

SPECIMEN SUBMISSION GUIDANCE FOR PATHOLOGIC EVALUATION OF TISSUE SPECIMENS FROM CASES OF LUNG INJURY ASSOCIATED WITH E-CIGARETTE, OR VAPING PRODUCT USE



Characterizing tissue pathology seen in cases of e-cigarette, or vaping, product use (including the device, liquid refill pads and cartridges) associated lung injury is important to further understand the spectrum of pathology and the pathogenesis of lung injury. Evaluation of both fresh and fixed tissues can help improve our understanding during this outbreak. CDC's Infectious Diseases Pathology Branch can provide assistance in the evaluation of fixed tissue specimens.

LUNG BIOPSY TISSUE SPECIMENS

The decision to perform a lung biopsy is at the discretion of the clinical treatment team. This will include consultation with pulmonary, critical care, or other specialties. Initial evaluation of biopsy tissues at the clinical institution should be guided by consultation with these specialties and pathology specialists. Evaluation can include lipid staining on

fresh lung tissues, histopathologic evaluation of formalin-fixed, paraffin-embedded (FFPE) tissues, and testing for possible infectious etiologies.

CDC can receive formalin-fixed (wet) tissues or FFPE lung tissue blocks for evaluation, if available from a biopsy procedure.

AUTOPSY TISSUE SPECIMENS

Autopsies should be considered for cases of lung injury associated with e-cigarette or vaping product use with a fatal outcome. At autopsy, medical examiners and coroners might also identify deaths among persons with a history of e-cigarette product use who had antecedent respiratory or gastrointestinal symptoms, acute respiratory failure, adult respiratory distress syndrome, or gross or microscopic pulmonary pathology. Examples of possible pathologic findings include but are not limited to organizing pneumonia, acute lung injury, diffuse alveolar damage, collections of foamy (vacuolated) histiocytes, unexplained pneumonitis or pneumonia, and alveolar hemorrhage.

Collection of fresh lung tissue for staining of lipids, formalin-fixed (wet) lung tissue, and submission of lung and other tissues for routine tissue processing, paraffin-embedding, and evaluation of histopathology should be considered. Infectious disease testing, including postmortem microbiology and molecular testing, should also be considered.

CDC can receive formalin-fixed (wet) tissues and FFPE tissue blocks from autopsy for evaluation. Because the pathology of acute lung injury and related death is often systemic, preferred specimens for submission to CDC also include tissues from other major organs:

1. Formalin-fixed (wet) tissues and FFPE tissue blocks from lung parenchyma, trachea, bronchi; respiratory tissues should be thoroughly sampled;
2. Liver;
3. Kidney;
4. Heart; and
5. Representative sampling of tissues from all other major organs including brain is recommended, especially any organs with gross or microscopic pathology.



EVALUATION AT CDC

If available, submission of formalin-fixed (wet) lung biopsy or autopsy tissues is encouraged. CDC's Infectious Diseases Pathology Branch can perform lipid staining on formalin-fixed (wet) lung tissues using osmium tetroxide prior to routine tissue processing and paraffin embedding. Routine tissue processing involves the application of alcohols, which remove lipids, to formalin-fixed (wet) tissues. As a result, lipid staining cannot be performed on FFPE lung tissue blocks because they have undergone this routine processing step. CDC's Infectious Diseases Pathology Branch will also review tissue histopathology and perform additional testing, including testing for possible infectious etiologies, as indicated.

SUBMISSION OF SPECIMENS

Pre-approval is required prior to submission of any tissue specimens. For pre-approval please contact Pathology@cdc.gov and LungDiseaseOutbreak@cdc.gov (See page 2–3 for additional guidance.)

FORMALIN-FIXED (WET) TISSUE

Place tissue in 10% buffered formalin for three days (72 hours) for biopsies, and a week for thinly-sliced autopsy tissues. The volume of formalin used to fix tissues should be 10x the volume of tissue.

