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PEARLS OF LABORATORY MEDICINE

Heparin-Induced Thrombocytopenia

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Heparin-Induced Thrombocytopenia (HIT)

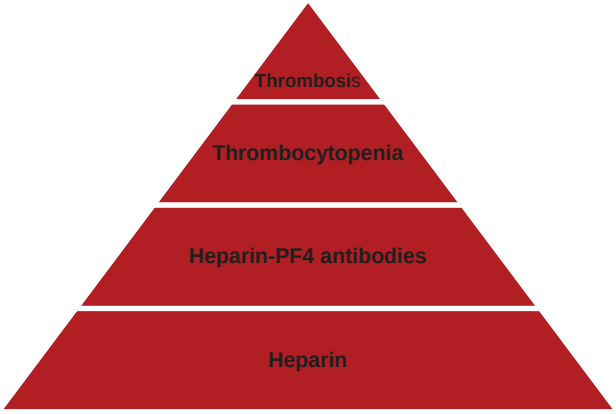
What is HIT?

- Immune mediated syndrome that occurs in a small percentage (1-5%) of heparin-exposed patients
- Caused by IgG antibodies to heparin-platelet factor 4 (PF4) complexes
- Immune complexes cause platelet activation
  - Thrombocytopenia
  - High risk of arterial or venous thrombosis
    - ~50% if untreated

Heparin-Induced Thrombocytopenia (HIT)

Pathogenesis

- PF4 released from platelet granules is positively charged
  - Able to form complexes with negatively charged heparin
- Heparin-PF4 complexes are antigenic in some patients, resulting in antibody formation
  - Immune complexes composed of IgG-heparin-PF4 activate platelets via the platelet Fcγ receptor for IgG



Heparin-Induced Thrombocytopenia (HIT)

Risk of HIT

- Higher risk in surgical patients than in medical patients
  - Related to amount of circulating PF4 and type and dose of heparin
  - Low risk in pregnancy
- Higher risk with unfractionated heparin (UFH) than low-molecular-weight heparin (LMWH)
  - Related to likelihood of complex formation, creation of HIT antigen

Heparin-Induced Thrombocytopenia (HIT)

HIT Diagnosis - Clinical

- Suspected when patient develops thrombocytopenia 5-10 days after heparin exposure
- Evaluation of clinical pre-test probability using clinical scoring system such as the 4T's system
  - Excellent negative predictive value (NPV) for low scores
    - NPV 97-99%
  - Poor positive predictive value (PPV) for intermediate or high scores
    - PPV for intermediate 10-20%
    - PPV for high 40-80%

Heparin-Induced Thrombocytopenia (HIT) 4T's scoring system

Thrombocytopenia
- 0 points; <30% drop or nadir <10 x 10^9/L
- 1 point; 30-50% drop or nadir 10-19 X 10^9/L
- 2 points; >50 drop and nadir >=20 X 10^9/L

Timing of platelet drop
- 0 points; <4 days with no recent exposure
- 1 point; suspect 5-10 days but not documented, >10 days, <=1 day with last exposure 30-100 days prior
- 2 points; 5-10 days or <= 1 day if exposed in past 30 days

Thrombosis
- 0 points; none
- 1 point; progressive, recurrent, silent, suspected but not proven
- 2 points; proven new thrombosis or skin necrosis or acute systemic reaction to heparin

Other causes of thrombocytopenia
- 0 points; definite
- 1 point; possible
- 2 points; none

Scoring
Low = <4 points
Intermediate = 4-6 points
High = 6-8 points

Heparin-Induced Thrombocytopenia (HIT)

HIT Diagnosis - Laboratory

- 2 categories of tests
  - Immunoassays to detect heparin-PF4 antibodies
    - ELISA is most common
  - Functional assays to detect platelet activation by HIT immune complexes

Heparin-Induced Thrombocytopenia (HIT)

HIT Diagnosis - ELISA

- Plates coated with heparin-PF4
- Detect antibodies in patient serum
  - IgG/IgM/IgA – polyspecific
  - IgG only – monospecific
- Diagnostic utility
  - Excellent NPV (~99%)
  - High sensitivity (95-100%)
  - Poor specificity (<90%, possibly well below 90%) and poor PPV since many patients who develop antibodies do not develop the HIT syndrome
  - Specificity somewhat improved with monospecific IgG ELISA

Heparin-Induced Thrombocytopenia (HIT)

HIT Diagnosis - ELISA

- Specificity improved by considering the optical density (OD) value of positive results
- Higher OD values correlate with capacity for platelet activation
  - 0.4 to 1.0: ~5% probability of HIT
  - 1.0 to 1.5: ~25% probability of HIT
  - >=2.0: ~90% probability of HIT
- Unclear if repeating positive samples in the presence of a high heparin concentration significantly improves ELISA specificity
  - Possible risk of false-negative results

Heparin-Induced Thrombocytopenia (HIT)

HIT Diagnosis – Functional platelet activation assays

- Sensitive (>90%) and more specific (>90%) for HIT than ELISA due to demonstration of antibody platelet activating properties
- Serotonin release assay (SRA) is the most commonly used test and golden standard
  - Further evaluate unexpected ELISA results
  - Further evaluate positive ELISA results

