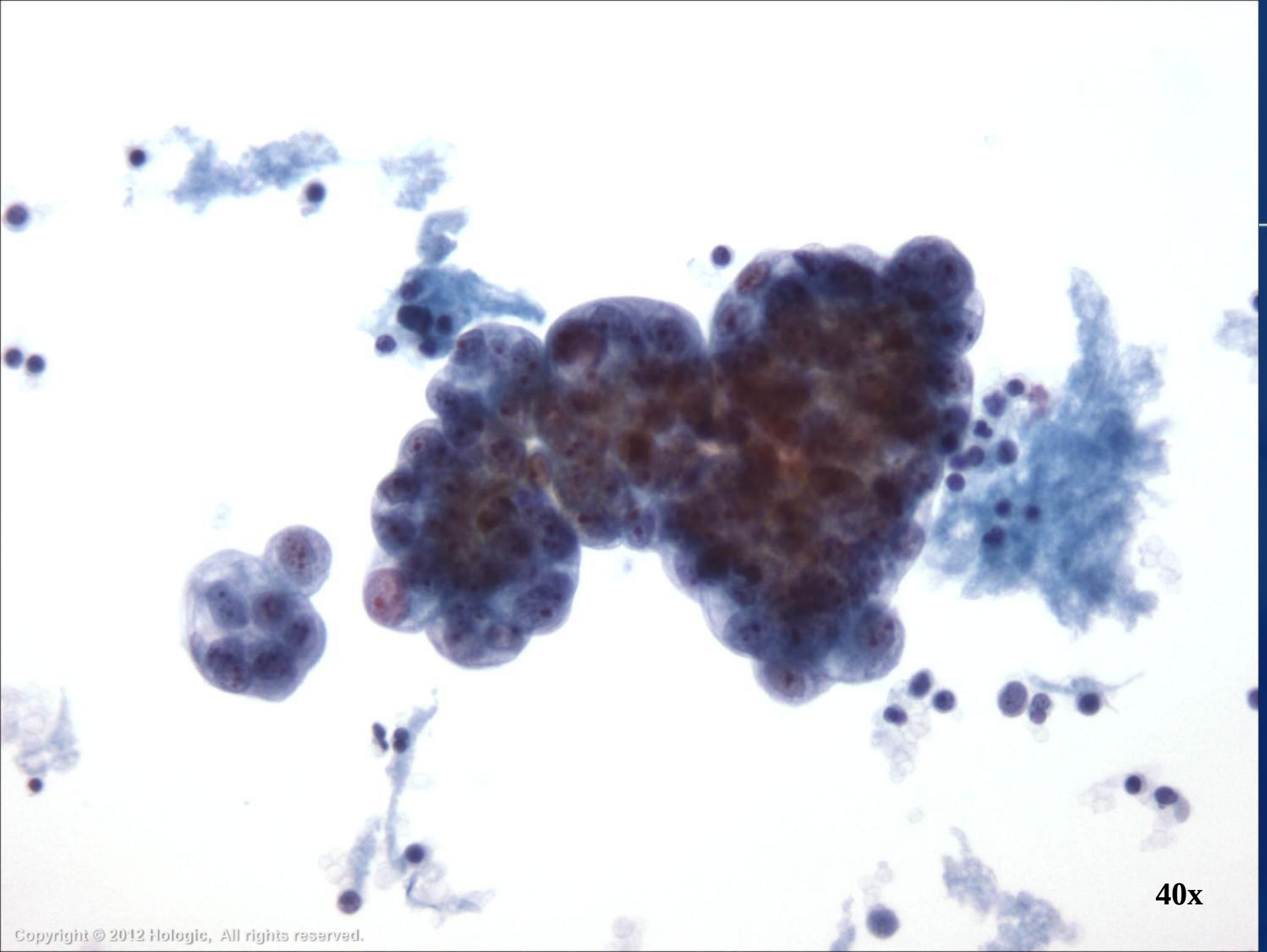

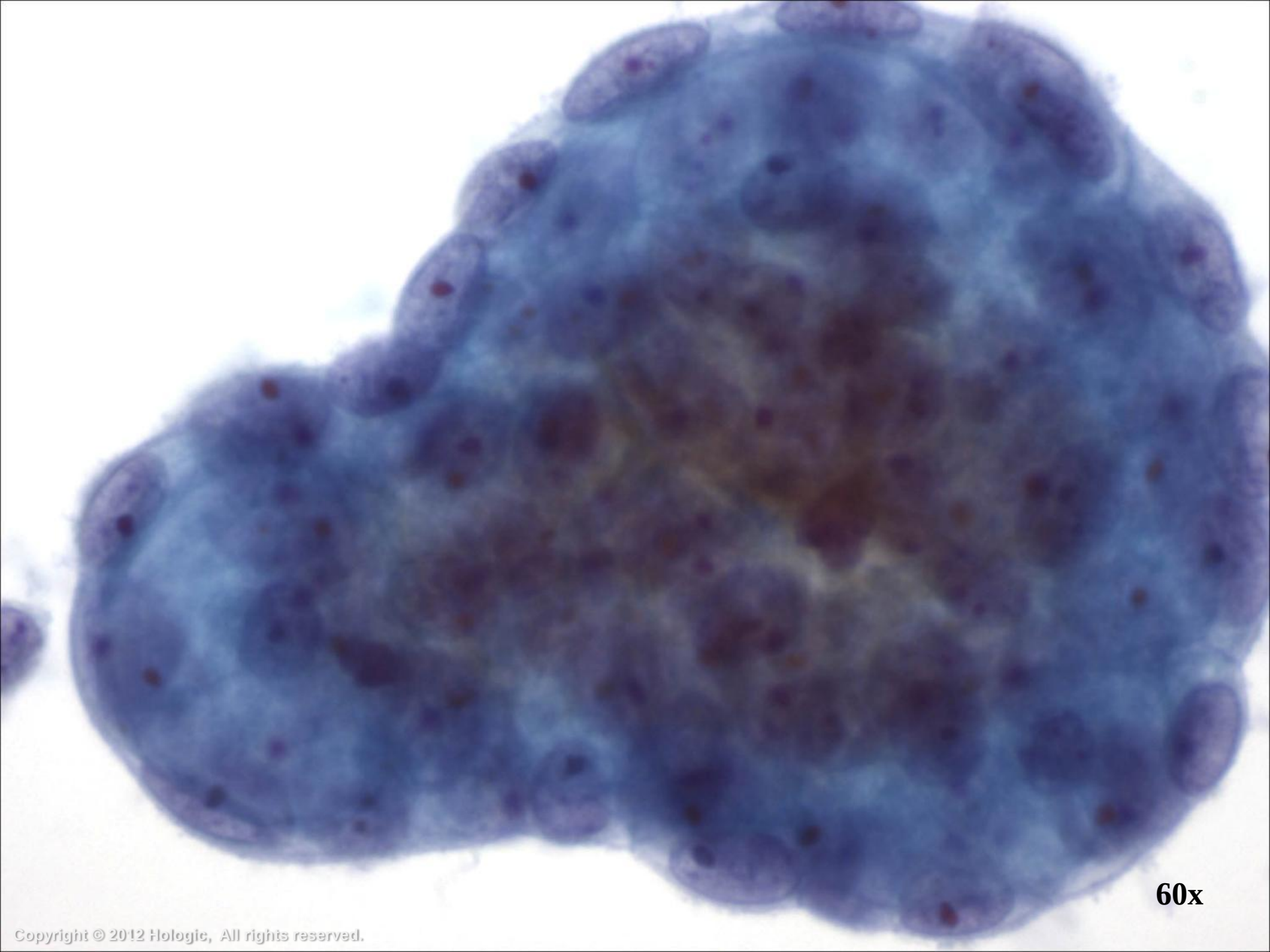

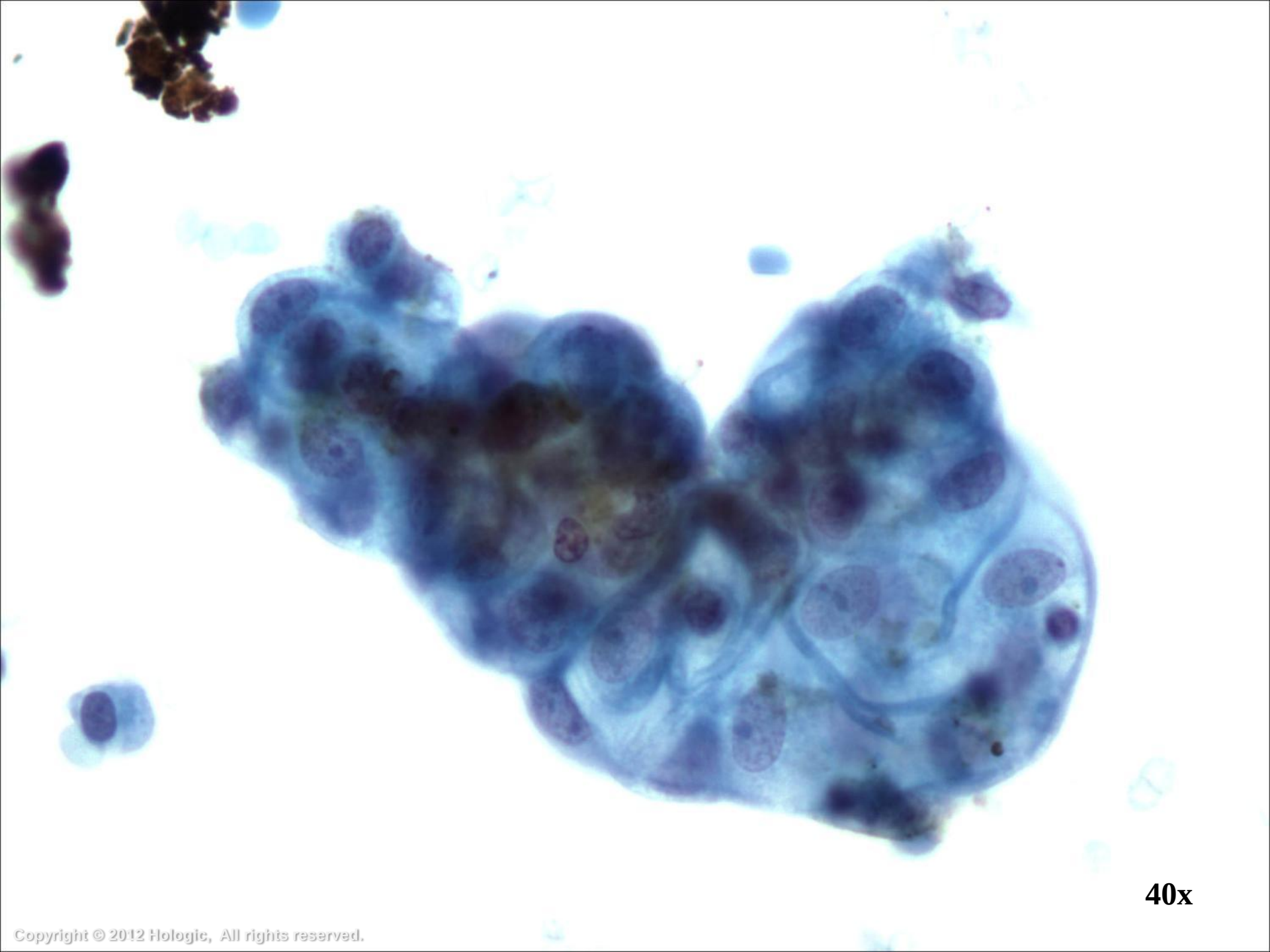

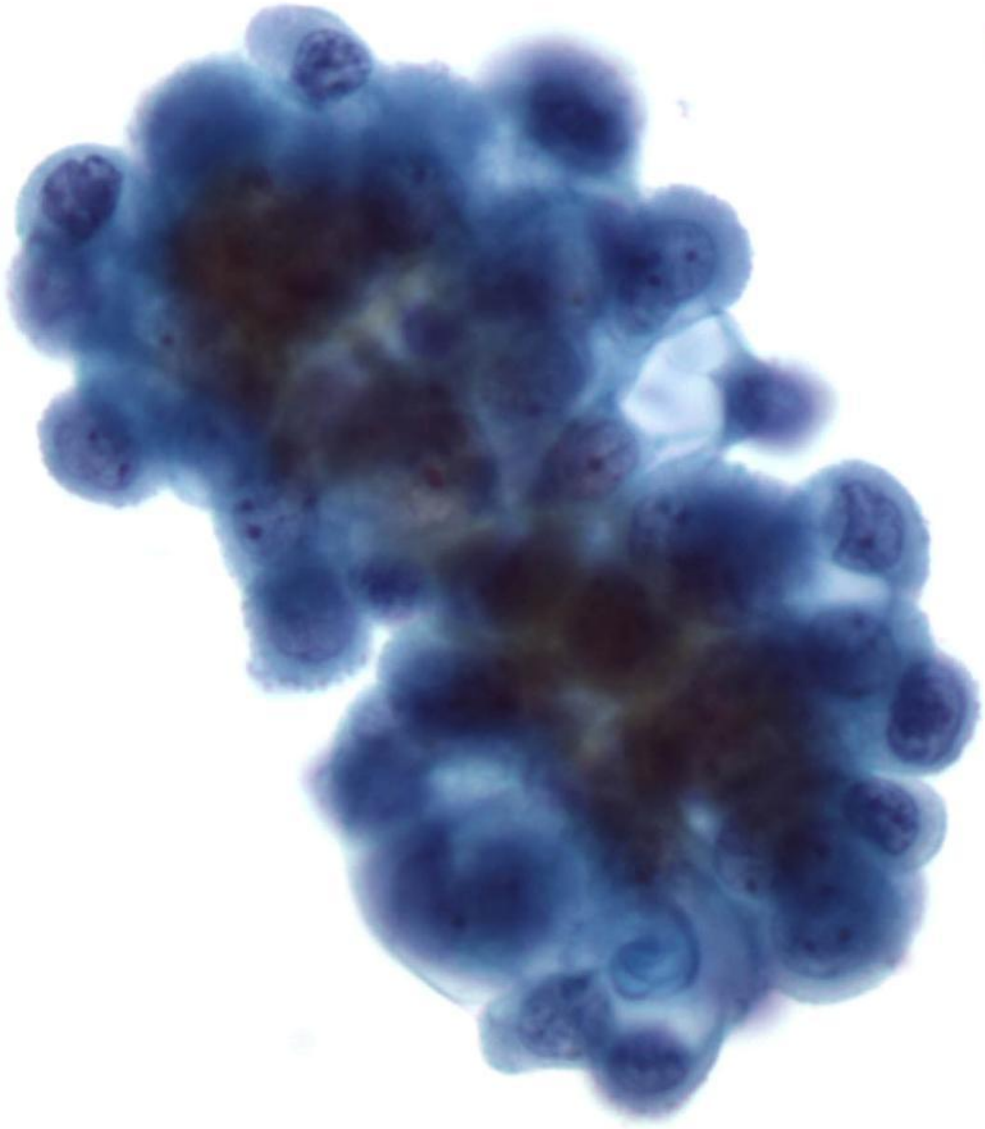

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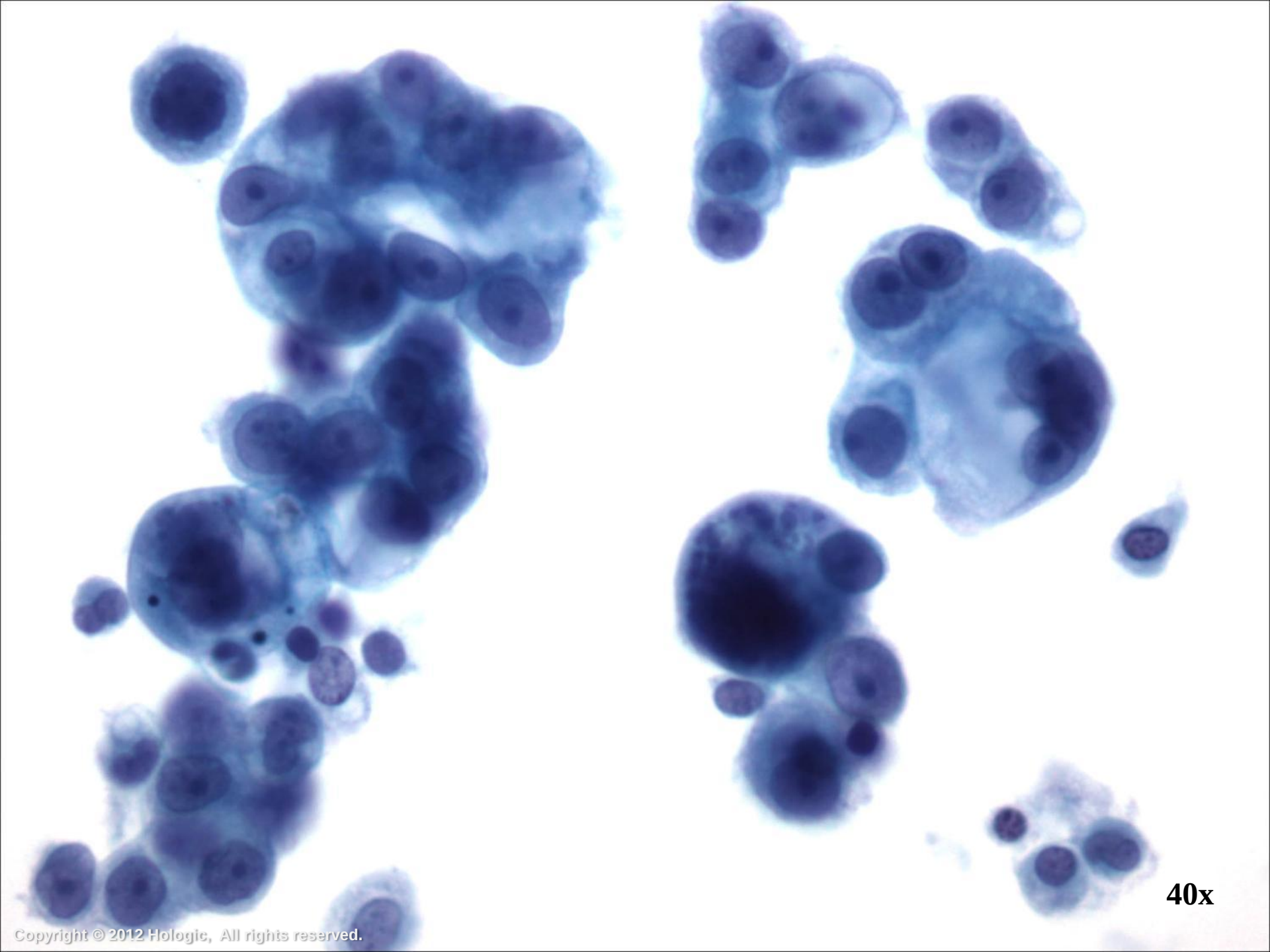

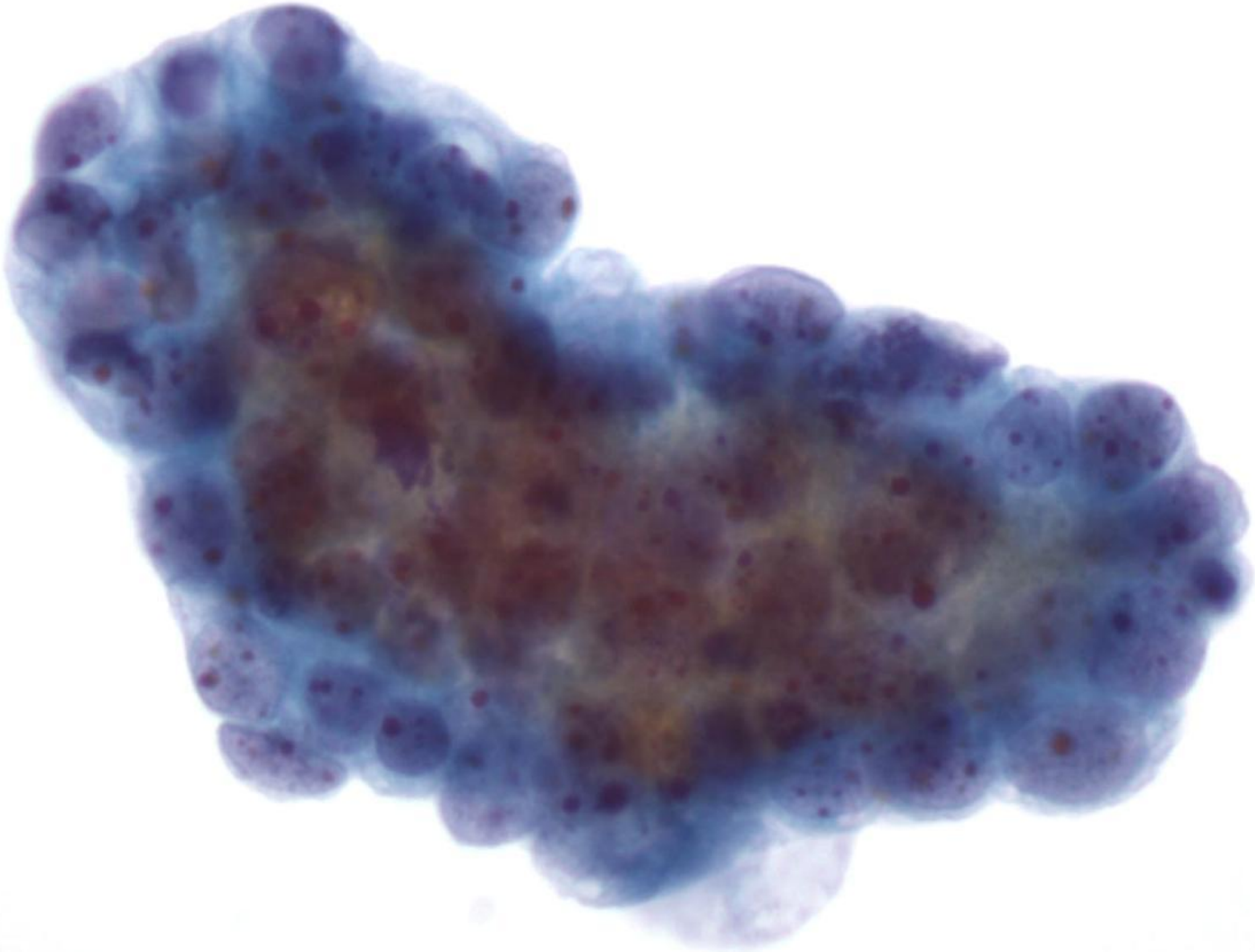

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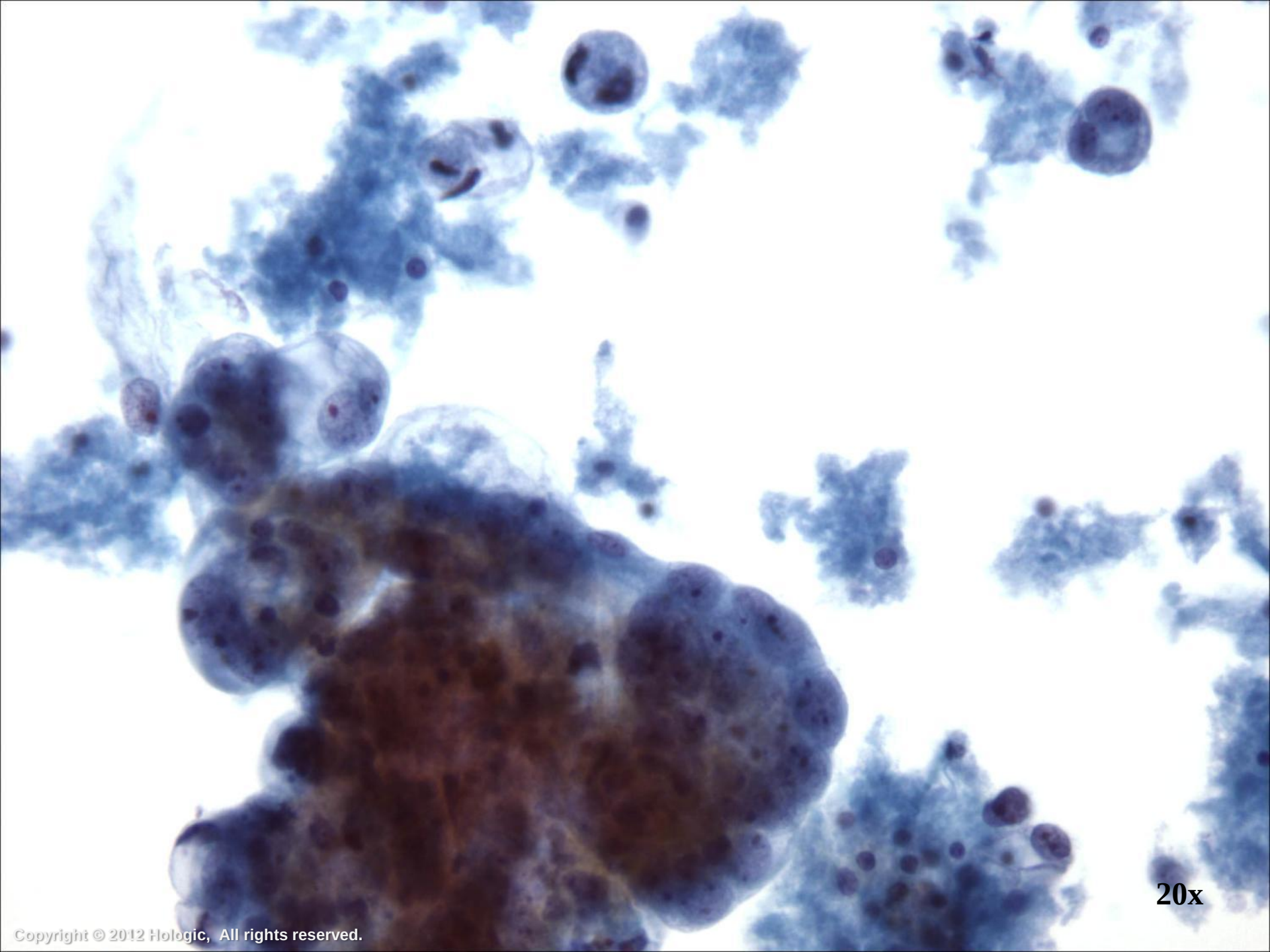