INFECTIOUS DISEASES PATHOLOGY BRANCH SPECIMEN SUBMISSION REQUIREMENTS

- A. Healthcare providers, pathologists, medical examiners, and coroners—please first report any suspected cases of lung injury associated with e-cigarette product use to your state, territorial, tribal, or local health department**
- B. Health departments—contact Pathology@cdc.gov and LungDiseaseOutbreak@cdc.gov with**
1. A brief clinical history
 2. A listing of available formalin-fixed specimen types including tissue source (e.g., lung biopsy, autopsy tissues) and if formalin-fixed (wet) tissue, FFPE tissue blocks, or slides are available
 3. Description of histopathologic findings in tissues to be submitted including results of any special stains, including lipid stains
 4. CDC Case ID (used by state epidemiologists for submitting case data to CDC) and State Case ID numbers. Email correspondence **should not** include patient identifiers such as name, date of birth, or medical record number. All applicable federal, state, and local regulations must be followed to adhere to patient confidentiality and privacy protections.
- C. After you receive approval by email to submit the case to CDC's Infectious Diseases Pathology Branch (IDPB)**
1. Electronically fill, save, and print both pages of the Specimen Submission Form CDC 50.34.
 - a. The form must be filled electronically to generate three bar-codes required for accessioning
 - b. E-mail addresses of the Original and Intermediate submitters (if any) are mandatory fields
 2. Select Test Order Code CDC-10365 (Pathologic Evaluation of Tissues for Possible Infectious Etiologies)
 3. Enter "E-cigarette, or Vaping, Product Use Associated Lung Injury" and provide CDC and State Case ID numbers in the Comments section on Page 2 of the CDC 50.34 form.
 4. In addition to the CDC 50.34 form, enclose the following in the specimen submission package:
 - a. Surgical pathology, autopsy report (preliminary is acceptable), or both
 - b. Relevant clinical notes, including admission History and Physical (H&P), discharge summary, if applicable

(continued on page 3)

D. Packaging and container guidance

1. If formalin-fixed (wet) tissues are being submitted, there are restrictions on the amount of formalin that can be shipped. Small quantities (inner container less than 30 ml, with a maximum net quantity of 1 L for the entire package) are allowable.
2. For shipping purposes, you may:
 - a. Drain off almost all the formalin, leaving enough to keep the tissue moist
 - b. Place leakproof jar(s) in double Ziploc bags prior to shipping, and add absorbent material to the outer bag (alternatively heat-sealable bags can be used)
 - c. Ship for overnight delivery if possible

E. Guidance for specimen labeling

1. Please ensure that the formalin-fixed (wet) tissue specimen containers/bags and formalin-fixed paraffin-embedded tissue blocks are labeled with two primary patient identifiers (i.e., first and last name, date of birth, unique patient identifier from time of collection, such as surgical pathology number, autopsy number, or medical record number)
2. The patient identifiers on the specimen containers/bags or specimens should match the patient identifiers listed on the CDC 50.34 Form, Surgical Pathology Report, Autopsy Report, and/or other medical records submitted with the specimen(s).
3. Specimens without adequate labeling are subject to delay in accessioning and testing.

F. Mailing contact information

1. Ship to Dr. Sherif Zaki, CDC, IDPB, 1600 Clifton Rd NE, MS: H18-SB, Atlanta, GA 30329-4027
2. Mail in suitable packaging for delivery Monday to Friday, excluding Federal holidays
3. Send tracking number to Pathology@cdc.gov
4. Tel: 404-639-3132, Fax: 404-639-3043, Email: Pathology@cdc.gov

For more information on shipping instructions and specimen handling, see:

<https://www.cdc.gov/laboratory/specimen-submission/detail.html?CDCTestCode=CDC-10365>

Digital slides or microscopic images can also be submitted for an informal, image-only consultation (i.e., no patient report will be issued). Contact Pathology@cdc.gov and see <https://www.cdc.gov/ncezid/dhcpp/idpb/epathology/index.html> for more information.

For more information on CDC's Infectious Diseases Pathology Branch, see: www.cdc.gov/pathology