Heparin-Induced Thrombocytopenia (HIT)

HIT Diagnosis – SRA

- Combine patient serum with donor platelets and heparin
  - HIT positive sera result in platelet activation and release of serotonin from dense granules
  - Released serotonin is detected and quantified as an indicator of the degree of platelet activation
    - A variety of methods can be used to quantify serotonin

Heparin-Induced Thrombocytopenia (HIT)

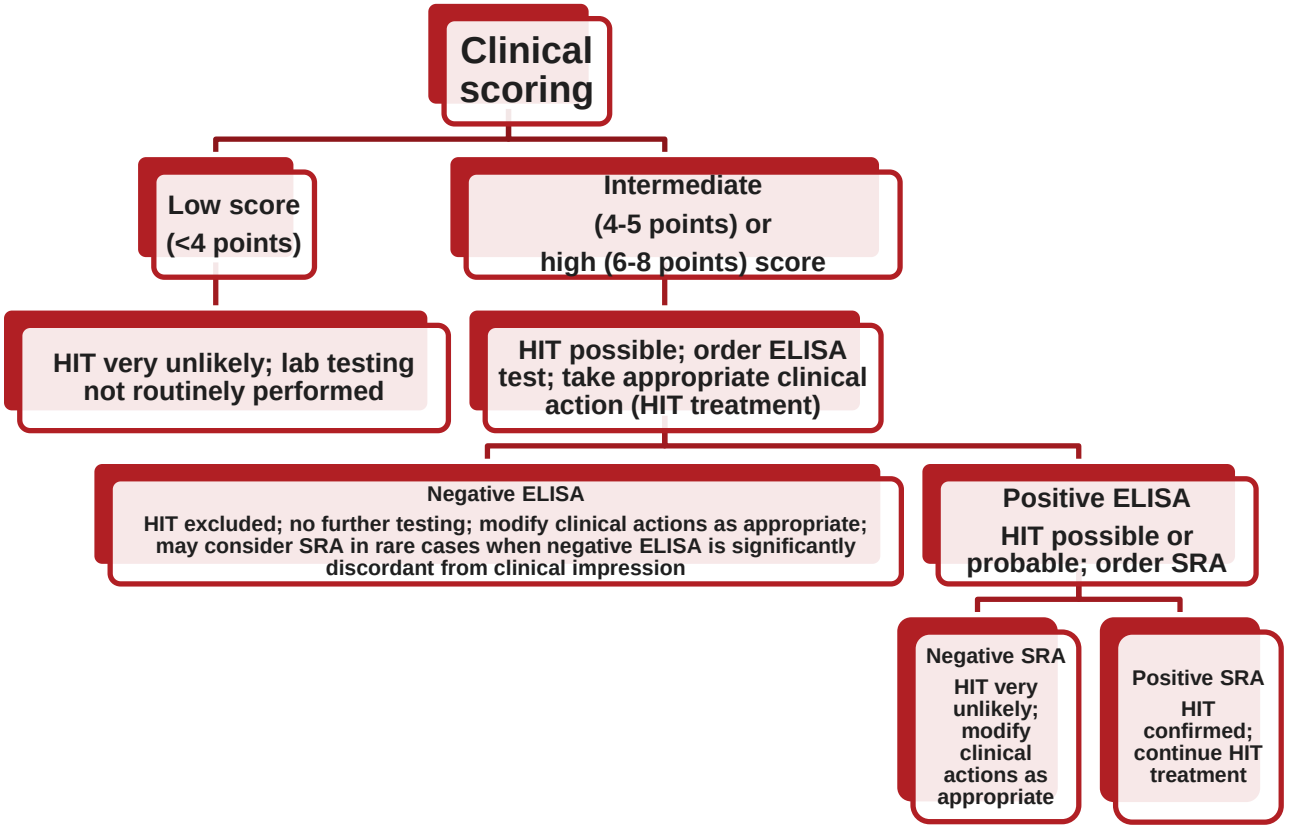
HIT Diagnosis – SRA results

- Released serotonin expressed as % release
  - Amount of serotonin released divided by total platelet serotonin X 100
  - >= 20% release represents platelet activation
- Each patient specimen is tested at 2 different heparin concentrations and % release values are obtained
  - Low heparin concentration (LH) – therapeutic, supports immune complex formation
  - High heparin concentration (HH) – supra-therapeutic, disrupts immune complex formation

Heparin-Induced Thrombocytopenia (HIT) SRA reaction patterns

LH % Release	HH % Release	Classification
>=20%	<20%	Positive
<20%	<20%	Negative
>=20%	>=20%	Indeterminate

Heparin-Induced Thrombocytopenia (HIT)
Diagnostic algorithm for suspected HIT



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Disclosures/Potential Conflicts of Interest

Upon Pearl submission, the presenter completed the Clinical Chemistry disclosure form. Disclosures and/or potential conflicts of interest:

- **Employment by the University of Utah Department of Pathology and ARUP Laboratories**

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