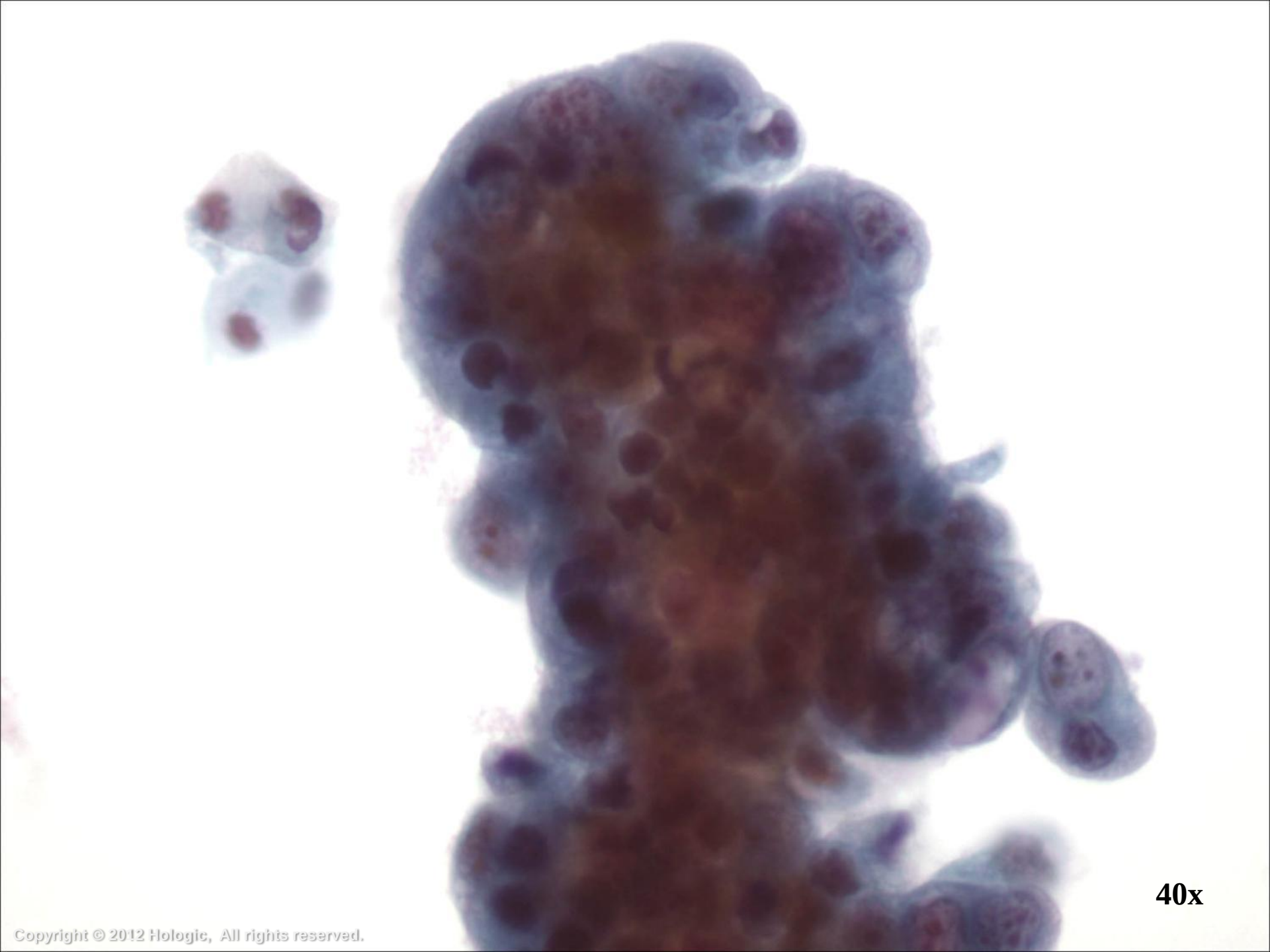

Suggested Immunochemistry Markers – Adenocarcinoma Ovary

Mucinous Adenocarcinoma

- CK 7 +
- CK 20 +
- BerEP4 +
- CA 125 +

Non-mucinous Adenocarcinoma

- CK 7 +
- CK 20 -
- ER/PR +
- CEA -

ThinPrep Non-Gyn

Note – Expected staining results; observed in most but not all cases.

