CDC encourages clinicians to report possible cases of lung injury associated with e-cigarette product use, or vaping, to their local or state health department for further investigation.

If e-cigarette product use, or vaping, is suspected as a possible cause for a patient's symptoms, a detailed history of the substances used, the devices used, and the sources of the devices and substances, should be obtained, as outlined in the Health Alert Network (HAN). Additionally, efforts should be made to determine if any remaining product and liquids used in the devices are available for testing.

While testing of products and liquids is ongoing, CDC has been asked to provide guidance on collection and storage of clinical samples. This may include retention of residual clinical samples collected for patient care or samples collected and stored from early in a patient's presentation to look for markers of exposure.

Healthcare providers and state public health authorities should work together to make decisions about the collection and storage of clinical samples. Healthcare providers should initiate communication promptly with their local or state public health laboratory to retain samples if further analysis is anticipated.

CDC will update this guidance based on the ongoing investigation.

SCOPE

The purpose of this document is to provide general sample collection and storage guidance for healthcare providers and public health laboratory personnel involved in the care of patients who meet the probable or confirmed case definitions for lung injury associated with e-cigarette product use, or vaping. Healthcare providers and public health laboratory personnel might also consider collecting and storing samples from patients suspected of having lung injury associated with e-cigarette product use, or vaping. These recommendations are not intended to direct laboratory guidance for patient care because these decisions are best made by the clinical treatment team.

Empiric collection instructions are limited to collection and storage of blood and urine. For clinicians treating patients who have undergone bronchoalveolar lavage (BAL), instructions are provided for preservation of BAL fluid. This guidance is provided in the event that specific chemicals, substances, or biomarkers linked to respiratory disease caused by the use of e-cigarette products, or vaping, are identified in the future.

Retention and storage of residual samples that were collected for other types of diagnostic screening and testing can also be considered. Healthcare providers should contact their hospital laboratories to identify and retain such samples before disposal. Healthcare providers should also coordinate long-term storage of samples with state public health authorities and state public health laboratories.

NOTE: CDC recommends that healthcare providers consult their local or state health department for the department's recommendations on collection and storage of clinical samples. The decision to collect clinical samples is at the discretion of healthcare providers and the treatment team. The decision to store samples is at the discretion of healthcare providers and local or state public health authorities.

For more information visit https://www.cdc.gov/lunginjury
A. Bronchoalveolar lavage fluid (BAL fluid) samples

1. Due to the invasive nature of BAL sampling, the decision about whether to perform BAL should be based on the clinical judgement of the treating clinicians.

2. **Timing.** BAL samples may be obtained at any time during the clinical course, but may be most informative if obtained prior to initiation of antimicrobial or steroid therapy. If antibiotics or steroids have been initiated, course, duration and timing relative to BAL should be noted.

3. **Sample collection.** Collect samples in sterile containers. BAL fluid should undergo culture and routine centrifugation followed by cellular analyses and cytopathology, including lipid and other staining, as clinically indicated at the local institution.

Lipoid pneumonia associated with inhalation of lipids in aerosols generated by e-cigarette products, or vaping, has been reported based on the detection of lipid-laden alveolar macrophages obtained by bronchoalveolar lavage (BAL) and lipid staining (e.g., Oil Red O).

Guidance for retaining residual BAL fluid samples after routine clinical evaluation:

1. Remaining uncentrifuged BAL fluid and supernatant from centrifuged BAL fluid should be labeled as such and be retained.

2. Up to 10 unstained cytology slides should be briefly fixed in formalin and retained for future evaluation.

3. Excess cell pellet after cytopathologic evaluation can be divided in half, with half being fixed in formalin and stored at room temperature for further cytopathologic evaluation and half frozen at -20 °C or lower for future chemical or lipid analysis.

4. Place remaining uncentrifuged fluid and centrifuged supernatant from centrifuged fluid into sterile vials with external caps and internal O-ring seals. If there is no internal O-ring seal, then seal tightly with the available cap and secure with Parafilm®. Label each sample container with the patient's name, ID number, the sample type, subsection of lung lavaged, and the date the sample was collected. **FREEZE** these samples at freezer temperatures of -20 °C or lower.

B. Blood samples

1) For each patient, collect up to 8 mL of blood in two (2) 4-mL PURPLE/LAVENDER-top (K2 -EDTA) glass or plastic tubes. If only 3-mL tubes are available, three (3) 3-mL tubes may be collected. (Note: **DO NOT** use gel separators.)

2) Mix contents of tubes by inverting them 8 - 10 times.

3) Label tubes in order of collection. Example: No. 1, No. 2, No. 3.

4) Place a barcode label on each tube so that the barcode looks like a ladder when the tube is upright.

5) Store blood samples at 1 °C to 10 °C. **DO NOT FREEZE.**

C. Urine samples

1) For each patient, store 40 to 60 mL of urine in a screw-cap urine cup.

2) Place barcoded label on the cup when the cup is upright; the barcode will look like a ladder.

3) Indicate on the cup how the sample was collected if the method was other than “clean catch” (example: catheterization).

4) Store urine samples in the freezer. Freezer temperatures of -20 °C or lower are recommended.
1) Establish communication protocols with local hospitals and other healthcare providers regarding appropriate sample collection guidance (see sample collection above).

2) Plan to transport collected samples on at least a twice-weekly basis (or more frequently if needed).

3) Follow your local in-house chain of custody protocols (i.e., shipping manifests).

4) Collect samples from local healthcare providers.

5) Separate plasma from whole blood cells. Centrifuge the purple/lavender top tubes for 15 minutes at 1000 to 1300 g-force to separate the plasma from whole blood cells. Check with the centrifuge rotor manual (or RCF to RPM table) for the proper RPM (e.g. 2400 RPM) to use with your specific rotor.

6) Aliquot plasma into cryotubes and apply labels.

7) **FREEZE** BAL fluid, urine, and plasma samples at freezer temperatures of -20 °C or lower.

8) Any formalin-fixed samples (BAL pellet, cytology slides) should be stored at **room temperature NOT FROZEN**.

9) Contact your state epidemiologist or outbreak principal investigator regarding next steps.

**Note:** Consult your local or state health department about the outbreak of lung disease associated with e-cigarette use, or vaping, as soon as possible. State health department officials seeking technical assistance with case reporting or epidemiologic issues can contact CDC at LungDiseaseOutbreak@cdc.gov. State health department officials seeking technical assistance with laboratory testing can discuss with their state health department laboratories or contact CDC at LungDiseaseOutbreak@cdc.gov.

**SHIPPING INSTRUCTIONS FOR CHEMICAL ANALYSES PERFORMED BY CDC’S DIVISION OF LABORATORY SCIENCES**

Shipments are accepted only from state public health laboratories or local and state health departments. Healthcare providers who would like to submit samples should coordinate with local or state health officials.

Health officials who would like to submit samples to CDC should email NCEHSampleLogistics@cdc.gov. Please list **“CDC Lung Injury Response”** in the title of all email correspondence.