MAYO CLINIC LABORATORIES

Test Definition: ASPAG

Aspergillus (Galactomannan) Antigen, Serum

Overview

Useful For

Aiding in the diagnosis of invasive aspergillosis

Assessing response to therapy

Method Name

Enzyme Immunoassay (EIA)

NY State Available

Yes

Specimen

Specimen Type

Serum SST

Ordering Guidance

For bronchoalveolar lavage specimens, order ASPBA / *Aspergillus* Antigen, Bronchoalveolar Lavage.

Specimen Required

Container/Tube: Serum gel (red-top tubes are **not acceptable**)

Specimen Volume: 1.5 mL

Collection Instructions:

1. Avoid exposure of specimen to atmosphere to prevent sample contamination from environment.
2. Centrifuge and send specimen in original tube. **Do not aliquot or open tube.**

Forms

If not ordering electronically, complete, print, and send 1 of the following forms with the specimen:

-General Request (T239)

-Infectious Disease Serology Test Request (T916)

Specimen Minimum Volume

1 mL

Reject Due To

Gross hemolysis	Reject
Gross lipemia	Reject

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Serum SST	Refrigerated (preferred)	14 days	SERUM GEL TUBE
	Frozen	14 days	SERUM GEL TUBE

Clinical & Interpretive

Clinical Information

Invasive aspergillosis (IA) is a severe infection that occurs in patients with prolonged neutropenia, following transplantation or in conjunction with aggressive immunosuppressive regimens (eg, prolonged corticosteroid usage, chemotherapy). The incidence of IA is reported to vary from 5% to 20% depending on the patient population. IA has an extremely high mortality rate of 50% to 80% due in part to the rapid progression of the infection (ie, 1-2 weeks from onset to death). Approximately 30% of cases remain undiagnosed and untreated at death.

Definitive diagnosis of IA requires histopathological evidence of deep-tissue invasion or a positive culture. This evidence is often difficult to obtain due to the critically ill nature of the patient and the fact that severe thrombocytopenia often precludes the use of invasive procedures to obtain a quality specimen. The sensitivity of culture in this setting is low, reportedly ranging from 30% to 60% for bronchoalveolar lavage fluid. Accordingly, the diagnosis is often based on nonspecific clinical symptoms (unexplained fever, cough, chest pain, dyspnea) in conjunction with radiologic evidence (computed tomography scan); a definitive diagnosis is often not established before fungal proliferation becomes overwhelming and refractory to therapy.

Recently, a serologic assay was approved by the US Food and Drug Administration for the detection of galactomannan, a molecule found in the cell wall of *Aspergillus* species. Serum galactomannan can often be detected a mean of 7 to 14 days before other diagnostic clues become apparent, and monitoring of galactomannan can potentially allow initiation of preemptive antifungal therapy before life-threatening infection occurs.

Reference Values

<0.5 index
Reference values apply to all ages.

Interpretation

A positive result supports a diagnosis of invasive aspergillosis (IA). Positive results should be considered in conjunction with other diagnostic procedures, such as microbiologic culture, histological examination of biopsy specimens, and radiographic evidence. See Cautions.

A negative result does not rule out the diagnosis of IA. Repeat testing is recommended if the result is negative but IA is clinically suspected. Patients at risk of IA should have a baseline serum tested and should be monitored twice a week for increasing galactomannan antigen levels.

Galactomannan antigen levels may be useful in the assessment of therapeutic response. Antigen levels decline in response to antimicrobial therapy.

Cautions

False-positive results are reported to occur at rates of 8% to 14% with this assay. For all positive patients, it is recommended that a new aliquot of the same specimen be repeated, as well as collection of a new specimen from the patient for follow-up testing. Two or more consecutive positive results should be obtained from separately collected specimens before the patient is considered to have a positive *Aspergillus* antigen test.

Numerous foods (pasta, rice, etc) contain galactomannan. It is thought that damage to the gut wall by cytotoxic therapy, irradiation, or graft-versus-host disease enables translocation of the galactomannan from the gut lumen into the blood and may be partially responsible for the high false-positive rate of this assay.

Other genera of fungi such as *Penicillium* and *Paecilomyces* have shown reactivity with the rat EBA-2 monoclonal antibody used in the assay. These species are rarely implicated in invasive fungal disease. Cross reactivity with *Alternaria* species has also been reported.

The specificity of the assay for *Aspergillus* species cannot exclude the involvement of other fungal pathogens with similar clinical presentations such as *Fusarium*, *Alternaria*, and *Mucorales*.

The performance of the assay has not been evaluated with neonate serum specimens or for use with plasma or other specimen types such as urine or cerebrospinal fluid.

The assay may exhibit reduced detection of galactomannan in patients with chronic granulomatous disease or autosomal dominant hyper-IgE syndrome (formerly known as Job syndrome).