Specific Patterns of Adenocarcinoma

- Endometrial
  - Papillary clusters or single malignant cells
  - Delicate cytoplasm that is scant to abundant
  - Coarse chromatin
  - Macronucleoli
  - Psammoma bodies may be seen

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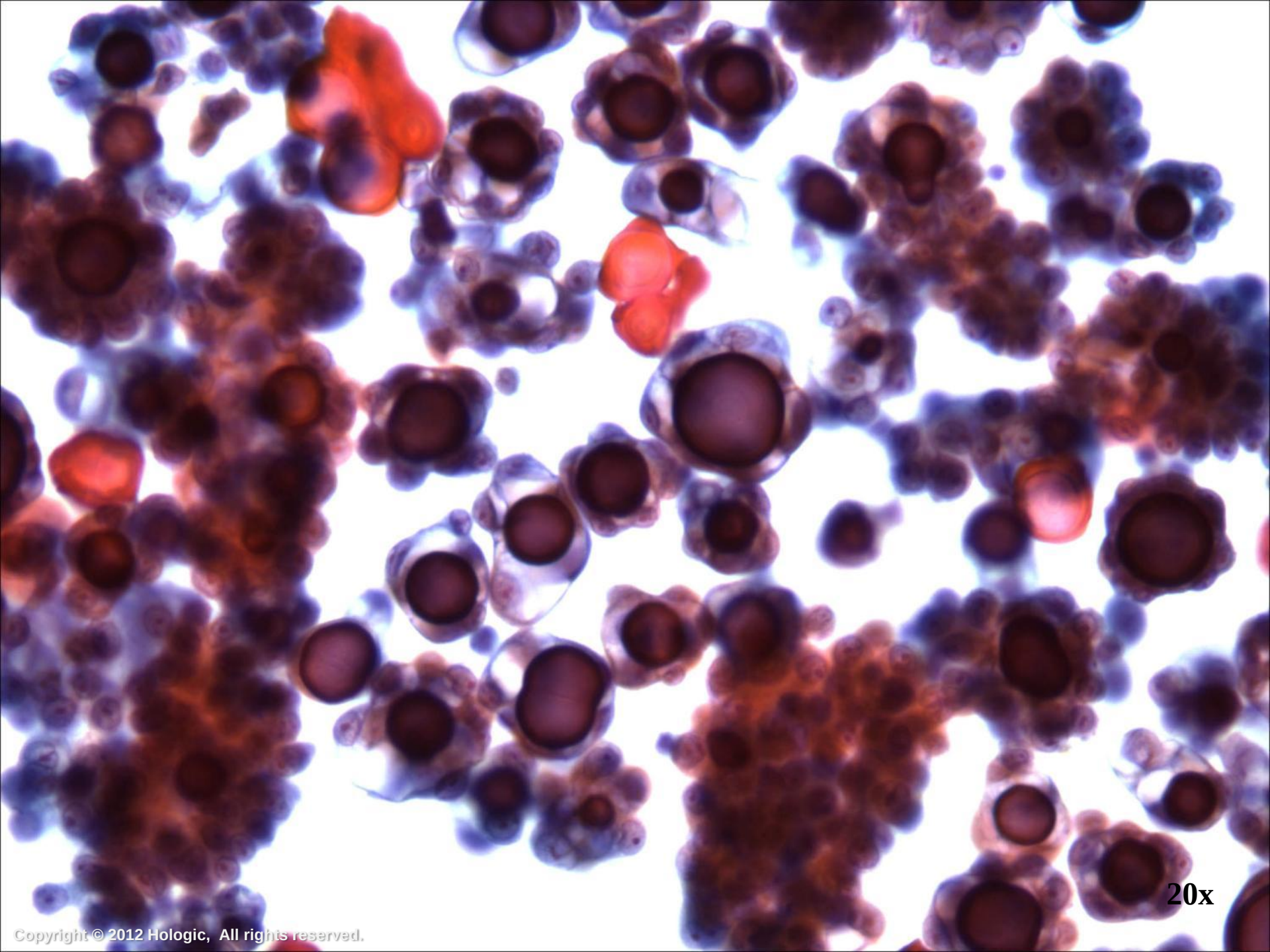

