Heparin-induced Thrombocytopenia (HIT)

Introduction

Heparin-induced thrombocytopenia (HIT) is a potentially catastrophic, antibody-mediated complication of heparin therapy caused by antibodies directed against platelet factor 4 (PF4) complexed with heparin or other polyanions.[1-7] HIT antibodies bind to PF4/heparin complexes on the platelet surface, resulting in platelet activation leading to thrombocytopenia that can be accompanied by life-threatening thrombosis, including ischemic limb necrosis, pulmonary embolism, myocardial infarction and stroke.

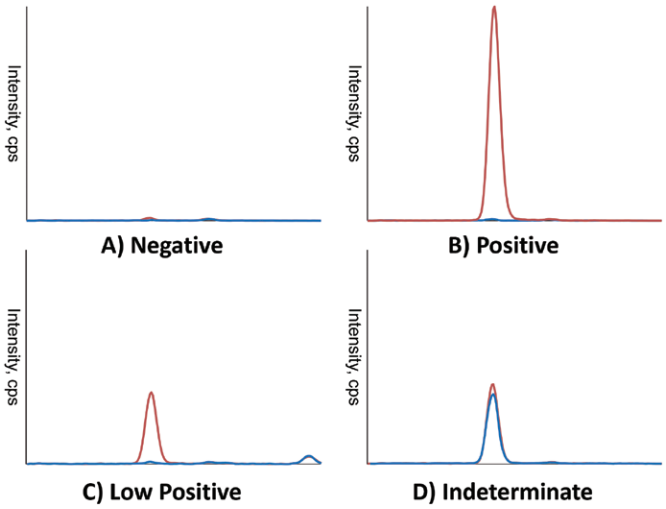
Moderate thrombocytopenia is common in the clinical settings where heparin is administered and most cases are not caused by antibodies to PF4/heparin complexes.[2-6] Differentiation of HIT from other potential causes of thrombocytopenia is difficult diagnostically and relies on a combination of clinical assessment and laboratory investigation.[2,6] Prompt diagnosis and management of HIT is critical to minimize the risk of life-threatening thrombosis. Patients diagnosed with or suspected of suffering from HIT must strictly avoid exposure to heparin (including heparin administered therapeutically or prophylactically, as well as that used to maintain patency of intravenous access and any catheter that may be heparin-coated) and transitioned to an alternative nonheparin anticoagulant as quickly as possible to prevent thromboembolic complications.

Laboratory method

Labcorp's **Heparin-dependent Platelet Antibody (Serotonin Release Assay) [150018]**, or SRA, employs washed donor platelets and detects their activation by measuring the release of endogenous serotonin that is induced by addition of patient serum in the presence of heparin. Serotonin levels are measured by a highly sensitive liquid chromatography tandem mass spectrometry method (LC/MS-MS). The classical signature for HIT is demonstrated via release of serotonin from platelets following coincubation with low-dose (therapeutic) heparin accompanied by a lack of serotonin release following coincubation with high-dose (supratherapeutic) heparin. A true HIT antibody causes serotonin release from platelets at therapeutic heparin concentrations. Much higher (supratherapeutic) heparin concentrations in the reaction mixture should prevent true HIT antibodies from complexing on the platelet surface, thus inhibiting the release of serotonin. Testing at both therapeutic and supratherapeutic heparin concentrations provides for optimal sensitivity and specificity of the assay. Patient specimen test results will produce one of four potential diagnostic signatures displayed in Figure 1, A-D. Each figure shows serotonin release in the presence of low heparin (in red) and high heparin (in blue). The accompanying chromatograms depict example serotonin release profiles for:

- **A negative SRA sample:** 1% serotonin release in low heparin and 0% release in high heparin
- **A positive SRA sample:** 83% serotonin release in low heparin and 2% release in high heparin
- **A low positive SRA sample:** 30% serotonin release in low heparin and 1% release in high heparin
- **An indeterminate SRA sample:** 31% serotonin release in low heparin and 29% release in high heparin

Figure 1: Characteristic Serotonin Release (%) Results in the Presence of Lowand High Dose Heparin Concentrations Measured by LC/MS-MS



The SRA requires 1 mL of serum frozen in a plastic transport tube. Results are interpreted as negative, indeterminate, low positive or positive. Percent endogenous serotonin release with low-dose and high-dose heparin are reported. A positive result requires >20% release of serotonin with low-dose heparin and ≤20% release in the presence of a high-dose of heparin. The result is considered low positive when the serotonin release in the presence of low-dose heparin falls between ≥20% and ≤30%.

Clinical Application

The investigation of HIT is challenging and requires correlation between clinical symptoms and laboratory assays. The most common assay performed is a serologic assay that detects the presence of HIT antibodies without regard for their functional ability. Several serologic assays that are relatively easy to perform are available commercially, and these assays are highly sensitive. The results of these assays have excellent negative predictive values, and a negative result can be used to exclude HIT in all but the most compelling clinical circumstances.[6] However, these assays suffer from low specificity and frequently yield positive results in the absence of clinical HIT. A positive result, especially of low titer, does not differentiate between pathogenic antibodies and clinically irrelevant antibodies.[6] Functional assays that measure platelet activation by HIT antibodies in the presence of heparin are considered gold-standard diagnostic laboratory tests due to their ability to detect the pro-coagulant, platelet activating potential of a patient's serum.[7] Due to complexity of performance, functional assays that use washed platelets are not widely available. The serotonin release assay using washed platelets is a well-established functional HIT assay that is both highly sensitive and specific for HIT.[1-6] Currently recommended HIT diagnostic algorithms published by the American Society for Hematology, British Committee for Standards in Haematology (BCSH) and American College of Chest Physicians Evidence-Based Clinical Practice Guidelines incorporate an estimate of clinical probability and use of a sensitive immunoassay to guide initial management with subsequent confirmatory testing by a more specific functional assay.[2,8,9]

Labcorp's SRA is a confirmatory assay that can be used in the evaluation of heparin-induced thrombocytopenia (HIT). Patients clinically suspected of having HIT, with a positive immunologic HIT assay, especially at high titer, may be tested with this confirmatory SRA assay. This assay should not be used as a screening assay for HIT. The diagnosis of heparin-induced thrombocytopenia should not be based on the results of the SRA only and should take into account the results of the immunologic HIT assay, presence and timing of thrombocytopenia in relation to heparin administration, the presence of thrombosis or other HIT-related sequelae, as well as determination of other causes of thrombosis. Release of native serotonin from platelets is less susceptible to interference from HLA class 1 antibodies than radio-labeled serotonin that is taken up by donor platelets.

Coagulation testing Labcorp offers

Test Name	Test No.
Heparin-dependent Platelet Antibody (Serotonin Release Assay)	**150018**
Heparin-induced Platelet Antibody (HIPA)	**150075**
Heparin-induced Platelet Antibody With Reflex to Serotonin Release Assay	**150031**

*For complete testing details, including specimen requirements and CPT codes, please consult the online Test Menu at www.labcorp.com.

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