Guidance for Adenovirus Testing and Typing of Patients Under Investigation

CDC Health Alert Network: Children with Acute Hepatitis of Unknown Etiology

HAN Archive - 00462 | Health Alert Network (HAN) (cdc.gov)

On Thursday, April 21, 2022, CDC issued the Health Alert Network (HAN) Health Advisory to notify clinicians and public health authorities of children identified with hepatitis (CDC Hepatitis) and adenovirus infection (CDC Adenovirus). A cluster of pediatric cases of significant liver injury with positive adenovirus infection have been identified and reported to CDC since November 2021. A possible association between pediatric hepatitis and adenovirus infection is currently under investigation after subsequent laboratory testing identified adenovirus type 41 infection in several cases. Clinicians are recommended to consider adenovirus testing and to report such cases to their state or jurisdictional public health authorities and to CDC.

A standard diagnostic workup for children with acute hepatitis should be done locally per treating clinicians.

CDC recommends including adenovirus testing in children with acute hepatitis

Because the potential relationship between adenovirus and acute hepatitis is still under a national epidemiologic investigation, and not limited to individual patient care, please consider collection and submission of the following specimen types (if available) for adenovirus detection.

- Blood specimen collected in purple top EDTA tube (whole blood, plasma) or serum; whole blood is preferred to plasma
- Respiratory specimen (nasopharyngeal swab in VTM/UTM, sputum, or bronchioalveolar lavage [BAL])
- Stool specimen (or rectal swab in VTM/UTM); whenever possible, a stool specimen is preferred to a rectal swab
- If a liver biopsy has already been performed as clinically indicated, or from native liver explant or autopsy:
    - Formalin-fixed, paraffin embedded (FFPE) liver tissue
    - Fresh liver tissue, frozen on dry ice or liquid nitrogen immediately or as soon as possible, and stored at ≤ -70°C

Nucleic acid amplification testing (NAAT, e.g. PCR) is preferred for adenovirus detection (currently not available for FFPE liver biopsy or native liver explant). Testing whole blood by PCR may be more sensitive than testing plasma by PCR and is preferred.

Instructions for diagnostic adenovirus testing of clinical specimens

Local/Clinical Laboratories

- Any clinical specimens that can be tested locally, should be, to ensure the most timely results for patient care.
    - Volume permitting, prepare one aliquot for diagnostic testing and one aliquot for adenovirus typing.
        - Aliquot for the diagnostic test according to the instructions in the applicable test order
        - Aliquot for adenovirus typing (minimum volume = 0.5 mL) and store frozen (use ≤ -70°C if available).
    - Any residual clinical specimens or aliquots that were **positive for adenovirus** and collected from pediatric cases with acute hepatitis should be kept frozen (use ≤ -70°C if available) until adenovirus typing can be completed.

- **For any diagnostic testing needs beyond the local capacity, we recommend contacting your state public health laboratory (SPHL) to determine if direct submission to reference laboratories or the SPHL is preferred.**

- *Reference laboratory submission*: Adenovirus PCR testing capacity for whole blood is less common than testing of respiratory or stool specimens and may require submission to a reference laboratory.
    - Reference Laboratories confirmed to do whole blood (in EDTA) adenovirus PCR include ARUP Laboratories and Quest Diagnostics.
        - Specimen containers should be sealed with Parafilm® and must be securely packed with absorbent material to prevent breakage and spillage.
        - Transport and ship at 4C° (refrigerated with cold packs).

- **DO NOT freeze the whole blood for diagnostic PCR testing**.

  - Quest Diagnostics: 0.35 mL minimum volume, specimen stability (48 hours room temperature; 7 days refrigerated); Adenovirus DNA, Quantitative Real-Time PCR | Test Detail | Quest Diagnostics

  - ARUP Laboratories: 0.50 mL minimum volume, specimen stability (24 hours room temperature; 5 days refrigerated); (Adenovirus, Quantitative PCR | ARUP Laboratories Test Directory), TAT 1-4 days.

- *SPHL Submission*: Any clinical specimens that need additional diagnostic testing, should be submitted to your **State Public Health Laboratory.** Specimens must also include **two primary patient identifiers** on the specimen container, in compliance with CLIA regulations, in case additional diagnostic tests are needed. Acceptable primary patient identifiers include:
  - Full patient name (First and last name)
  - Date of birth
  - A unique ID from the time of specimen collection (secondary unique identifiers do not qualify, such as a state public health lab ID)

State Public Health Laboratories (SPHLs)

- Most specimens received by the SPHL should have been tested locally; diagnostic testing by the SPHL will only be required for a subset of specimens where local/reference lab testing was not available.
  - Any residual clinical specimens or aliquots that were **positive for adenovirus** and collected from pediatric cases with acute hepatitis **should be stored** frozen (use ≤ -70°C if available) for adenovirus typing.