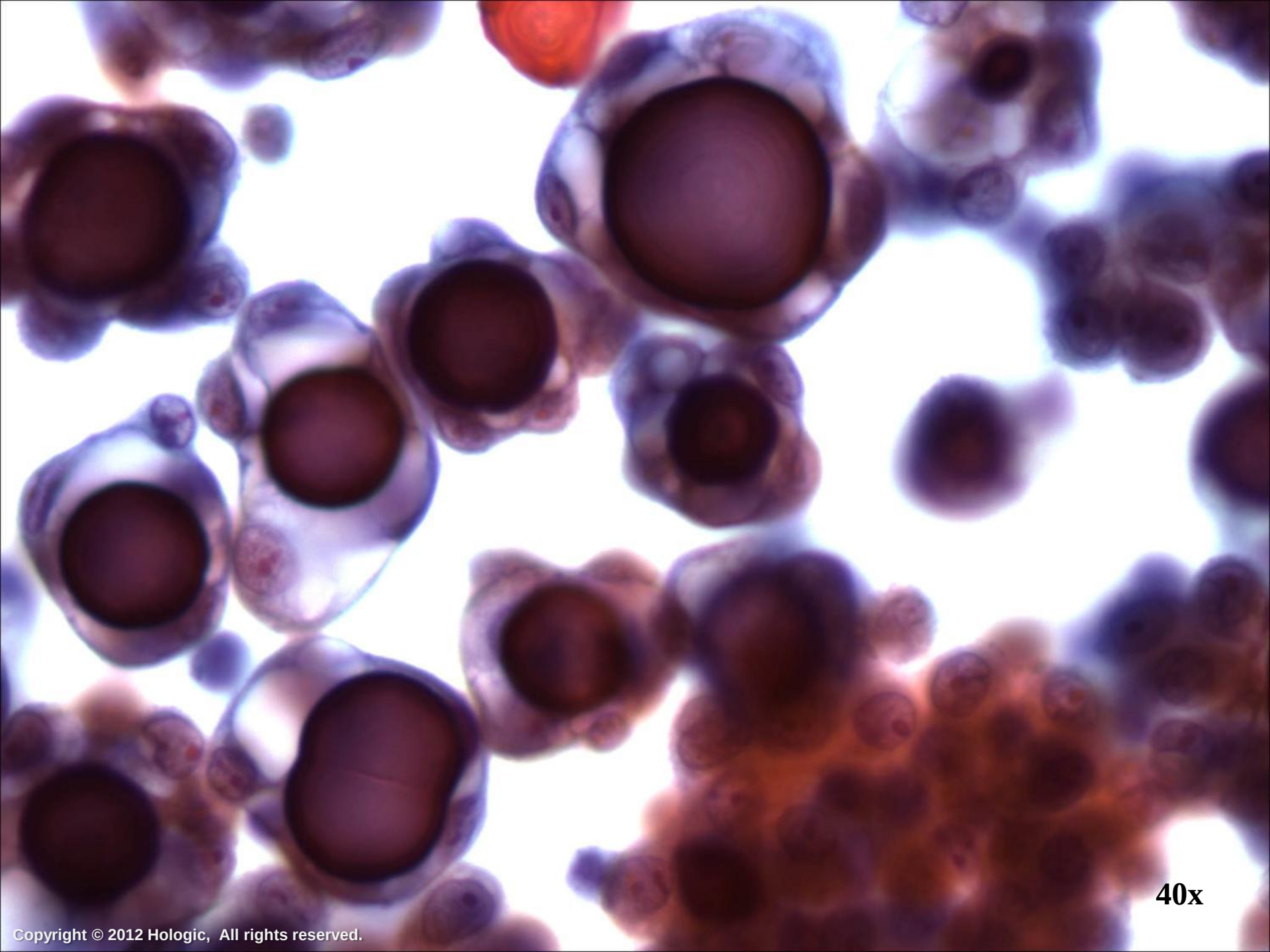

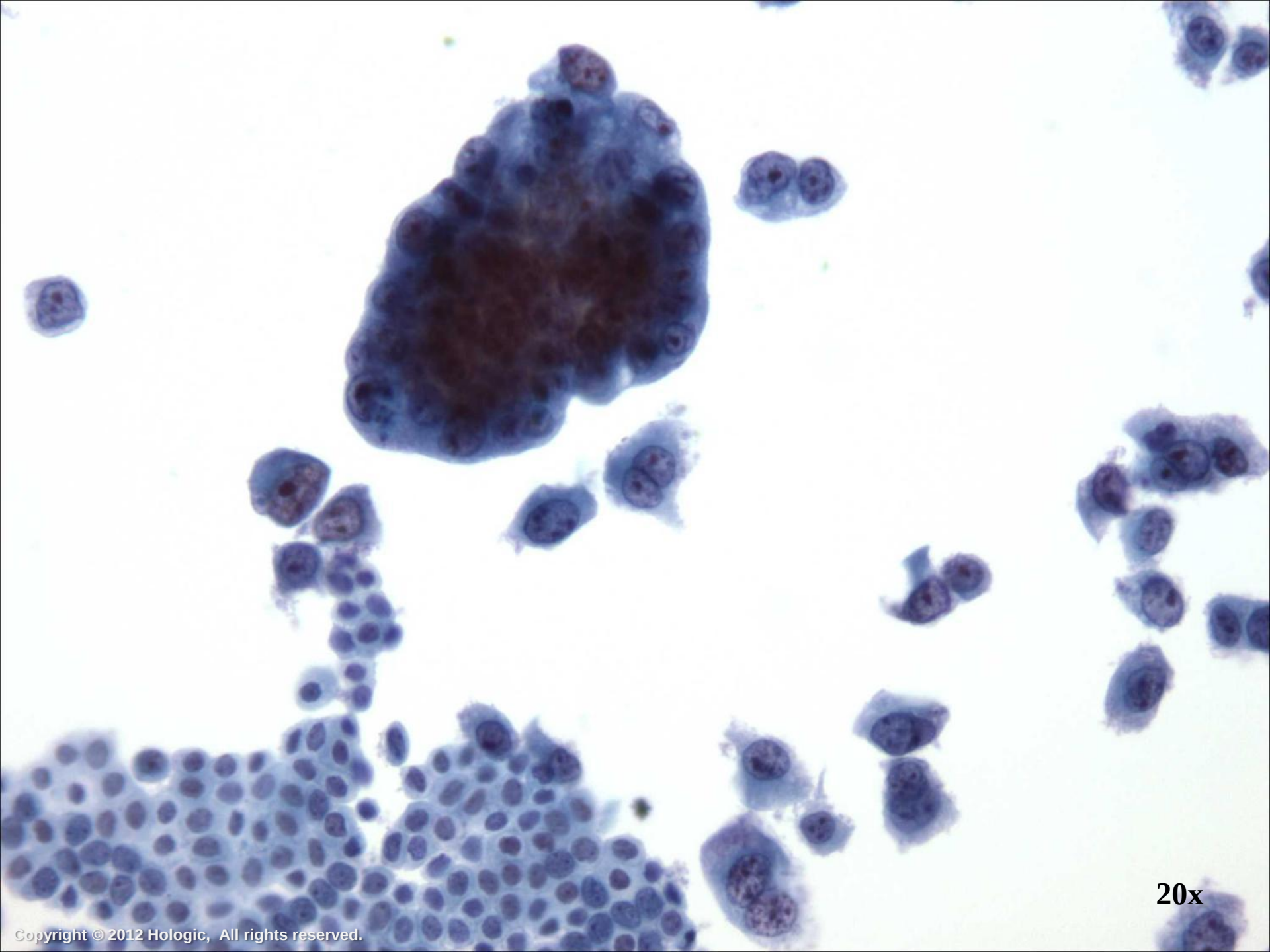

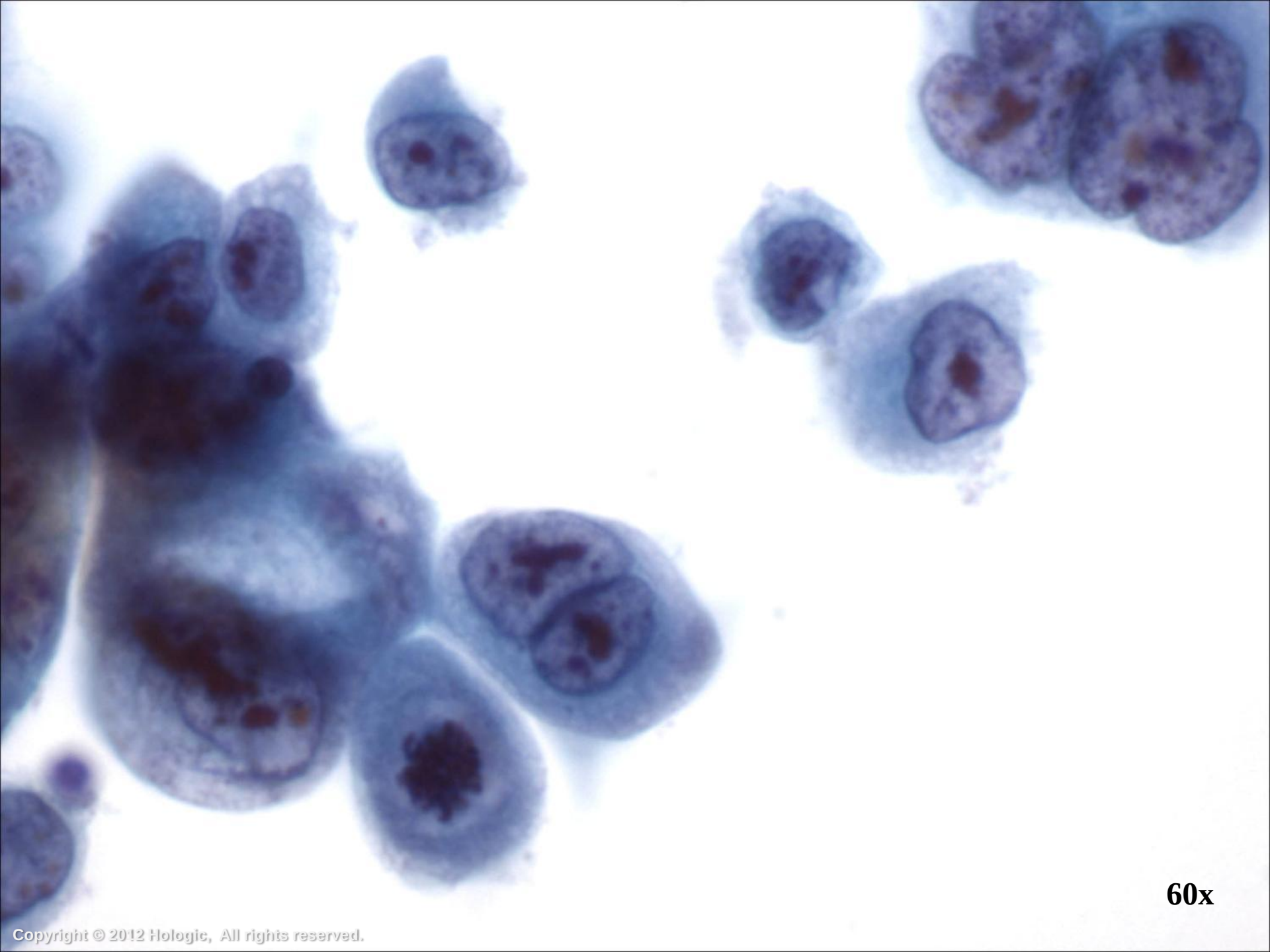

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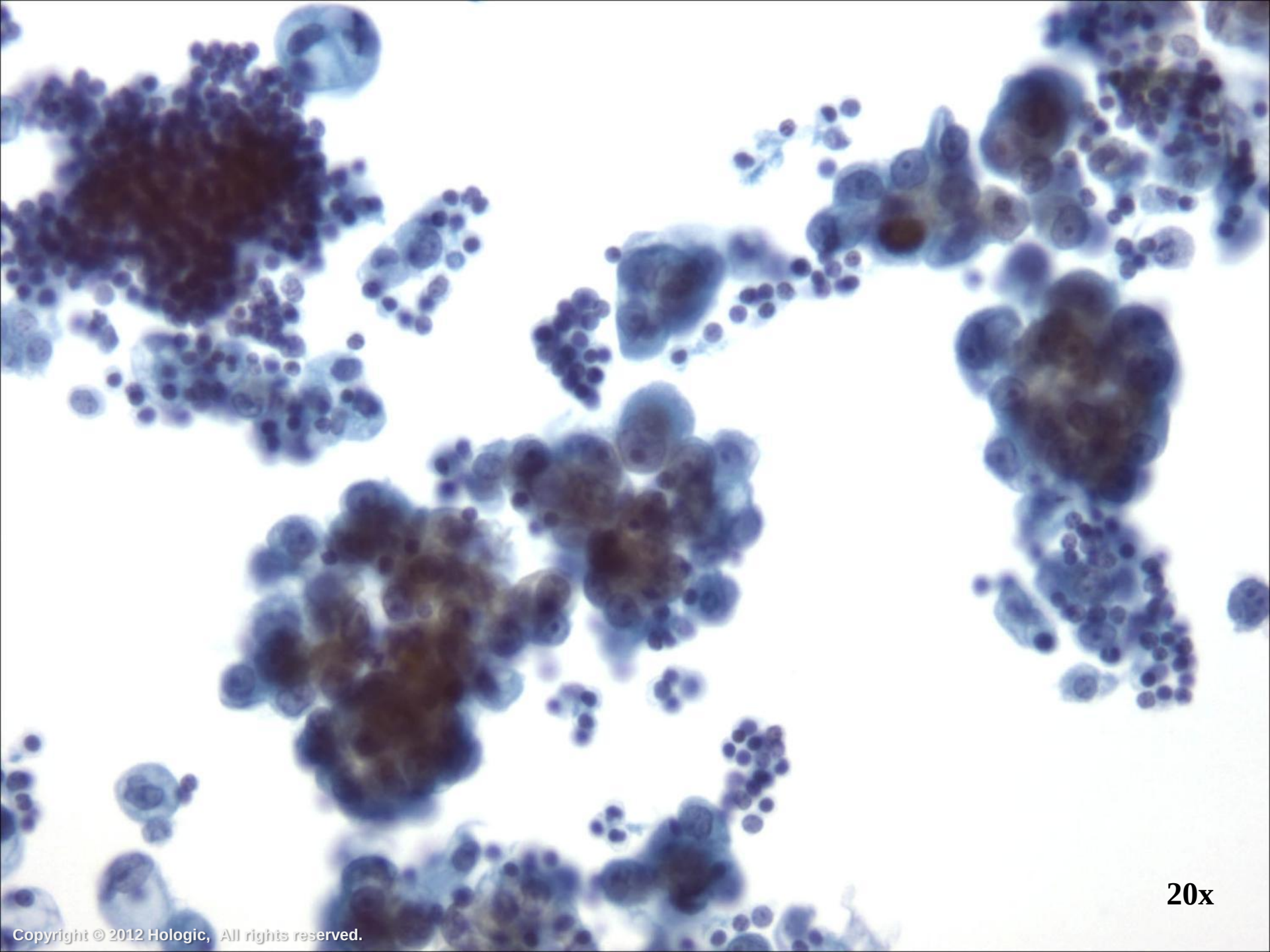

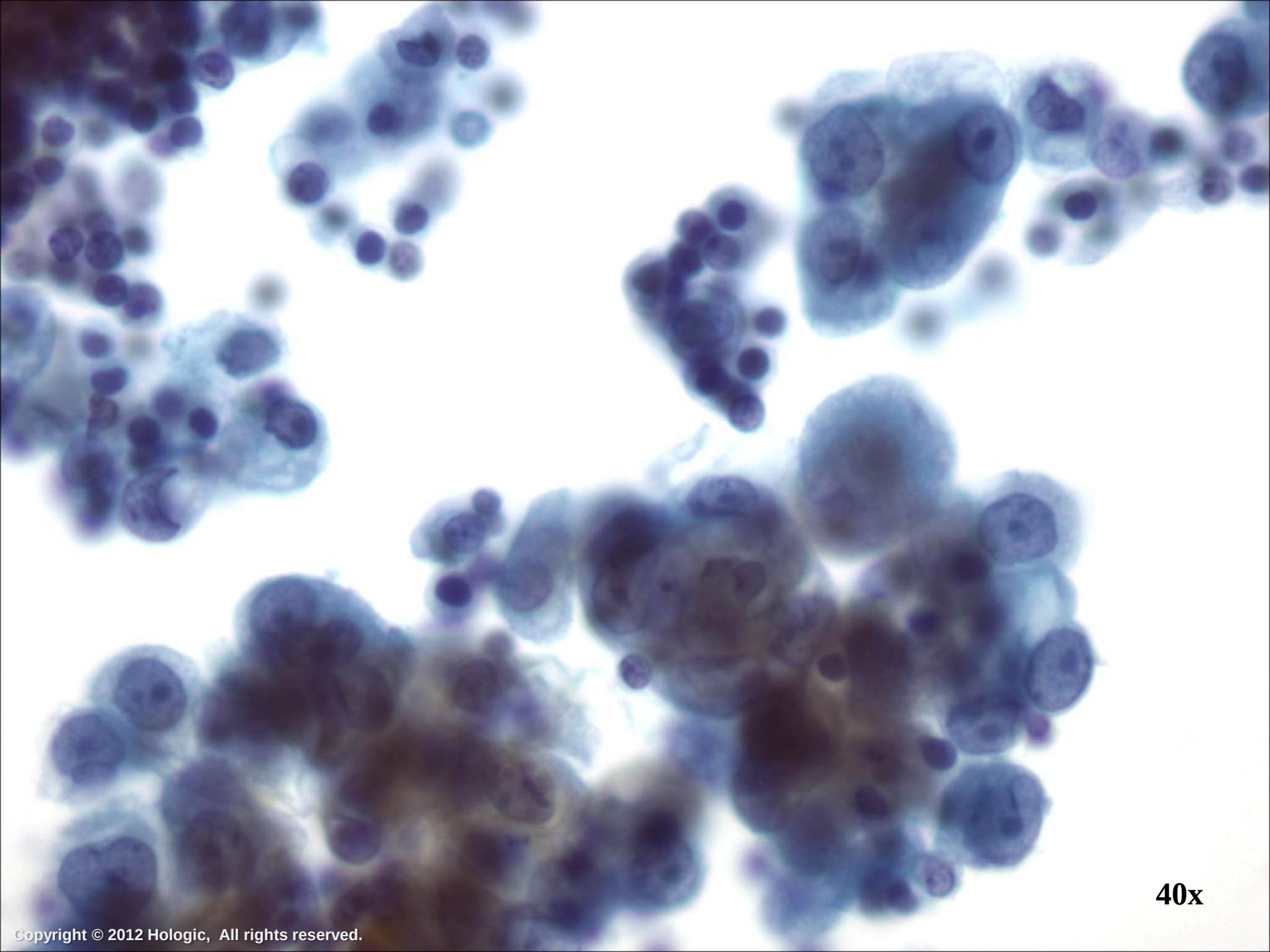

Suggested Immunochemistry Markers – Adenocarcinoma Endometrium

- CK 7 +
- CK 20 -
- Vimentin +
- B72.3 +

ThinPrep Non-Gyn

Note – Expected staining results; observed in most but not all cases.

Squamous Cell Carcinoma

- Cytologic features:
  - Large fragments and/or single cells
  - Cytoplasm may be dense and occasionally orangeophilic
  - Hyperchromatic nuclei
  - Coarse chromatin
  - Nucleoli not prominent

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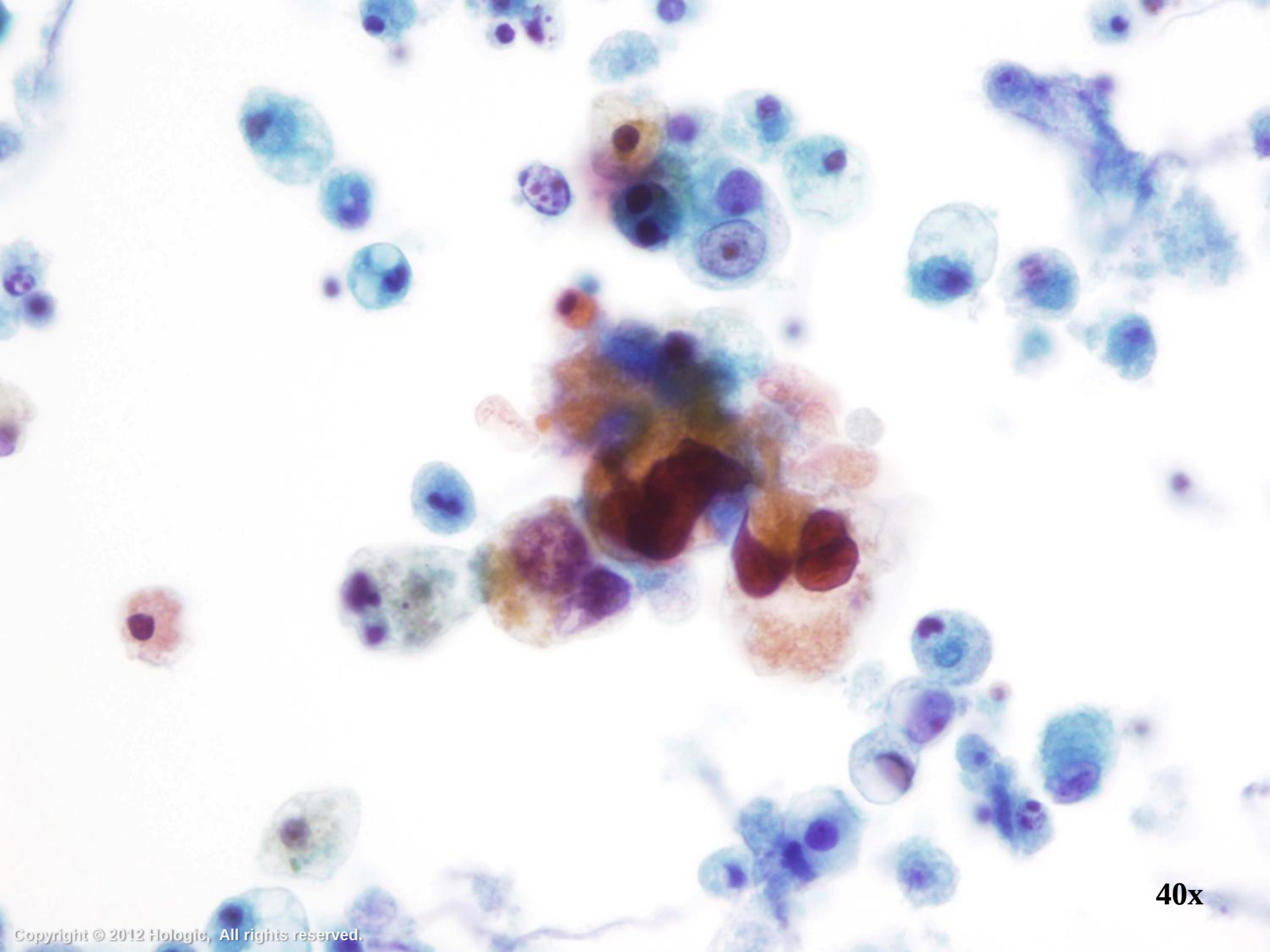

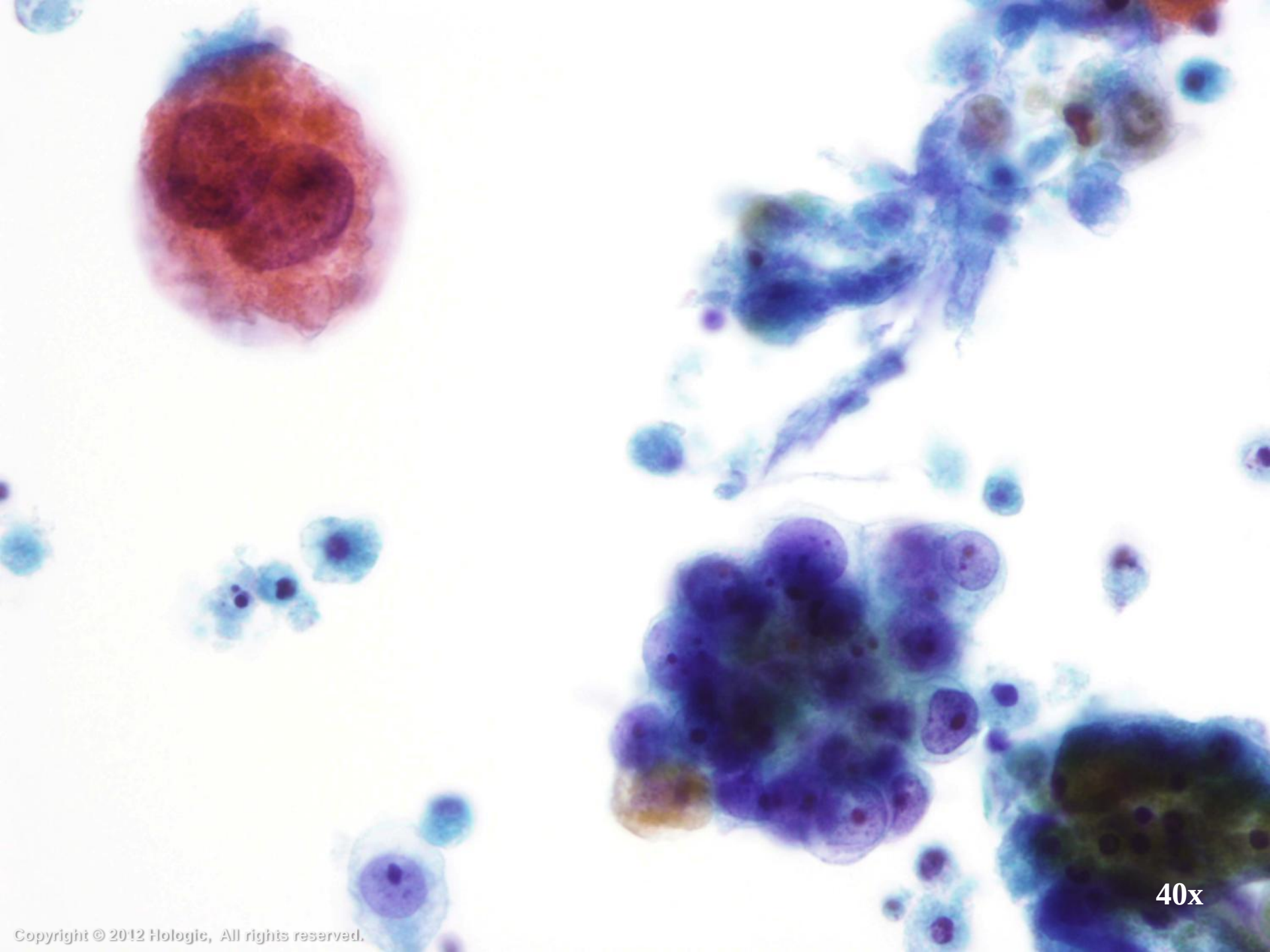

Suggested Immunochemistry Markers – Squamous Carcinoma

- CK 5/6 +
- CEA +
- p63* +

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* p63 staining is especially useful in non-keratinizing SCC

Note – Expected staining results; observed in most but not all cases.

