Overview

Useful For
Diagnosis of antithrombin deficiency, acquired or congenital

Monitoring treatment of antithrombin deficiency disorders, including infusion of antithrombin therapeutic concentrate

Special Instructions
- Coagulation Guidelines for Specimen Handling and Processing

Method Name
Chromogenic Assay

NY State Available
Yes

Specimen

Specimen Type
Plasma Na Cit

Advisory Information
Coagulation testing is highly complex, often requiring the performance of multiple assays and correlation with clinical information. For that reason, consider ordering AATHR / Thrombophilia Profile, Plasma and Whole Blood.

Specimen Required

Specimen Type: Platelet-poor plasma
Collection Container/Tube: Light-blue top (3.2% sodium citrate)
Submission Container/Tube: Plastic vial
Specimen Volume: 1 mL

Collection Instructions:
1. For complete instructions, see Coagulation Guidelines for Specimen Handling and Processing in Special Instructions.
2. Centrifuge, transfer all plasma into a plastic vial, and centrifuge plasma again.
3. Aliquot plasma into a plastic vial leaving 0.25 mL in the bottom of centrifuged vial.
4. Freeze plasma immediately (no longer than 4 hours after collection) at -20 degrees C or, ideally < or =-40 degrees C.

Additional Information:
1. A double-centrifuged specimen is critical for accurate results as platelet contamination may cause spurious results.
2. Each coagulation assay requested should have its own vial.
3. Heparin treatment may lower plasma antithrombin.

Forms
If not ordering electronically, complete, print, and send a Coagulation Test Request (T753) with the specimen.

Specimen Minimum Volume
0.5 mL
Test Definition: ATTF
Antithrombin Activity, P

Reject Due To

<table>
<thead>
<tr>
<th>Condition</th>
<th>Action</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gross hemolysis</td>
<td>Reject</td>
</tr>
<tr>
<td>Gross lipemia</td>
<td>Reject</td>
</tr>
<tr>
<td>Gross icterus</td>
<td>Reject</td>
</tr>
</tbody>
</table>

Specimen Stability Information

<table>
<thead>
<tr>
<th>Specimen Type</th>
<th>Temperature</th>
<th>Time</th>
<th>Special Container</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plasma Na Cit</td>
<td>Frozen</td>
<td>14 days</td>
<td></td>
</tr>
</tbody>
</table>

Clinical and Interpretive

Clinical Information

Antithrombin is a member of the serine protease inhibitor (serpin) superfamily. It is the principal plasma anticoagulant serpin mediating inactivation of serine protease procoagulant enzymes, chiefly thrombin and coagulation factors Xa and IXa.(1) Heparin and certain other naturally occurring glycosaminoglycans markedly enhance the anticoagulant activity of antithrombins (approximately 1000-fold) by providing a template to catalyze formation of covalently bonded, inactive complexes of serine protease and antithrombin that are subsequently cleared from circulation. Antithrombin is the mediator of anticoagulant activity of heparin.

The antithrombin gene on chromosome 1 encodes a glycoprotein with a molecular weight of approximately 58,000 D, which is synthesized in the liver and is present in a relatively high plasma concentration (approximately 2.3 mcmol/L). The biological half-life of antithrombin is 2 to 3 days.

Hereditary antithrombin deficiency, a relatively rare autosomal dominant disorder, produces a thrombotic diathesis (thrombophilia). Individuals with hereditary antithrombin deficiency are usually heterozygous with plasma antithrombin activity results of approximately 40% to 70%. These patients primarily manifest with venous thromboembolism (deep vein thrombosis: DVT, and pulmonary embolism: PE) with the potential of development as early as adolescence or younger adulthood. More than 100 different alterations have been identified throughout the gene producing either the more common type I defects (low antithrombin activity and antigen) or the rarer type II defects (dysfunctional protein with low activity and normal antigen).(2) Homozygous antithrombin deficiency appears to be incompatible with life.

The incidence of hereditary antithrombin deficiency is approximately 1:2000 to 1:3000 in general populations, although minor deficiency (antithrombin activity =70-75%) may be more frequent (approximately 1:350-650). In populations with venous thrombophilia, approximately 1% to 2% of individuals have antithrombin deficiency. Among the recognized hereditary thrombophilic disorders (including deficiencies of proteins C and S, as well as activated protein C: APC-resistance [factor V Leiden variant]), antithrombin deficiency may have the highest phenotypic penetrance (greater risk of venous thromboembolism). Arterial thrombosis (eg, stroke, myocardial infarction) has occasionally been reported in association with hereditary antithrombin deficiency.

Hereditary deficiency of antithrombin activity can also occur because of defective glycosylation of this protein in individuals with carbohydrate-deficient glycoprotein syndromes (CDGS).(3) Antithrombin activity assessment may be useful as an adjunct in the diagnosis and management of CDGS.