The concomitant use of antifungal therapy in some patients with invasive aspergillosis may result in reduced sensitivity of the assay.

False-positive galactomannan results are possible in patients receiving PLASMA-LYTE for intravenous hydration or if PLASMA-LYTE is used during bronchoscopy for the collection of bronchoalveolar lavage fluid.

Specimens containing *Histoplasma* antigen may cross-react in the *Aspergillus* galactomannan assay.

Supportive Data

In clinical studies submitted for the US Food and Drug Administration-approval process, the sensitivity of the test was reported to be 81% for proven/provable invasive aspergillosis (N=31 patients), and the specificity was 89% (N=148 patients). The positive and negative predictive values were reported as 68% and 96% respectively, based on an average prevalence of 14% in the study population. In a low-prevalence population (5%), the positive predictive value decreases to 31%; the negative predictive value remains at 96%.(Package insert: Platelia *Aspergillus* EIA, Bio-Rad Laboratories; 06/2003)

Clinical Reference

1. Maertens J, Verhaegen J, Lagrou K, Boogaerts M: Screening for circulating galactomannan as a noninvasive diagnostic tool for invasive aspergillosis in prolonged neutropenic patients and stem cell transplantation recipients: a prospective evaluation. Blood. 2001 March 15;97(6):1604-1610

2. Pinel C, Fricker-Hidalgo H, Lebeau B, et al: Detection of circulating *Aspergillus fumigatus* galactomannan: value and limits of the Platelia test for diagnosing invasive aspergillosis. J Clin Microbiol. 2003 May;41(5):2184-2186

3. Swanink CM, Meis JF, Rijs AJ, Donnelly JP, Verweij PE: Specificity of a sandwich enzyme-linked immunosorbent assay for detecting *Aspergillus* galactomannan. J Clin Microbiol. 1997 Jan;35(1):257-260

4. Ansorg R, van den Boom R, Rath PM: Detection of *Aspergillus* galactomannan antigen in foods and antibiotics. Mycoses. 1997 Dec;40(9-10):353-357
5. Connolly P, Durkin M, Wheat LJ, et al: Rapid diagnosis of systemic and invasive mycoses. Clinical Microbiology Newsletter. 2007 Jan;29(1):1-5
6. Thompson GR, Patterson TF: *Aspergillus* species. In: Bennett JE, Dolin R, Blaser MJ, eds. Mandell, Douglas, and Bennett's Principles and Practice of Infectious Diseases. 9th ed. Elsevier; 2020:3103-3116

Performance

Method Description

The Platelia *Aspergillus* enzyme immunoassay (EIA) is a 1-stage immunoenzymatic sandwich microplate assay that detects galactomannan in human serum. The assay uses the rat monoclonal antibody EBA-2, which is directed against *Aspergillus* galactomannan. The monoclonal antibody is used to coat the wells of the microplate and bind the antigen and as the detector antibody in the conjugate reagent (peroxidase-linked monoclonal antibody).

Serum Samples are heat-treated in the presence of EDTA to dissociate immune complexes and to precipitate serum proteins that could possibly interfere with the test. The treated serum samples and conjugate are added to the wells coated with the monoclonal antibody and incubated. A monoclonal antibody-galactomannan-monoclonal antibody/peroxidase complex is formed in the presence of *Aspergillus* antigen.

The strips are washed to remove any unbound material, and the substrate solution is added, which will react with the complex bound to the well to form a blue color reaction. The enzyme reaction is stopped by the addition of acid, which changes the blue color to yellow. The optical absorbance of specimens and controls is determined with a spectrophotometer set at 450 nm and 620/630 nm wavelengths.

Negative, cutoff (low-positive), and high-positive controls are analyzed each time the assay is performed. The presence or absence of *Aspergillus* galactomannan antigen in the test sample is determined by calculation of an index for the specimen. The index is the optical density (OD) value of the specimen divided by the mean OD of wells containing the cutoff control serum (low-positive control).(Package insert: Platelia *Aspergillus* EIA. Bio-Rad Laboratories; 10/2020)

PDF Report

No

Day(s) Performed

Monday through Friday, Sunday

Report Available

1 to 4 days

Specimen Retention Time

2 weeks

Performing Laboratory Location

Rochester

Fees & Codes

Fees

- Authorized users can sign in to Test Prices for detailed fee information.
- Clients without access to Test Prices can contact Customer Service 24 hours a day, seven days a week.
- Prospective clients should contact their account representative. For assistance, contact Customer Service.

Test Classification

This test has been cleared, approved, or is exempt by the US Food and Drug Administration and is used per manufacturer's instructions. Performance characteristics were verified by Mayo Clinic in a manner consistent with CLIA requirements.

CPT Code Information

87305

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
ASPAG	Aspergillus Ag, S	44357-2

Result ID	Test Result Name	Result LOINC® Value
84356	Aspergillus Ag, S	44357-2