Small Cell Carcinoma

- Cytologic features:
  - Tissue aggregates with molding
  - Single file arrangements can be seen
  - Small, rounded cells
  - Coarse chromatin
  - Inconspicuous nucleoli

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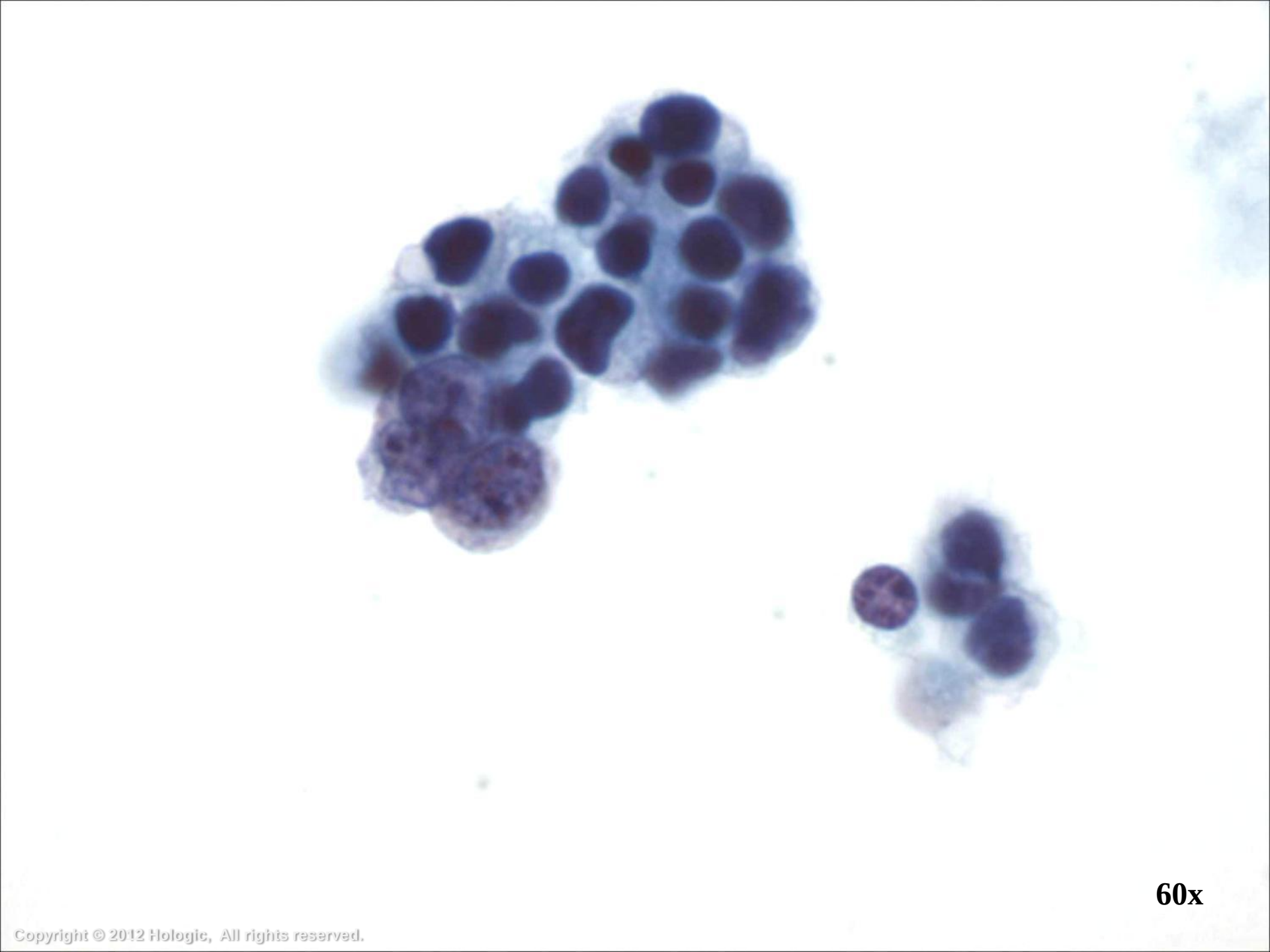

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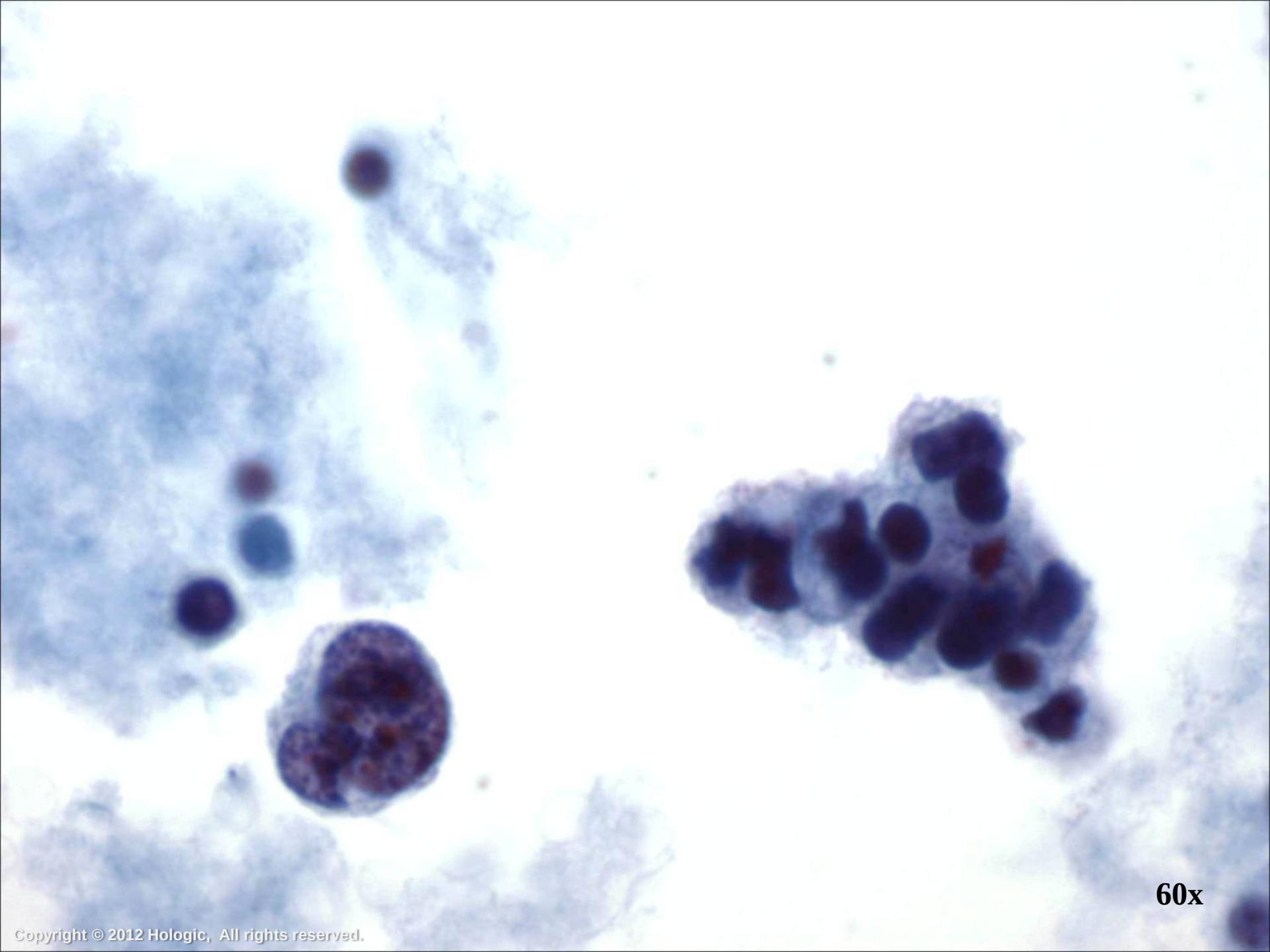