- For specimens requiring testing at the SPHL, volume permitting, prepare one aliquot for diagnostic testing and one aliquot for adenovirus typing.
  - Aliquot for the diagnostic test according to the instructions in the applicable test order
  - Aliquot for adenovirus typing (minimum volume = 0.5 mL) and store frozen (use ≤ -70°C if available).

- If a whole blood adenovirus PCR test is not available at your laboratory, please submit to a reference laboratory (instructions for ARUP Laboratories and Quest submissions are described above).

- If your laboratory does not have an account set up for ARUP Laboratories or Quest Diagnostics or if you receive a specimen type for diagnostic testing but don't have a test available, please notify ncirddvdgast@cdc.gov, respvirus@cdc.gov, and infectious.diseases@aphl.org to help coordinate the diagnostic testing.

Instructions for adenovirus typing submissions of positive clinical specimens

All adenovirus typing submissions MUST be shipped through a SPHL.

Adenovirus typing is a surveillance-use only test. Results will be reported back to the SPHL only.

Local/Clinical Laboratories

- **DO NOT SUBMIT SPECIMENS DIRECTLY TO WADSWORTH CENTER FOR ADENOVIRUS TYPING. THESE MUST BE SHIPPED THROUGH THE STATE PUBLIC HEALTH LABORATORY**.

- Any residual clinical specimens or aliquots that were **positive for adenovirus** and collected from pediatric cases with acute hepatitis **should be stored frozen** (use ≤ -70° if available) for adenovirus typing.

- Submit specimens on dry ice to your **state public health laboratory**, including any local test results and a notification that these specimens are associated with the **"Adenovirus/Hepatitis Investigation in Children."**

State Public Health Laboratories (SPHL)

- Clinical laboratories are being requested to send all adenovirus-positive specimens to SPHLs for adenovirus typing

- Any specimens known to be **adenovirus-positive** by a PCR test should be **kept frozen (≤ -70°C)** upon receipt.
    - The minimum volume requirement for adenovirus typing is 0.5 mL
    - The majority of adenovirus positive specimens will be identified locally; only a subset of diagnostic testing will be facilitated by the SPHL

- Notify your State Epidemiologist POC and CDC (ncirddvdgast@cdc.gov and respvirus@cdc.gov) when you have specimens available for adenovirus typing; CDC will be coordinating all shipments to Wadsworth Center.

- Ship on dry ice; shipments should **NOT be sent until pre-approval is received from CDC**.

VIROLOGY LABORATORY
ATTENTION: ADENO/HEPATITIS
WADSWORTH CENTER. DAVID AXELROD INSTITUTE
120 New Scotland Ave
Albany, NY, 12208

Instructions for submission of virus isolates

- If any virus isolates are received or obtained through the culturing of clinical specimens related to the pediatric hepatitis cases, please notify CDC (ncirddvdgast@cdc.gov and respvirus@cdc.gov) for submission and additional characterization.

Instructions for submission of fixed tissues for pathology testing

Local/Clinical Laboratories

Fixed liver tissue samples (liver biopsy, explants, or autopsy) will receive routine histopathological examination and may be tested by immunohistochemistry for adenovirus and other pathogens at the clinical institution. When these samples are collected as clinically indicated, FFPE liver tissue can be submitted to CDC for additional pathologic characterization and infectious disease testing.

Notify your state public health department and pathology@cdc.gov regarding potential submissions; all submissions will **require pre-approval**. Do not send specimens to CDC until pre-approval and submission instructions are provided by pathology@cdc.gov

Fixed liver tissue from patients who meet the following criteria can be submitted to CDC:

- Pediatric patients with hepatitis of unknown etiology meeting the current case definition
  AND, for which
- Formalin-fixed, paraffin embedded liver tissue specimens are available that a) demonstrate histopathologic evidence of hepatitis AND b) have been submerged in formalin for ≤ 2 weeks prior to embedding in paraffin.

State Public Health Laboratories (SPHL)

- CDC can receive fixed liver tissues samples for additional pathologic characterization and infectious disease testing.

- Acceptable sample criteria: formalin-fixed, paraffin embedded liver tissue specimen that demonstrate histopathologic evidence of hepatitis and have been submerged in formalin for ≤2 weeks prior to embedding in paraffin AND from Pediatric patients with hepatitis of unknown etiology meeting the current outbreak case definition

- Please contact CDC Pathology (pathology@cdc.gov) upon receiving tissue specimen for pre-approval and specific submission instructions. Do not send specimens until pre-approval is provided by pathology@cdc.gov

- See Pathologic Evaluation of Fixed Tissues for Possible Infectious Etiologies (CDC-10365) for additional information.