MAYO CLINIC LABORATORIES

Test Definition: PSTF

Protein S Antigen, Plasma

Overview

Useful For

Investigation of patients with a history of thrombosis

Profile Information

Test Id	Reporting Name	Available Separately	Always Performed
PSF	Protein S Ag, Free, P	No	Yes

Reflex Tests

Test Id	Reporting Name	Available Separately	Always Performed
PST	Protein S Ag, Total, P	No	No

Testing Algorithm

If free protein S antigen is decreased, then total plasma protein S antigen will be performed at an additional charge.

Special Instructions

- Coagulation Guidelines for Specimen Handling and Processing

Method Name

PSF, PST: Latex Immunoassay (LIA)

NY State Available

No

Specimen

Specimen Type

Plasma Na Cit

Specimen Required

Specimen Type: Platelet-poor plasma

Patient Preparation:

1. Patient **should not** be receiving anticoagulant treatment (eg, warfarin, heparin). Warfarin will lower protein S. If not possible for medical reasons, note on request.

a. If medically feasible, for 4 to 6 hours before specimen collection, **do not** administer intravenous heparin.

b. If medically feasible, for 10 to 14 days before specimen collection, **do not** administer subcutaneous heparin or warfarin**.**

2. Patient **should not** be receiving fibrinolytic agents (streptokinase, urokinase, tissue plasminogen activator [tPA]).
3. It is recommended that specimens be collected pretransfusion. If patient has been transfused, **a specimen should not be collected for 48 hours.**
Collection Container/Tube: Light-blue top (3.2% sodium citrate)
Submission Container/Tube: 13-mm x 75-mm, 5-mL, plastic vials (polypropylene preferred)
Specimen Volume: 1 mL Platelet-poor plasma in 2 plastic vials, each containing 0.5 mL
Collection Instructions:
1. For complete instructions, see Coagulation Guidelines for Specimen Handling and Processing.
2. Centrifuge, transfer all plasma into a plastic vial, and centrifuge plasma again.
3. Aliquot 0.5 mL of plasma into 2 plastic vials, leaving 0.25 mL in the bottom of centrifuged vial.
4. Immediately freeze plasma (no longer than 4 hours after collection) at -20 degrees C or, ideally at -40 degrees C or below.
5. Send specimens in the same shipping container.
Additional Information: A double-centrifuged specimen is critical for accurate results as platelet contamination may cause spurious results.

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

Specimen Minimum Volume
Platelet-poor plasma: 0.5 mL

Reject Due To

Gross hemolysis	Reject
Gross lipemia	Reject
Gross icterus	Reject

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Plasma Na Cit	Frozen	14 days	

Clinical & Interpretive

Clinical Information
Protein S is a vitamin K-dependent glycoprotein present in platelets and synthesized within the liver and endothelial cells. Protein S works as part of the natural anticoagulant system by acting as a cofactor to activated protein C (APC) in the proteolytic inactivation of procoagulant factors Va and VIIIa. In addition, protein S has direct APC-independent anticoagulant activity by inhibiting formation of the prothrombin and tenase complexes, possibly due to its high affinity for anionic phospholipid membranes. In human plasma, protein S forms a complex with the compliment regulatory protein, C4b-binding protein (C4bBP). Of the total plasma protein S, approximately 60% circulates bound to C4bBP while the remaining 40% circulates as "free" protein S. Only free protein S has anticoagulant function. C4bBP is composed of 6

or 7 alpha-chains and 1 or no beta-chain (C4bBP-beta). Different C4bBP isoforms are present in plasma, but only C4bBP-beta binds protein S.

Congenital protein S deficiency is an autosomal dominant disorder that is present in 2% to 6% of patients with venous thrombosis. Patients with protein S deficiency have an approximately 10-fold increased risk of venous thrombosis. In addition, they may also experience recurrent miscarriage, complications of pregnancy (preeclampsia, abruptio placentae, intrauterine growth restriction, and stillbirth) and possibly arterial thrombosis.

Three types of protein S deficiency have been described according to the levels of total protein S antigen, free protein S antigen, and protein S activity in plasma. Types I and III protein S deficiency are much more common than type II (dysfunctional) protein S deficiency. Type III protein S deficiency appears to be partly due to variants within the protein S binding region for C4bBP-beta.

Homozygous protein S deficiency is rare but can present as neonatal purpura fulminans, reflecting severe disseminated intravascular coagulation/intravascular coagulation and fibrinolysis (DIC/ICF) caused by the absence of plasma protein S.