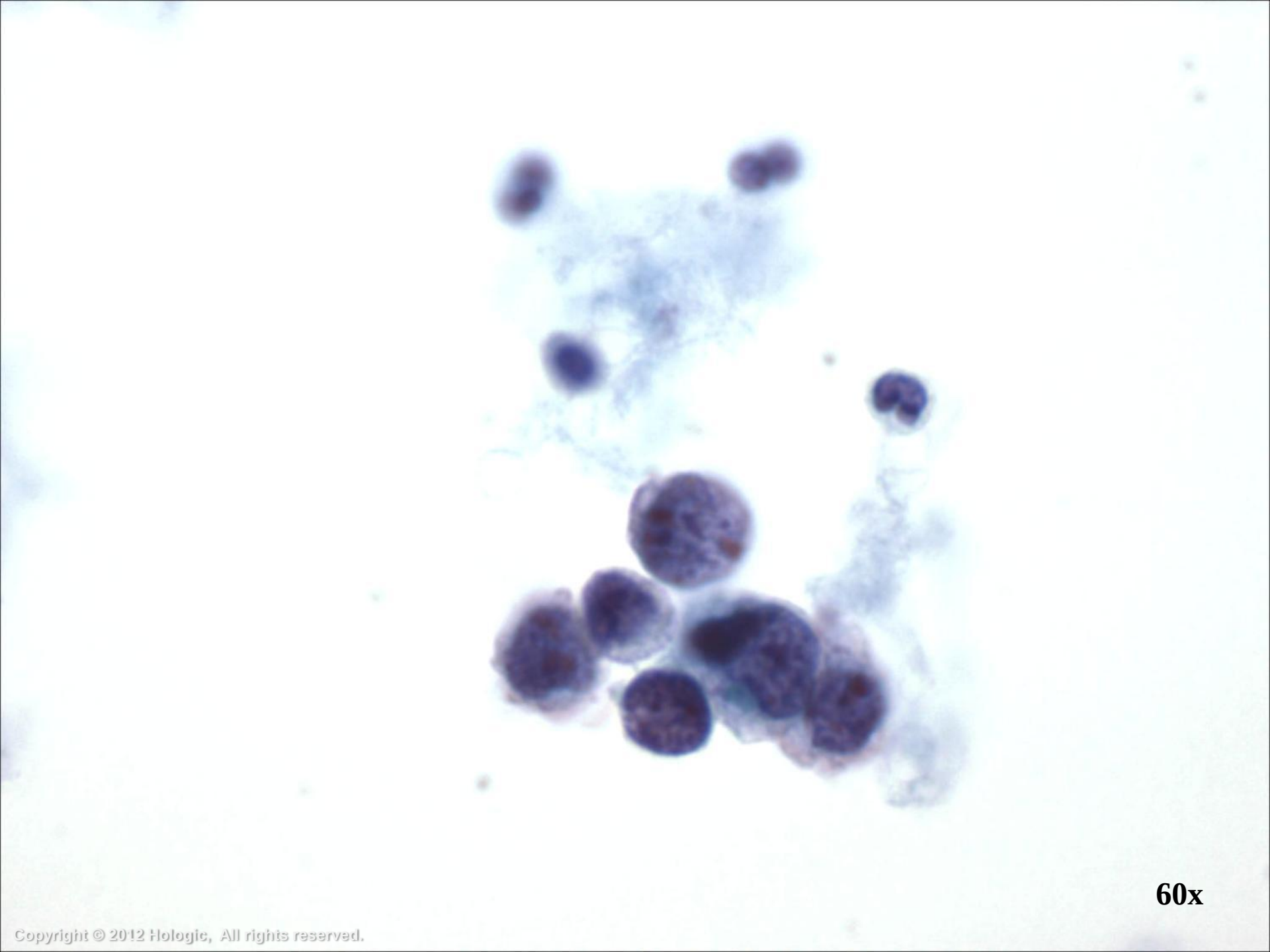

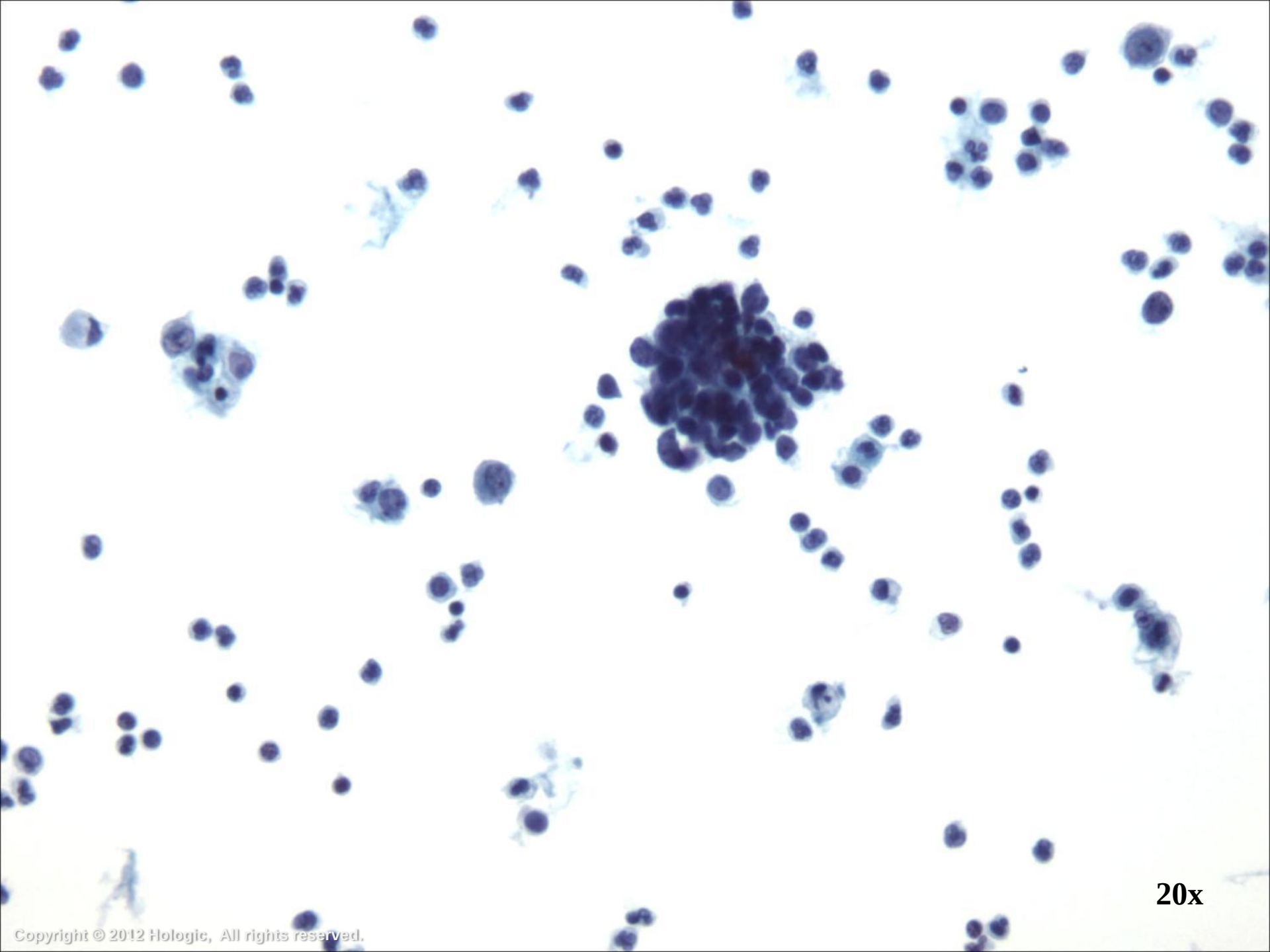

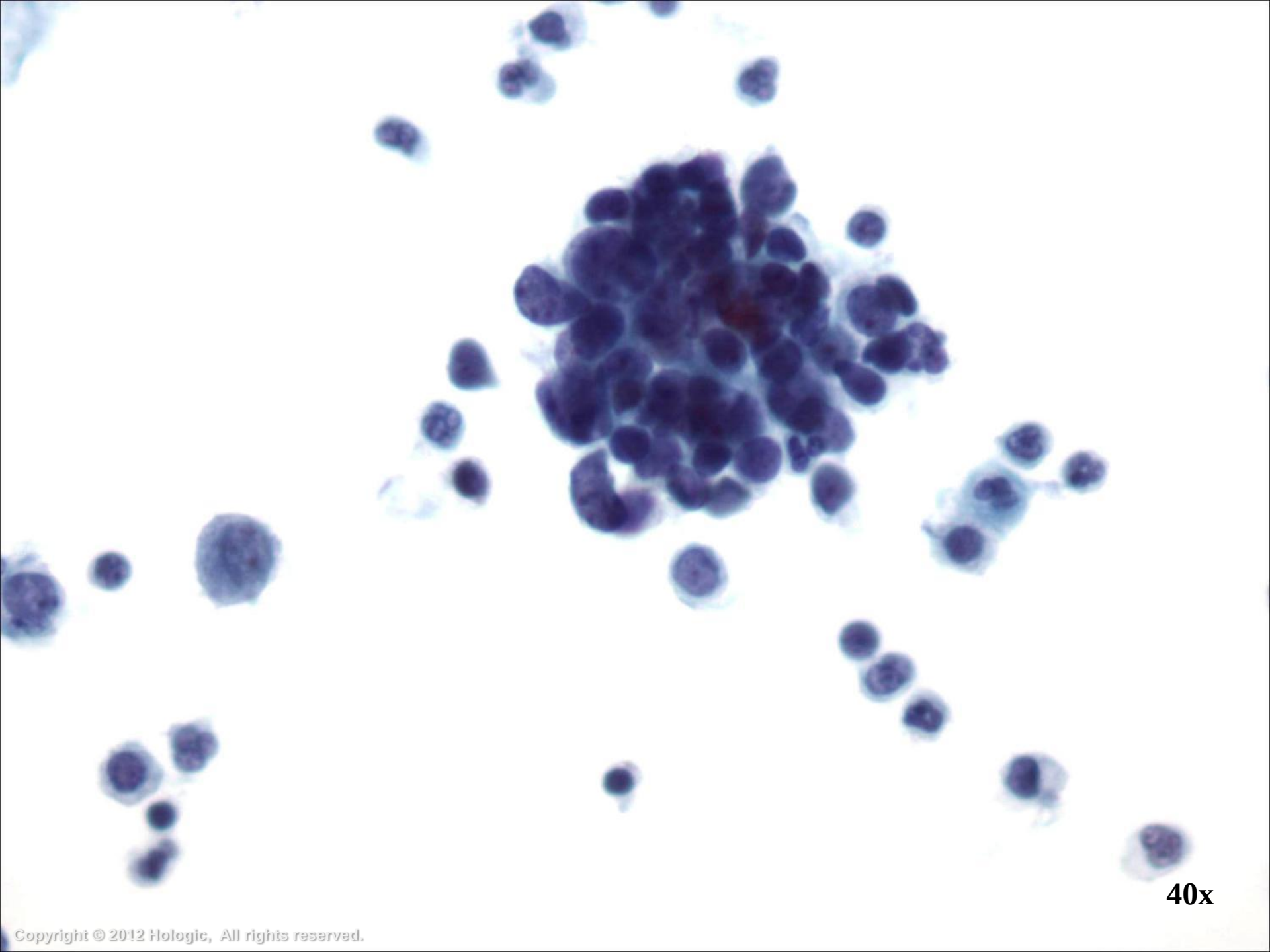

Suggested Immunochemistry Markers – Small Cell Carcinoma

- CK 7 +
- CK 20 -
- TTF-1 +
- Chromogranin +
- Synaptophysin +
- CD56 +

ThinPrep Non-Gyn

Note – Expected staining results; observed in most but not all cases.

Lymphoma/Leukemia

- Non-Hodgkin Lymphoma is most common and accounts for 10-15% of malignant effusions
- In children, hematopoietic neoplasms are the primary common cause of a malignant effusion
- Majority of patients will have biopsy proven disease before effusion develops
- Reed-Sternberg cells in Hodgkin disease are rarely found in effusions

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Lymphoma/Leukemia

- Classification of the cells is possible based on:
  - Size of cells
  - Degree of nuclear abnormalities (cleaved, non-cleaved)
  - Resemblance of cells to lymphoblasts or Burkitt cells
- Cytologic features:
  - Single cell, monomorphic cell pattern
  - Small cells with scant, delicate cytoplasm
  - High N/C ratios
  - Irregular nuclear membranes (cleaves, nuclear knobs)
  - Prominent nucleoli

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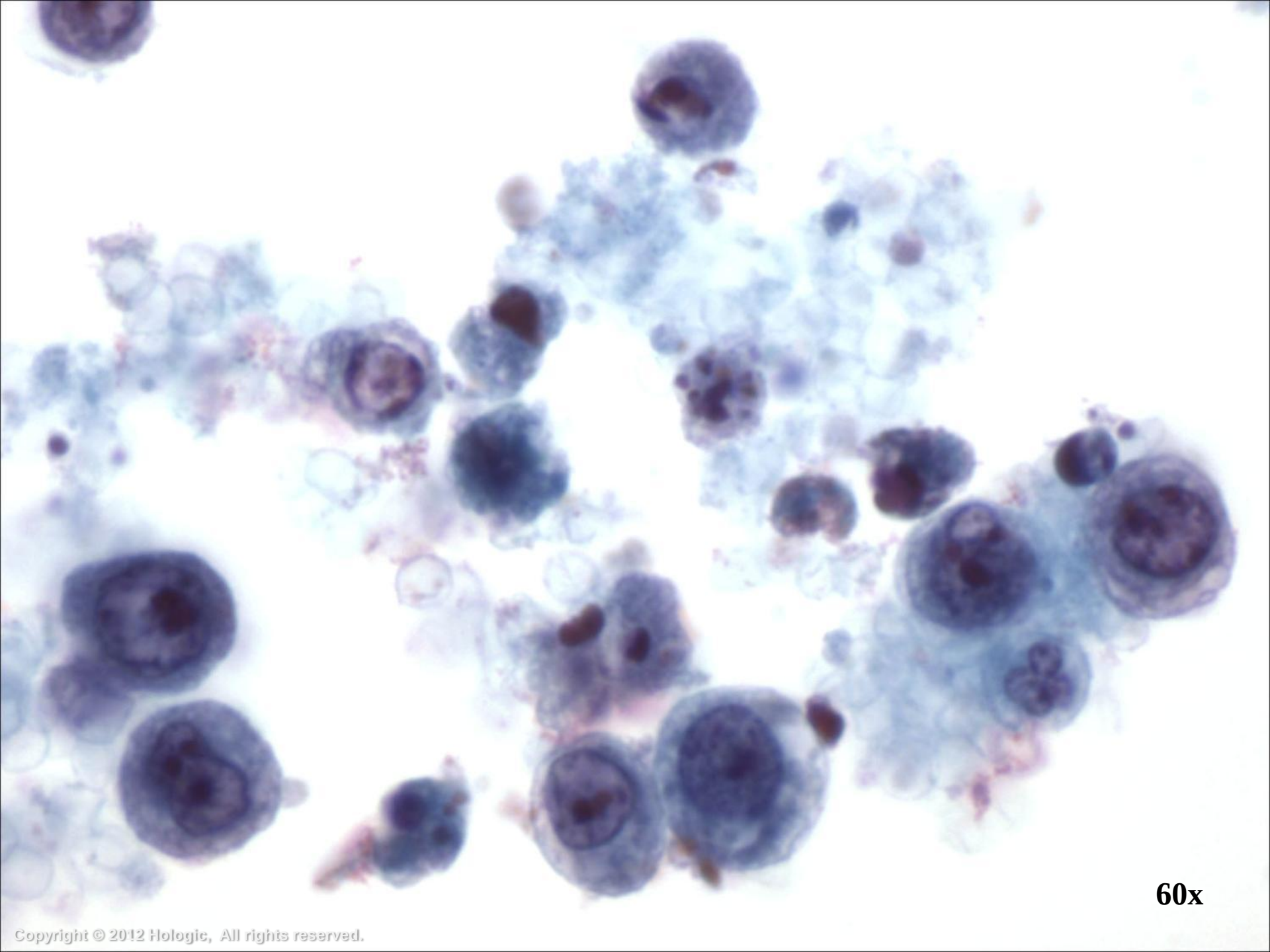

