# What test is the best for diagnosing infectious mononucleosis?

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## EVIDENCE-BASED ANSWER

Tests for antibodies to Epstein-Barr viral capsid antigen (EBVCA) or Epstein-Barr nuclear antigen (EBNA) are the most sensitive, are highly specific, and are also the most expensive for diagnosing infectious mononucleosis (strength of recommendation [SOR]: **C**, based on validating cohort study). Heterophile antibody tests have similar specificity and are cheaper, but are less sensitive in children or in adults during the early days of the illness (SOR: **C**, based on validating cohort study). The polymerase chain reaction (PCR) assay for Epstein-Barr virus DNA is more sensitive than the heterophile antibody test in children, is highly specific, but is also expensive (SOR: **C**, based on validating cohort study). The percentages of atypical lymphocytes and total lymphocytes on a complete blood count (CBC) provide another specific and moderately sensitive, yet inexpensive, test (SOR: **C** based on validating cohort study).

## CLINICAL COMMENTARY

**Initial testing with a CBC is a reasonable strategy**

Diagnosis of infectious mononucleosis by currently available testing remains somewhat problematic, especially early in the course of the illness. Initial testing with a CBC—looking for atypical lymphocytes (which after several days replace the early granulocytic response) and a heterophile antibody titer—is a reasonable strategy.

Most testing for infectious mononucleosis is antibody rather than antigen-related. Thus, delayed or serial testing is more accurate as it takes days to weeks for full antibody response to develop. If the clinical picture remains consistent with a mononucleosis-like syndrome and serial EBV testing is negative, then other illnesses such as cytomegalovirus and toxoplasmosis should be considered.

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## Evidence summary

**EBV-specific antibody tests.** A validating cohort study assessed the sensitivity and specificity of 6 commercial test kits for detection of Epstein-Barr virus-specific antibodies (EBVCA and EBNA). The study compared antibody levels in 139 serum specimens from patients with recent primary EBV infections (confirmed by both a positive heterophile antibody test and an EBV antibody pattern compatible with recent infection) and in 40 specimens from healthy normal controls. The average sensitivity of the antibody tests was 97%

**TABLE**

**Comparison of various tests for infectious mononucleosis**

| TEST | SN | SP | LR+ | LR– | COST |
|---|---|---|---|---|---|
| Patients with clinically suspected IM and: | | | | | |
| Antibody to VCA or EBNA | 97 (95-99) | 94 (89-99) | 16 | 0.03 | $64–$232 |
| Heterophile antibody—latex agglutination | 87 (79-95) | 91 (82-99) | 9.7 | 0.14 | $36–$64 |
| Heterophile antibody—solid-phase immunoassay | 83 (71-95) | 97 (94-99) | 28 | 0.18 | $36–$64 |
| Atypical lymphocytes (CBC with differential) | | | | | $37–$50 |
| >10% | 75 | 92 | 9.4 | 0.27 | |
| >20% | 56 | 98 | 28 | 0.44 | |
| >40% | 25 | 100 | 50 | 0.75 | |
| Total lymphocytes (peripheral smear) | | | | | $20–$44 |
| >50% lymphocytes | 66 | 84 | 4.1 | 0.40 | |
| >50% lymphocytes with >10% atypical lymphocytes | 61 | 95 | 12 | 0.41 | |
| PCR for EBV DNA | 75 (62–78) | 95.5 (79–99.8) | 16.67 | 0.26 | $64–$232 |

SN, sensitivity; SP, specificity; LR, likelihood ratio; IM, infectious mononucleosis; VCA, viral capsid antigen; EBNA, Epstein-Barr nuclear antigen; CBC, complete blood count; PCR, polymerase chain reaction

**FAST TRACK**

**Most tests for infectious mononucleosis are antibody-related; thus, delayed or serial testing is more accurate**

(95%–99%) and average specificity was 94% (86%–100%).[1]

**Heterophile antibody tests.** Two validating cohort studies assessed the accuracy of several commercially available test kits for the detection of heterophile antibodies (eg, "Mono spot" tests). The first compared 6 kits using either a latex agglutination or a solid-phase assay against a "gold standard" of serologic verification for 53 serum samples from primary EBV infection, 26 samples from EBV immune patients, and 21 samples from healthy donors.[2] Serologic verification used immunoflourescence to determine: the absence of IgG but presence of IgM, the presence of IgG but absence of IgM, or the absence of both antibodies (respectively) against EBVCA. The second study used a similar method to test 6 more heterophile kits using blood samples from 140 patients.[1] The sensitivity of heterophile antibody testing is lower in children under 12 (25%–50%) and early in the illness (25% false-negative rate in first week).[3]

**PCR assay for EBV DNA.** Another validating cohort study evaluated PCR testing for EBV DNA among children (average age 9 years, 4 months), 28 with infectious mononucleosis, 25 who were EBV seronegative, and 26 who were seropositive. Children with acute infectious mononucleosis were diagnosed by symptoms, >10% atypical lymphocytes, and a positive heterophile antibody test. The PCR found a sensitivity and specificity of 75% and 98% at 1 week.[4] Testing earlier, especially in children, is expected to decrease the sensitivity

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due to the lower maturity of the immune system response.

**Lymphocyte and atypical lymphocyte count.** A validating cohort study compared peripheral blood samples in 181 patients aged >16 years with a clinical diagnosis of infectious mononucleosis confirmed by a positive heterophile antibody test with those from 181 similar patients with a negative test. An increased percentage of lymphocytes and atypical lymphocytes were associated with higher sensitivity and specificity for infectious mononucleosis.[5]

## Recommendations from others

In the appropriate clinical situation, the Centers for Disease Control and Prevention recommends verifying the diagnosis of infectious mononucleosis with a CBC and heterophile antibody test. If the heterophile test result is negative, additional testing such as EBV DNA tests may be necessary.[6]

### REFERENCES


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