Acquired deficiency of antithrombin is much more common than hereditary deficiency. Acquired deficiency can occur...
due to:

- Heparin therapy (catalysis of antithrombin consumption)
- Intravascular coagulation and fibrinolysis (ICF) or disseminated intravascular coagulation (DIC), and other consumptive coagulopathies
- Liver disease (decreased synthesis and/or increased consumption) or with nephritic syndrome (urinary protein loss)
- L-asparaginase chemotherapy (decreased synthesis)
- Other conditions

In general, the clinical implications (thrombotic risk) of antithrombin deficiency in these disorders are not well defined, although antithrombin replacement in severe disseminated intravascular coagulation/intravascular coagulation and fibrinolysis (DIC/ICF) is being evaluated. Assay of antithrombin activity may be of diagnostic or prognostic value in some acquired deficiency states.

**Reference Values**

Normal values: 80-130%

Normal, full-term newborn infants have lower levels (> or ≥35-40%) that reach normal values by age 90 days. Premature infants (30-36 weeks gestation) have lower levels that reach normal values by age 180 days.

**Interpretation**

Antithrombin deficiencies due to inherited causes are much less common than those due to acquired causes (see Clinical Information). Diagnosis of hereditary deficiency requires clinical correlation, with the prospect of repeat testing (including antithrombin antigen assay), and family studies (with appropriate counseling). DNA-based diagnostic testing may be helpful, see ATTI / Antithrombin Deficiency, SERPINC1 Gene, Next-Generation Sequencing, Varies.

The clinical significance (thrombotic risk) of acquired antithrombin deficiency is not well established, but accumulating information suggests possible benefit of antithrombin replacement therapy in carefully selected situations.

Antithrombin deficiency, acquired or congenital, may contribute to the phenomenon of "heparin therapy resistance" (requirement of larger heparin doses than expected for achievement of therapeutic anticoagulation responses). However, it may more often have other pathophysiology, such as "acute-phase" elevation of coagulation factor VIII or plasma heparin-binding proteins.

Increased antithrombin activity is of unknown hemostatic significance. Direct factor Xa inhibitors, rivaroxaban (Xarelto), apixaban (Eliquis), and edoxaban (Savaysa) may falsely elevate the antithrombin activity and mask a diagnosis of antithrombin deficiency.

**Cautions**

Antithrombin functional result is affected by:

- Heparin (unfractionated or low-molecular-weight) >4 U/mL
- Alpha-1-antitrypsin >4 mg/mL
- Alpha-2-macroglobulin >10 mg/mL
Test Definition: ATTF
Antithrombin Activity, P

- Heparin cofactor II >4 U/mL
- Hemoglobin >500 mg/dL
- Bilirubin >40 mg/dL
- Triglycerides >2300 mg/dL

Heparin therapy may temporarily decrease plasma antithrombin activity into the abnormal range.

Antithrombin activity in serum specimens may be significantly lower than in plasma.

Clinical Reference

Performance

Method Description
Patient plasma, containing antithrombin, is mixed and incubated with reagent containing factor Xa and excess heparin. Factor Xa activity in the reagent is rapidly inhibited by antithrombin. Residual factor Xa activity is then measured using an amidolytic activity assay. This occurs when residual factor Xa lyses chromogenic substrate N-alpha-benzoyloxycarbonyl-D-arginyl-L-glycyl-L-arginine-p-nitroaniline-dihydrochloride and subsequently releases p-nitroaniline (detected at 405 nm) in a level that is inversely proportional to the amount of antithrombin in the sample. This method is based on inhibition of factor Xa and, therefore, only higher amounts of heparin cofactor II, alpha-2-macroglobulin, or alpha-1-antitrypsin will influence the assay. (Package insert: HemosIL Liquid Antithrombin. Instrumentation Laboratory Comp 06/2017)

PDF Report
No

Day(s) and Time(s) Test Performed
Monday through Friday

Analytic Time
1 day
Test Definition: ATTF
Antithrombin Activity, P

Maximum Laboratory Time
3 days

Specimen Retention Time
7 days

Performing Laboratory Location
Rochester

Fees and 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 Regional Manager. For assistance, contact Customer Service.

Test Classification
This test has been modified from the manufacturer's instructions. Its performance characteristics were determined by Mayo Clinic in a manner consistent with CLIA requirements. This test has not been cleared or approved by the U.S. Food and Drug Administration.

CPT Code Information
85300

LOINC® Information

<table>
<thead>
<tr>
<th>Test ID</th>
<th>Test Order Name</th>
<th>Order LOINC Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>ATTF</td>
<td>Antithrombin Activity, P</td>
<td>27811-9</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Result ID</th>
<th>Test Result Name</th>
<th>Result LOINC Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>ATTF</td>
<td>Antithrombin Activity, P</td>
<td>27811-9</td>
</tr>
</tbody>
</table>