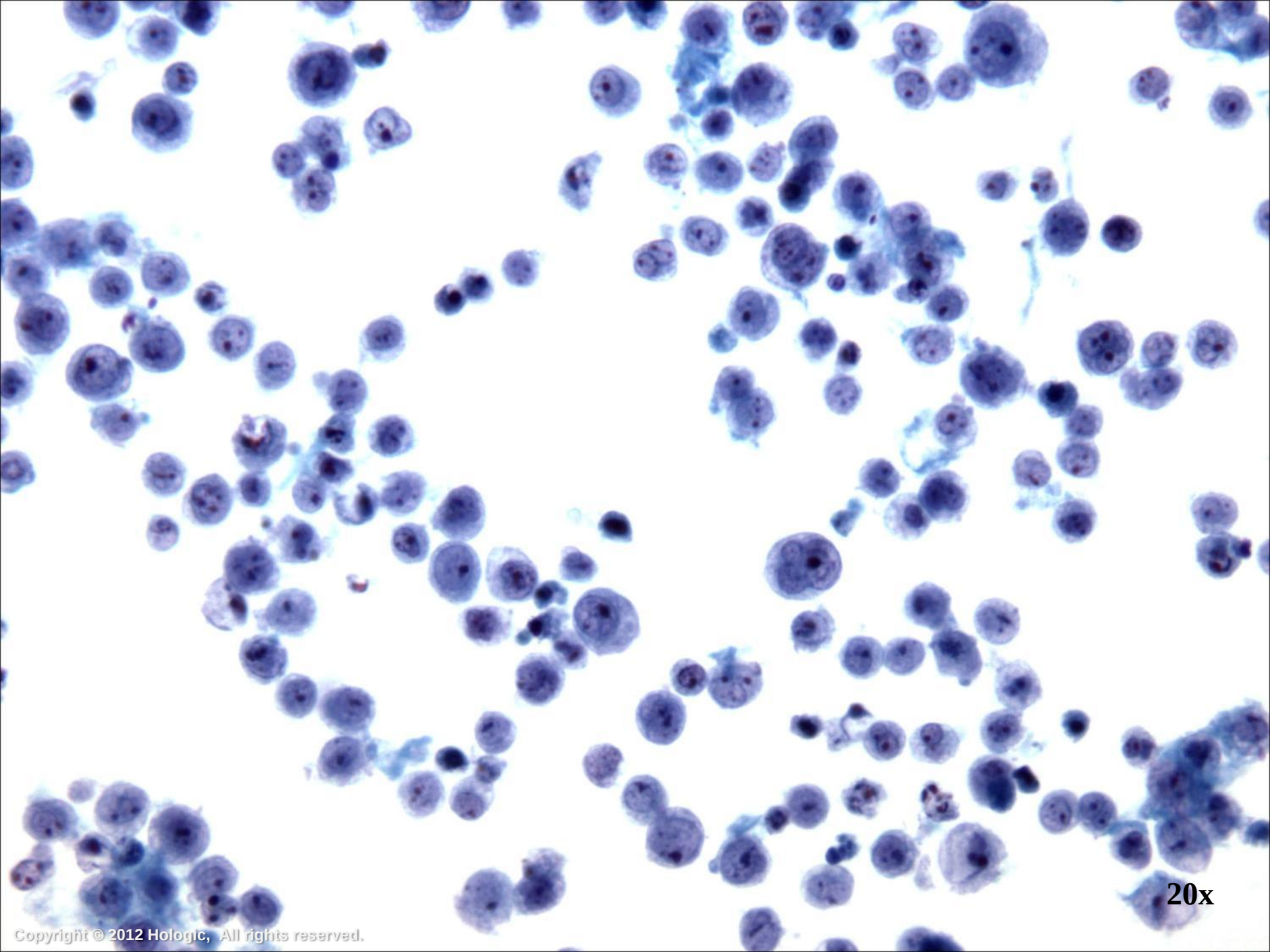

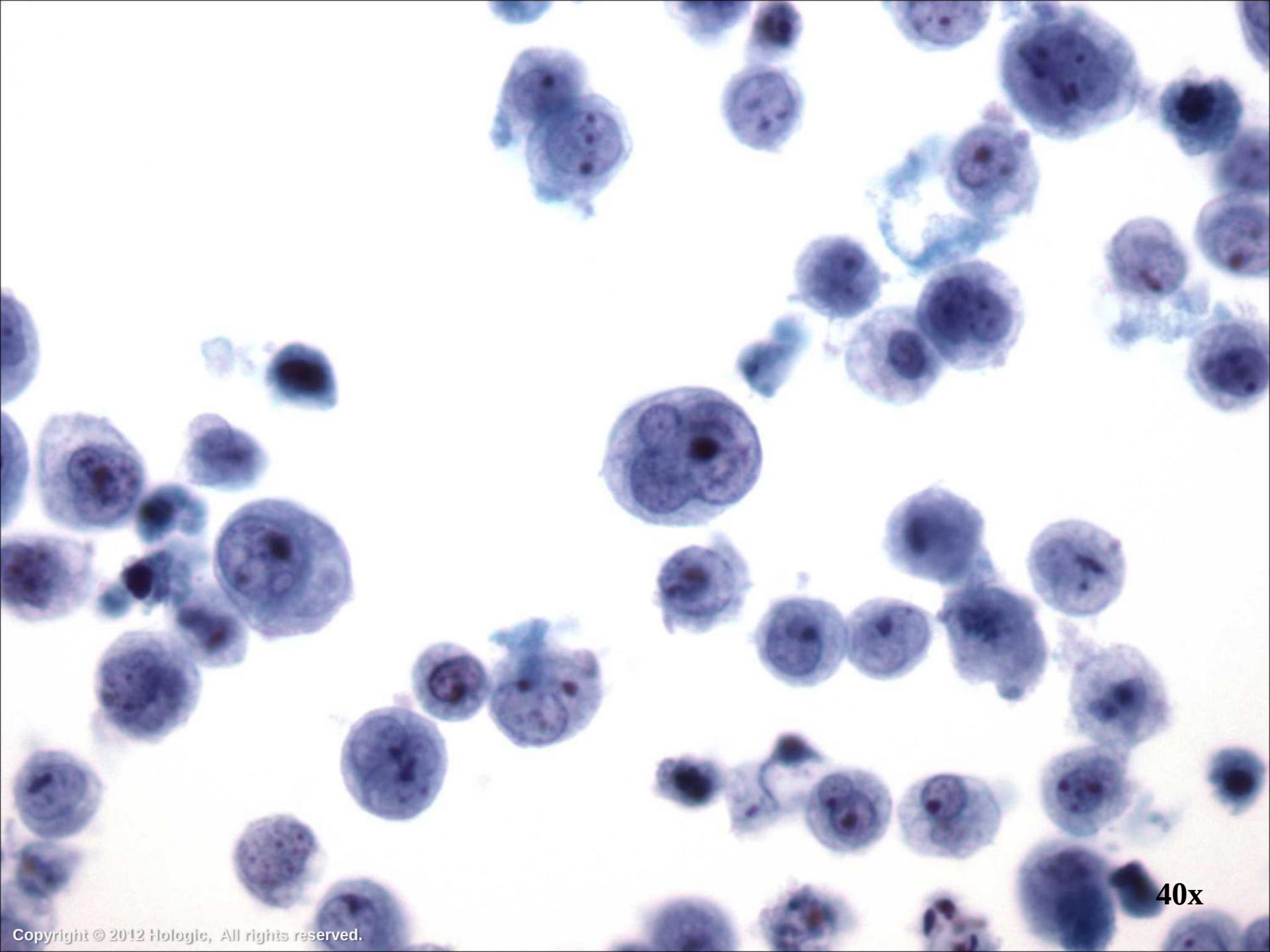

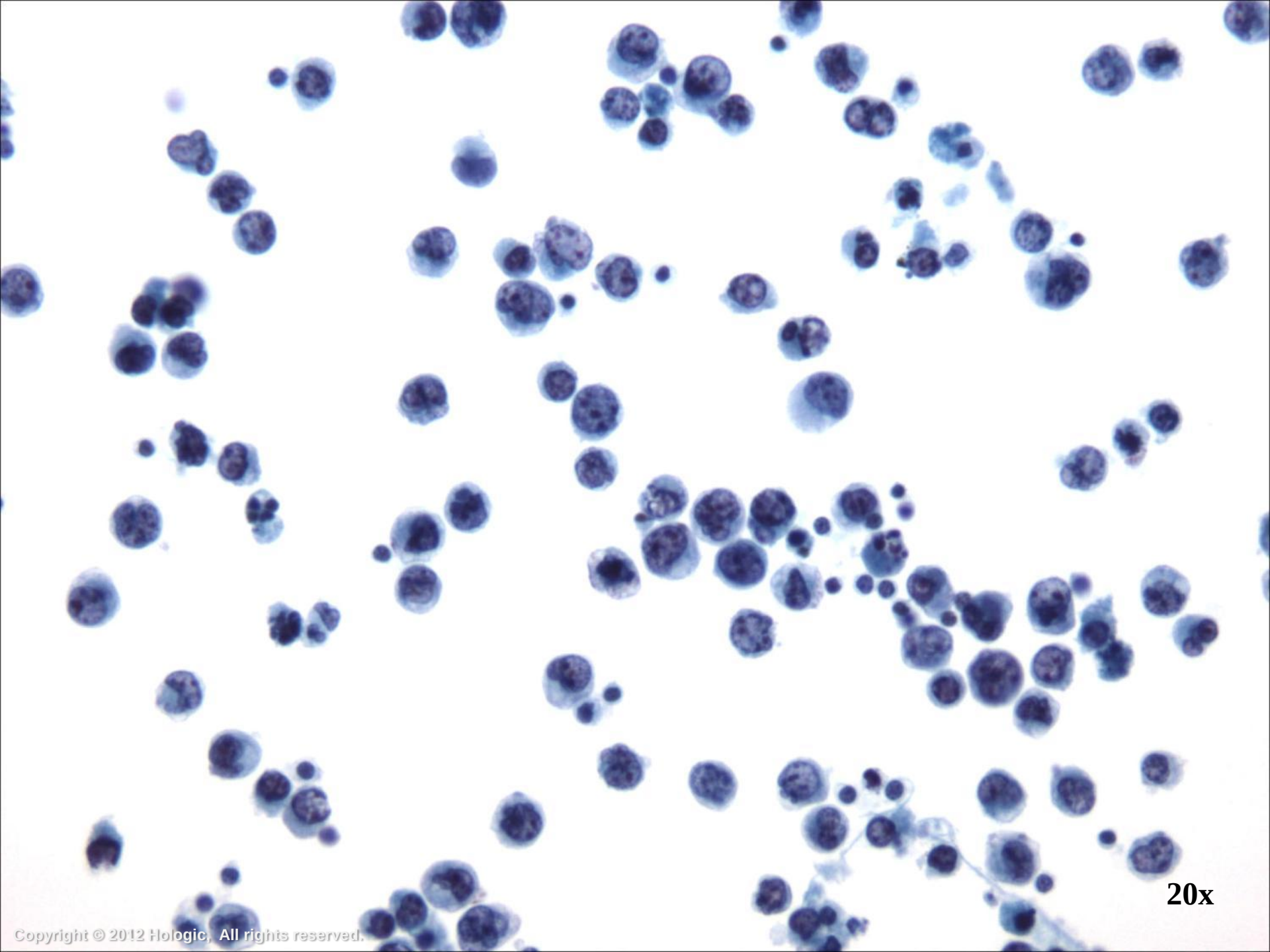

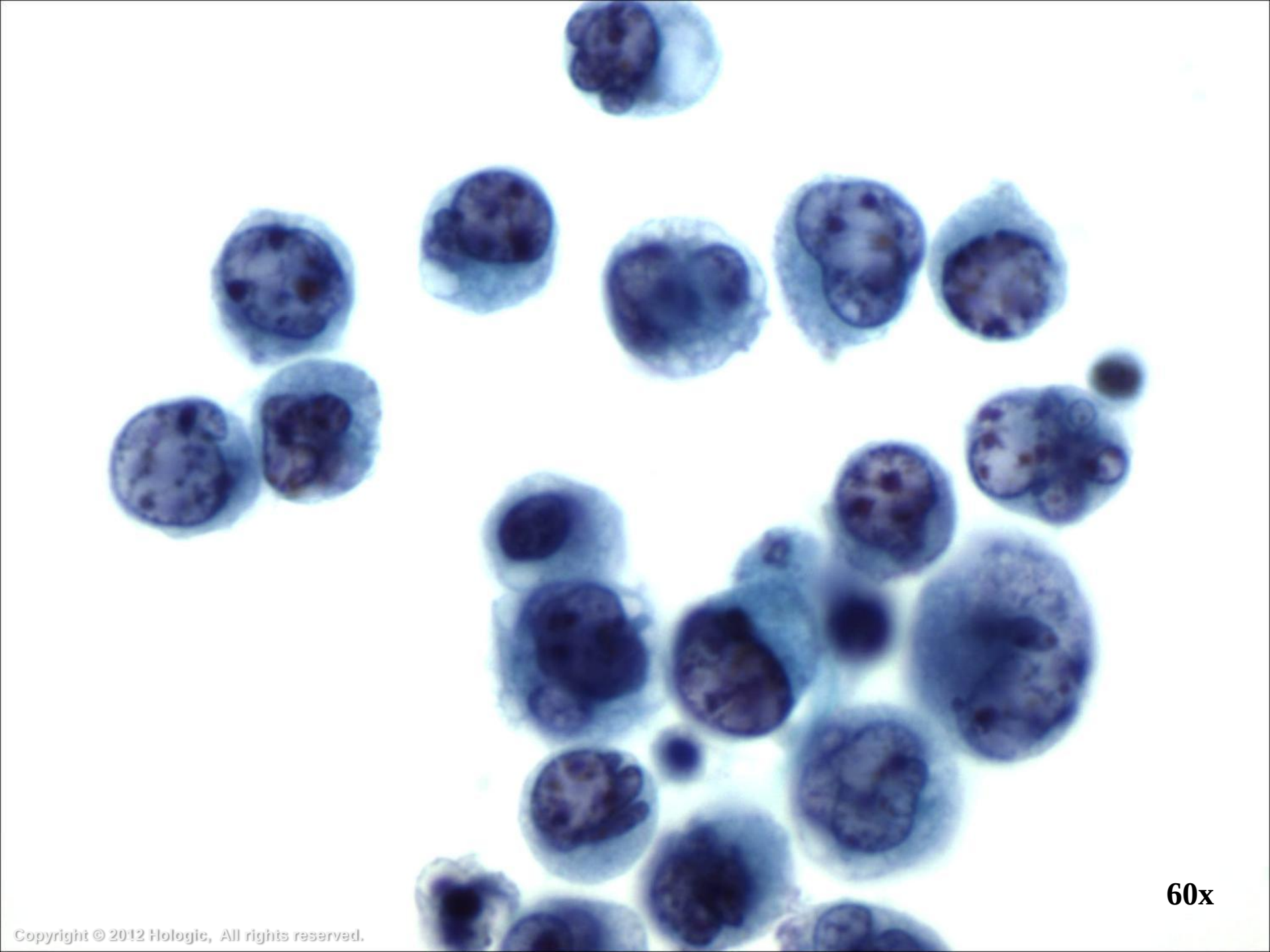

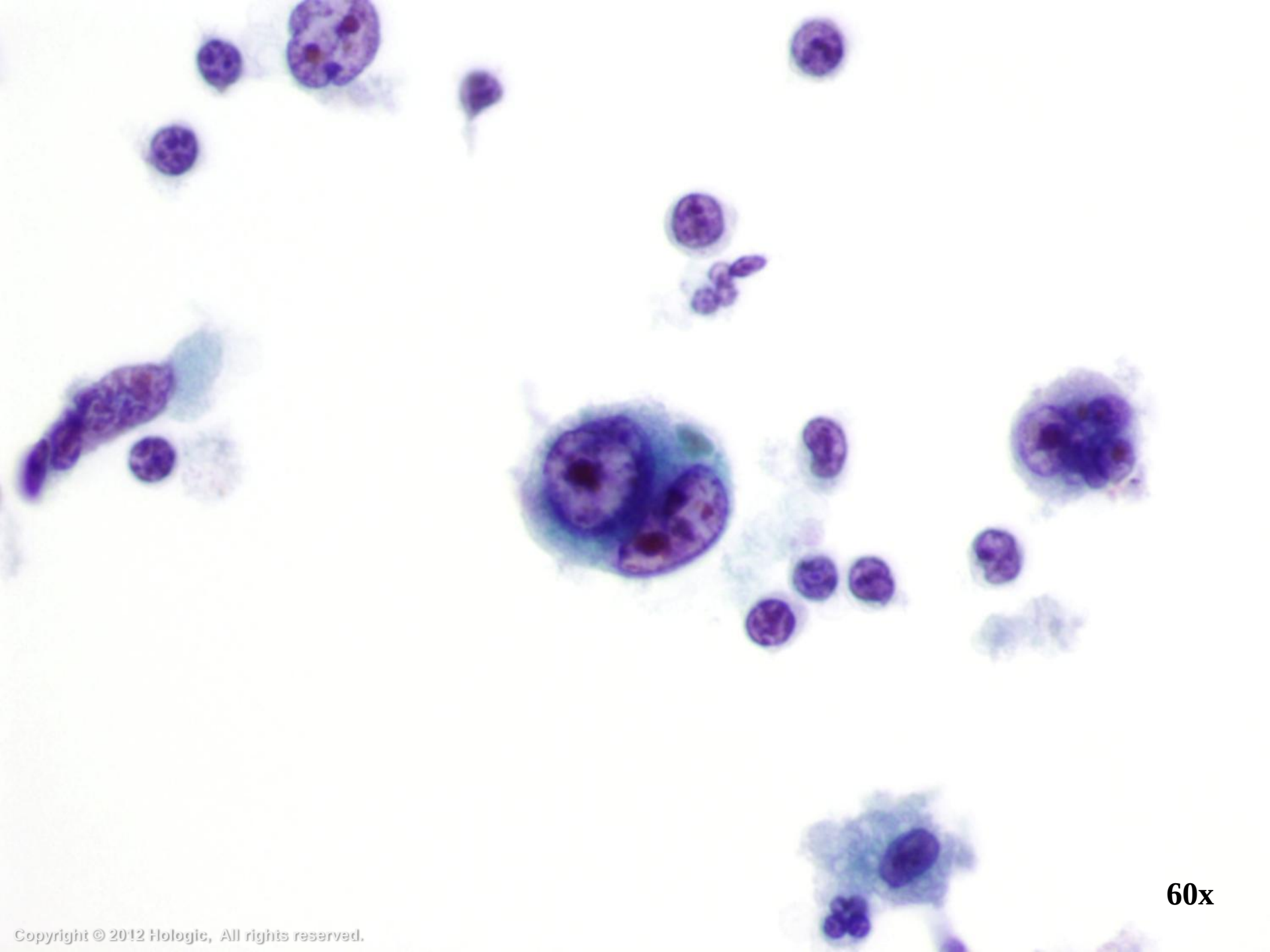

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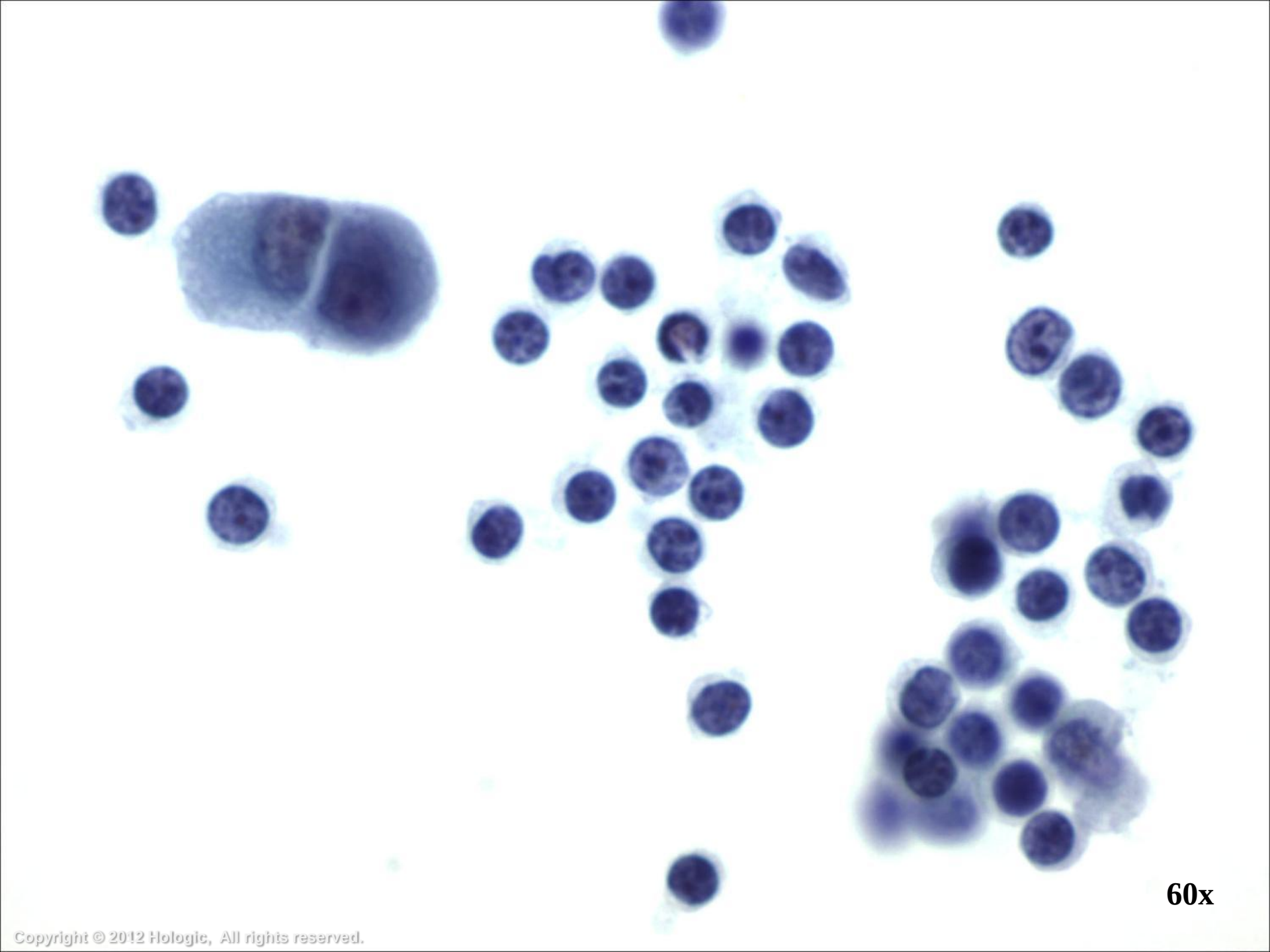

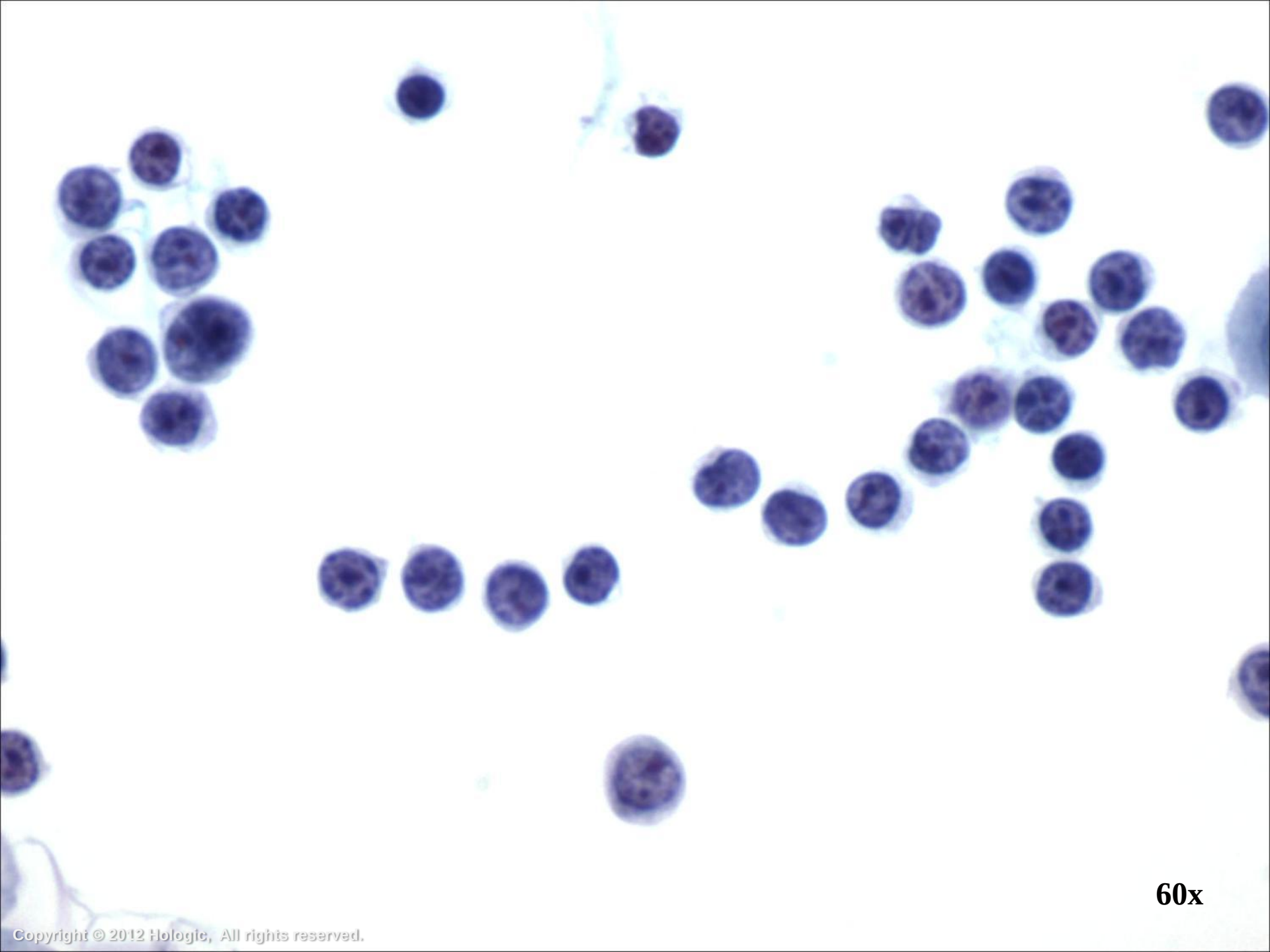

Suggested Immunochemistry Markers – Basic Lymphoid Markers

B Cell Markers

- CD5 +/-
- CD20 +
- LCA (CD45) +
- BCL-2 +

T Cell Markers

- CD3 +
- CD5 +
- LCA (CD45) +

Hodgkin Lymphoma Markers

- CD15 +
- CD30 +
- LCA (CD45) -

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Note – Expected staining results; observed in most but not all cases.