Acquired deficiency of protein S has causes that are generally of unknown hemostatic significance (ie, uncertain thrombosis risk) and is much more common than hereditary protein S deficiency. Acquired protein S deficiency can present through vitamin K deficiency, oral anticoagulant therapy, liver disease, DIC/ICF, thrombotic thrombocytopenia purpura, pregnancy, estrogen therapy, nephritic syndrome, and sickle cell anemia. As an acute-phase reactant, plasma C4bBP levels increase with acute illness and may cause acquired free protein S deficiency.

Measurement of plasma free protein S antigen is performed as the initial testing for protein S deficiency. When the free protein S antigen level is below the age- and sex-adjusted normal range, reflexive testing will be performed for total plasma protein S antigen.

Reference Values

TOTAL
Males: 80-160%
Females:
<50 years: 70-160%
> or =50 years: 80-160%

FREE
Males: 65-160%
Females:
<50 years: 50-160%
> or =50 years: 65-160%

Normal, full-term infants or healthy premature infants may have decreased levels of total protein S (15-50%); but because of low levels of C4b-binding protein, free protein S may be normal or near the normal adult level (> or =50%). Total protein S reaches adult levels by 90 to 180 days postnatal.*

*See Pediatric Hemostasis References section in Coagulation Guidelines for Specimen Handling and Processing

Interpretation

Protein S values vary widely in the normal population and are age- and sex-dependent.

Table. Types of Heterozygous Protein S Deficiency

Type	Protein S antigen free	Protein S antigen total	Protein S activity
I	Low	Low	Low
II	Normal	Normal	Low
III	Low	Normal	Low

Protein S and C4b-binding protein (C4bBP) are coordinately regulated, and an increased total protein S antigen and low free protein S antigen most commonly reflect acute or chronic inflammation or illness with an associated increase in plasma C4bBP.

For patients in whom hereditary protein S deficiency is strongly suspected and the free plasma protein S antigen level is normal, consideration should be given to testing of free protein S activity, S_FX / Protein S Activity, Plasma, for detecting type II protein S deficiency (which is rare).

An increased total protein S antigen is of uncertain clinical significance because free protein S antigen levels are usually normal, in such situations. However, the total protein S antigen level may be helpful in distinguishing acquired versus congenital protein S deficiency. High normal or increased total protein S antigen and reduced free protein S antigen suggests acquired protein S deficiency, as may be seen in pregnancy or inflammation. In contrast, low normal or decreased total protein S antigen and reduced free protein S antigen suggests vitamin K deficiency or a warfarin (Coumadin) effect but also could reflect congenital protein S deficiency (type I or III).

Vitamin K deficiency, oral anticoagulant therapy, presence of liver disease, or disseminated intravascular coagulation/intravascular coagulation and fibrinolysis are common acquired causes of protein S deficiency, which is of uncertain significance when such conditions are present. Concomitant assay of coagulation factor II activity may be helpful in differentiating congenital protein S deficiency from oral anticoagulation effects, but supportive data are currently suboptimal.

Differentiation of congenital and acquired protein S deficiency requires clinical correlation and may require repeated laboratory study of the patient and selected family members in some instances. DNA-based testing may be helpful; see GNPRS / Protein S Deficiency, *PROS1* Gene, Next-Generation Sequencing, Varies.

Cautions

Total protein S antigen results are potentially affected by:
-Heparin (unfractionated or low-molecular-weight) >4 U/mL
-Hemoglobin >2 g/L
-Bilirubin >100 mg/L
-Rheumatoid factor >300 IU/mL; may lead to an overestimation of the result
-Antirabbit antibodies; certain subjects may have aberrant results
-Lipemic specimen may lead to an overestimation of level

Free protein S antigen results are potentially affected by:
-Heparin (unfractionated or low-molecular-weight) >4 U/mL

-Hemoglobin >200 mg/dL
-Bilirubin >25 mg/dL
-Triglycerides >1500 mg/dL
-Platelets >10(10)/L
-Rheumatoid factor >900 IU/mL
-Factor V Leiden variant (activated protein C resistance, APC-R)

Supportive Data

A retrospective review of Mayo Special Coagulation Laboratory data found that of 584 patients tested sequentially, only 4 patients demonstrated a pattern of normal free protein S antigen with decreased total protein S antigen. Three of these patients were receiving oral anticoagulant therapy and 1 had liver disease. There were 8 patients with probable congenital protein S deficiency. Of this group, all had significantly reduced levels of free protein S antigen and normal or mildly reduced levels of total protein S antigen. We conclude that omission of routine measurement of total protein S antigen and substituting measurement of free protein S antigen with reflexive testing of total protein S antigen only for decreased free protein S antigen would not decrease clinical sensitivity of this assay system for detecting hereditary protein S deficiency.

