

*BloodCenter of Wisconsin offers three Platelet Factor Four
Enzyme Linked Immunosorbent Assays (PF4 ELISA) for detection of
Heparin-Dependent Platelet Antibodies.*

BACKGROUND:

Heparin-induced thrombocytopenia (HIT) associated with thrombosis is an immune complex mediated disorder that can cause morbidity and mortality in patients receiving heparin therapy. Prompt diagnosis is paramount to appropriate patient management. The diagnosis of HIT is suspected when:

- a sustained decline in the platelet count occurs during heparin therapy
- the platelet count recovers after heparin is discontinued and
- no other causes of thrombocytopenia are evident.

METHOD:

The PF4 ELISA is a solid phase assay based on the reaction of patient antibody with complexes of PF4 and a polyanionic compound (PVS), which is used as a substitute for unfractionated heparin (UFH). Bound patient antibody is detected with a secondary antibody specific for human IgG or IgA or IgM. Positive reactions are confirmed by demonstrating inhibition of antibody binding with excess heparin.

LIMITATIONS:

- Weakly reactive heparin-dependent antibodies, while not present in normal control sera, can be detected in the sera of patients with recent or current exposure to heparin but have a lower likelihood of being associated with clinical HIT compared with more potent antibodies.
- The assays are not appropriate for detection of antibodies with low molecular weight heparin.

REASONS FOR REFERRAL:

PF4 ELISA - IgG - Detection of heparin-dependent platelet antibodies in patients demonstrating clinical findings consistent with HIT with or without thrombosis.

PF4 ELISA - IgA and PF4 ELISA - IgM - Detection of rare clinically relevant antibodies of the IgM and IgA isotypes in patients demonstrating compelling clinical findings consistent with HIT, yet have a weak or negative result in the PF4 ELISA - IgG.

REFERENCE INTERVAL:

- Positive - Optical density value > 0.400 that inhibits by 49% or more for IgG and IgA and 30% or more for IgM in the presence of high dose (100 U/ml) UFH.
- O.D. values are reported. The first value demonstrates antibody binding, and the second value demonstrates inhibition by heparin.
- Results will be reported as negative, see interpretation, positive, or strong positive for heparin-dependent antibody.

SPECIMEN REQUIREMENTS:

5 ml refrigerated serum

Sample must be received within 7 days of draw date. Older samples are acceptable if they have been frozen.

SHIPPING REQUIREMENTS:

Place the specimen and the test requisition form in sealed plastic bags, and surround with cold packs in a Styrofoam container. Place the sealed Styrofoam container in a sturdy cardboard box and tape securely. Ship the package in compliance with your overnight carrier guidelines. Label with the following address:

Client Services/Platelet and Neutrophil Immunology Laboratory
BloodCenter of Wisconsin
638 N. 18th St.
Milwaukee, WI 53233
800-245-3117 x6250

TURNAROUND TIME:

Heparin Dependent Platelet Antibody - IgG (PF4 ELISA): 1-2 days

Heparin Dependent Platelet Antibody - IgA (PF4 ELISA): 2-4 days

Heparin Dependent Platelet Antibody - IgM (PF4 ELISA): 2-4 days

CPT CODES:

Heparin Dependent Platelet Antibody - IgG (PF4 ELISA): 86022

Heparin Dependent Platelet Antibody - IgA (PF4 ELISA): 86022

Heparin Dependent Platelet Antibody - IgM (PF4 ELISA): 86022

REFERENCES:

- 1) Sheridan D, Carter C, Kelton JG. A diagnostic test for heparin-induced thrombocytopenia. *Blood* 1986;67:27-30.
- 2) Eichler P, Rashke R, Lubenow N, Meyer O, Shwind P, Greinacher A. The new ID-heparin/PF4 antibody test for rapid detection of heparin-induced antibodies in comparison with functional and antigenic assays. *Brit J Haematol* 2002;116:887-891.
- 3) Arepally G, et al. Comparison of PF4/heparin ELISA assay with the 14C-serotonin release assay in the diagnosis of heparin-induced thrombocytopenia. *AJCP* 1995;104:648-654.
- 4) Collins JL, Aster RH, Moghaddam M, Piotrowski M, Strauss TR, McFarland JG. Diagnostic testing for heparin-induced thrombocytopenia (HIT): an enhanced platelet factor 4 complex enzyme linked immunosorbent assay (PF4 ELISA). *Blood* 1997;90 (Suppl 1):461a.
- 5) Visentin TP, Ford SE, Scott JP, Aster RH. Antibodies from patients with heparin induced thrombocytopenia/thrombosis are specific for platelet factor 4 complexed with heparin or bound to endothelial cells. *JCI* 1994, 93:81-88.
- 6) Eichler JD, et al. Incidence and clinical significance of anti-PF4/heparin antibodies of the IgG, IgM and IgA class in 755 consecutive patient samples referred for diagnostic testing for heparin-induced thrombocytopenia. *Eur J Haematol* 2006; 76:420
- 7) Warkentin TE, et al. Laboratory testing for the antibodies that cause heparin-induced thrombocytopenia: How much class do we need? *J Lab Clin Med* 2005; 146:341-346
- 8) Greinacher A, et al. Heparin-induced thrombocytopenia: A prospective study on the incidence, platelet-activating capacity and clinical significance of anti-PF4/heparin antibodies of the IgG, IgM, and IgA classes. *J Thromb Haemost* 2007; 5:1666-73
- 9) Warkentin TE, et al. Quantitative interpretation of optical density measurements using PF4-dependent enzyme immunoassays. *J Thromb haemost* 2008, 6:1304-1312
- 10) Lo GK, et al. What is the potential for over-diagnosis of heparin-induced thrombocytopenia? *Am J or Hematol.* 2007; 82:1037-1043
- 11) Lo GK, et al. Evaluation of pretest clinical score (4 T's) for the diagnosis of heparin-induced thrombocytopenia in two clinical settings. *J Thromb Haemost* 2006; 4:759-765
- 12) Visentin GP, et al. Heparin is not required for detection of antibodies associated with heparin-induced thrombocytopenia/thrombosis. *J Lab Clin Med* 2001; 138:22-31
- 13) Sheridan D, et al. A diagnostic test for heparin-induced thrombocytopenia. *Blood* 1986; 67:27-30
- 14) Amiral J, et al. Pathogenicity of IgA and/or IgM antibodies to heparin-PF4 complexes in patients with heparin-induced thrombocytopenia. *British Journal of Haematology* 1996; 92:954-9