Malignant Melanoma

- Cytologic features:
  - Usually single, large cells
  - Large, eccentric nuclei
  - Frequent intranuclear cytoplasmic invaginations
  - Binucleation is common
  - Very prominent nucleoli
  - Melanin pigment presence is diagnostic

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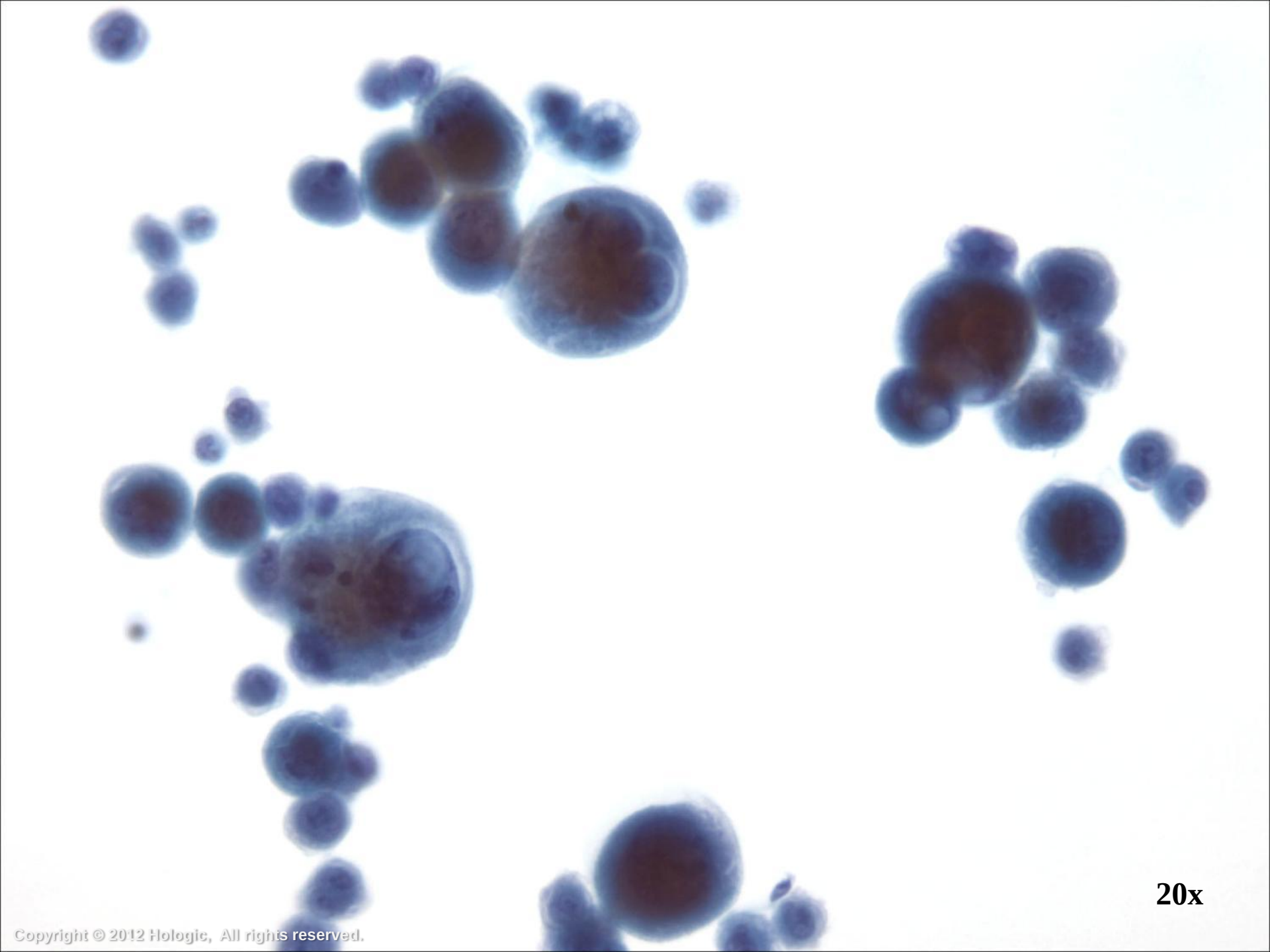

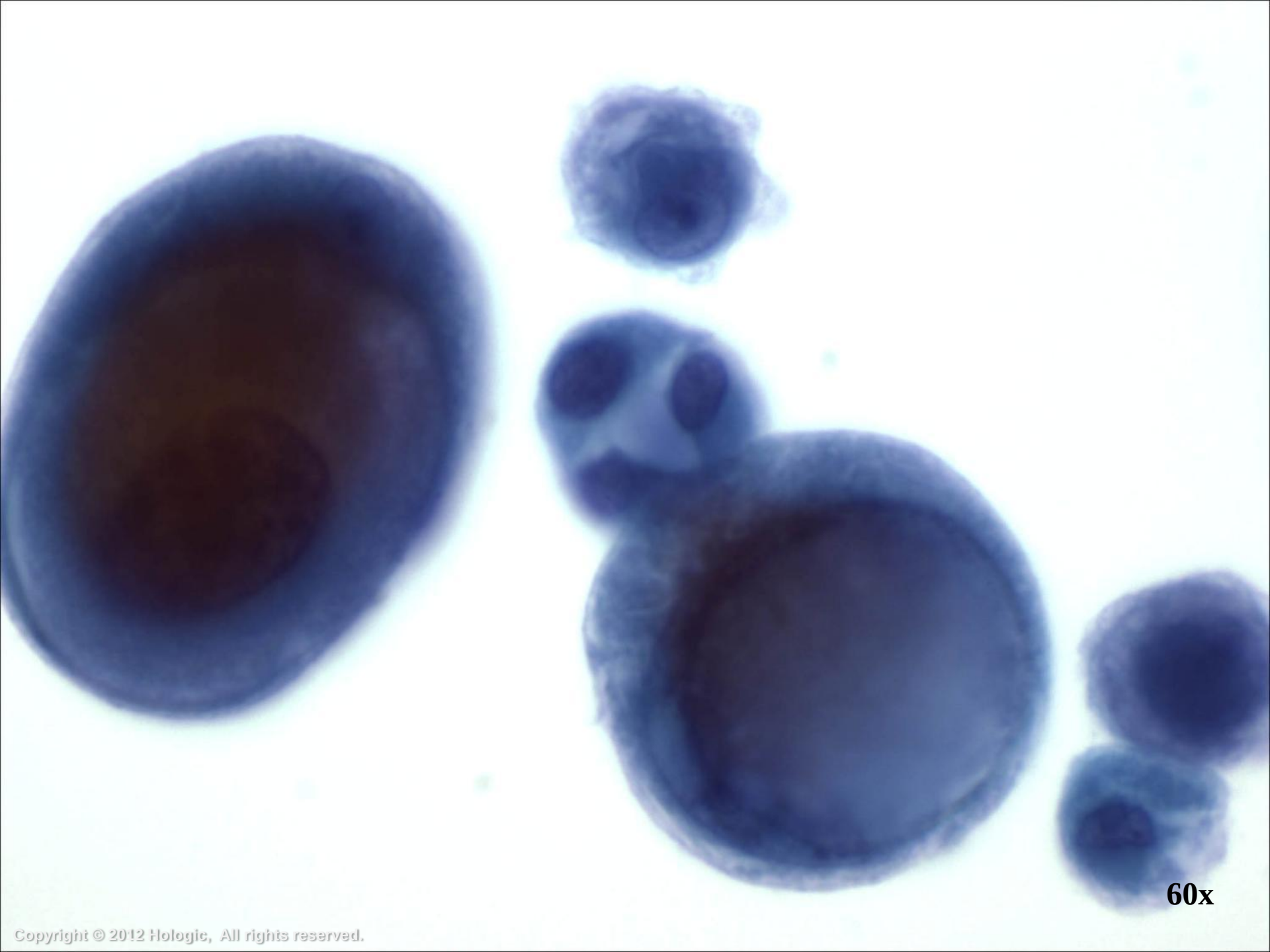

Suggested Immunochemistry Markers – Melanoma

- S100 +
- HMB45 +
- Melan-A +
- MART-1 +

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Note – Expected staining results; observed in most but not all cases.

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