Clinical Reference

1. Borgel D, Gandrille S, Aiach M. Protein S deficiency. Thromb Haemost. 1997 July;78(1):351-356
2. De Stefano V, Finazzi G, Mannucci PM. Inherited thrombophilia: pathogenesis, clinical syndromes, and management. Blood. 1996 1;87(9):3531-3544
3. Zoller B, Garcia de Frutos P, Dahlback B. Evaluation of the relationship between protein S and C4b-binding protein isoforms in hereditary protein S deficiency demonstrating type I and type III deficiencies to be phenotypic variants of the same genetic disease. Blood. 1995;85(12):3524-3531
4. Grandrille S, Borgel D, Ireland H, et al. Protein S deficiency: a database of mutations. For the Plasma Coagulation Inhibitors Subcommittee for the Scientific and Standardization Committee of the International Society on Thrombosis and Haemostasis. Thromb Haemost. 1997;77(6):1201-1214
5. Wolf M, Boyer-Neumann C, Peynaud-Debayle E, Marfaing-Koka A, Amiral J, Meyer D. Clinical applications of a direct assay of free protein S antigen using monoclonal antibodies. A study of 59 cases. Blood Coagul Fibrinolysis. 1994;5(2):187-192
6. Laroche P, Plassart V, Amiral J. Rapid quantitative latex immunoassays for diagnosis of thrombotic disorders. Thromb Haemost. 1989:62:379
7. Goodwin AJ, Rosendaal FR, Kottke-Marchant K, Bovill EG. A review of the technical, diagnostic, and epidemiologic considerations for protein S assays. Arch Pathol Lab Med. 2002;126(11):1349-1366
8. Sales M, Begona A, Rosen S. IL Test Free Protein S: A diagnostic tool for protein S deficiency. Instrumentation Laboratories; Hemostasis Monograph
9. Serra J, Sales M, Chitolie A, et al. Multicentre evaluation of IL Test Free PS: a fully automated assay to quantify free protein S. Thromb Haemost. 2002;88(6):975-983
10. Marlar RA, Gausman JN, Tsuda H, Rollins-Raval MA, Brinkman HJM. Recommendations for clinical laboratory testing for S deficiency: Communication from the SCC committee plasma coagulation inhibitors of the ISTH. J Thromb Haemost. 2021;19(1):68-74

Performance

Method Description

Total

This assay is performed using the Diagnostica Stago LIATEST Protein S Kit on the Beckman Coulter ACL TOP. Protein S total antigen is determined using automated latex immunoassay methodology. This methodology is comprised of a reagent with microlatex particles coated with specific antihuman total protein S antibodies. Patient plasma containing total protein S antigen is combined with the latex reagent causing the antibody-coated latex particles to agglutinate and form aggregates. The aggregates form diameters greater than the wavelength of the light (405 nm) passing through causing absorption of the light. This change in absorption is measured over time and reported as delta optical density (OD). The increase in absorption is proportional to the concentration of protein S total antigen present in the patient plasma.

Free

This assay is performed using the HemosIL Free Protein S kit on the Beckman Coulter ACL TOP. The assay uses latex immunoassay methodology to determine the presence of free protein S. It consists of 2 latex reagents, one being latex particles coated with purified human C4BP and the other is latex particles coated with a monoclonal antibody directed against human protein S. Patient plasma is combined with the purified C4BP which reacts with a high affinity for free protein S in the patient plasma. The free protein S adsorbed on the C4BP latex triggers the agglutination reaction with the second latex reagent. The aggregates form diameters greater than the wavelength of the light (405 nm) passing through causing absorption of the light. This change in absorption is measured over time and reported as delta OD. The increase in absorption is proportional to the concentration of free protein S antigen present in the patient plasma.

PDF Report

No

Day(s) Performed

Monday through Friday

Report Available

1 to 3 days

Performing Laboratory Location

Mayo Clinic Jacksonville Clinical Lab

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 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 US Food and Drug Administration.

CPT Code Information

85306-Free

85305-Total (if appropriate)

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
PSTF	Protein S Ag, P	87557-5

Result ID	Test Result Name	Result LOINC® Value
PSF	Protein S Ag, Free, P	27821-8