Review

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Heparin-Induced Thrombocytopenia (HIT) Syndrome

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Abstract

Heparin has been available as a clinical treatment and prevention for thromboembolic disease for half a century. Known complications of heparin therapy include bleeding, allergic reactions, osteoporosis, and thrombocytopenia. Heparin-induced thrombocytopenia (HIT) is a common and potentially grave adverse effect of heparin treatment. HIT is unusual among drug-induced thrombocytopenias in that it is more apt to cause thrombosis than bleeding. HIT-associated thrombosis can result in arterial and venous thrombosis leading to stroke, myocardial infarction, limb gangrene, amputation and even death. HIT pathogenesis is thought to involve antibody binding to an epitope on the platelet factor 4 (PF4)-heparin complex. The antibody bound complex then binds FcγRII receptors on the platelet surface, which activates blood-coagulation pathways and concomitantly produces extensive platelet activation and aggregation. HIT diagnosis is based on the presence of thrombosis and diagnostic laboratory tests including immunoassays for HIT antibodies and functional tests, such as the ^{14}C-serotonin release assay. Heparin treatment should be withdrawn immediately upon diagnosis of HIT, and the patient should be subjected to an alternative treatment for at least 5 days unless the HIT diagnosis is disproven. Once the patient has been stabilized, warfarin treatment should commence while the patient is still receiving the alternative anticoagulant therapy.

Key words: Thrombocytopenia, heparin, heparin-induced thrombocytopenia, thrombosis.

Introduction

There are two types of heparin-induced thrombocytopenia (HIT) (Table 1) with distinct etiologies. HIT type I is an early-onset thrombocytopenia that is rarely associated with clinical complications and generally resolves without any change in treatment and the mechanism of the thrombocytopenia is nonimmune in nature. (1) HIT type II is a clinical-pathological syndrome which is strongly associated with thrombotic events and requires immediate heparin withdrawal often with administration of an alternative anticoagulant therapy. (2) HIT type II is also known as heparin-associated thrombocytopenia, heparin-associated thrombocytopenia and thrombosis, and white clot syndrome. (3) White clot syndrome refers to the platelet-rich arterial thrombosis (rather than a fibrin-rich venous thrombosis) that is often observed in patients with HIT type II. (4) For the remainder of this review, the term HIT will refer selectively to HIT type II. HIT can be defined as an unexplained thrombocytopenia at 50% or lower of previously determined platelet counts during exposure to unfractionated heparin (UFH) or low molecular weight heparin (LMWH), (5) accompanied by the presence of antibodies against the heparin-platelet factor 4 (PF4) complex. (6)

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Epidemiology

The clinically reported incidences of HIT vary widely. In a review of prospective trials, the reported estimated frequencies of HIT ranged from 1 to 30% among patients receiving UFH and were about 2% in patients receiving LMWH. (7) In a more rigorous review, the estimated incidence was less than 3% with UFH, and 0% with LMWH. (8) The high variability in the incidence of HIT could be due to differences in the heparin preparations, differences in the diagnostic serologic assay used and differences in the patient populations. **Table 2** summarizes the incidence of HIT relative to cohort demographics and heparin treatment type based on prospective studies that used established definition of HIT (≥50% platelet count decrease or thrombotic event, with a positive antibody assay or satisfactory clinical criteria for HIT).

Most patients that develop HIT were administered intravenous or subcutaneous heparin therapy prophylactically or as treatment for a thrombotic event. (8) HIT can result from delivery of a relatively small quantity of heparin. Indeed patients have been known to develop HIT after receiving as little as 250 U from a heparin flush or even after being fit with heparin-coated catheters. (9,10)

HIT occurrence is affected by several factors including the type of heparin administered (bovine UFH > porcine UFH > LMWH), the duration of heparin administration (higher risk from days 5 to 14), gender (female > male), (4) the HIT definition adopted, and the type of test used for detecting heparin-dependent antibodies. (11) As shown in **Table 2** as much as 6.5% of orthopedic surgery patients receiving UFH develop HIT, (5,12) while ≤ 1% of patients develop HIT under other circumstances. (11-18) A recent meta-analysis determined that the overall risk of developing HIT after the prophylactic use of UFH or LMWH was 2.6% and 0.2%, respectively, in medical and surgical patients. (19) In a prospective cohort study involving 1754 consecutive medical patients treated with LMWH, the incidence of HIT was 0.8 percent. (20) HIT developed more often in patients with prior heparin exposure (1.7% vs 0.3% with no prior exposure; OR 4.9; 95% CI 1.5-16). An analysis of data from seven prospective studies determined that HIT risk was higher with UFH than with LMWH (RR 5.3; 95% CI 2.8-9.9), higher in surgical than in medical patients (RR 3.2; 95% CI 2.0-5.4), and higher in females than in males (RR 2.4; 95% CI 1.4-4.1). (20) The patient group with the highest risk for HIT was female surgical patients receiving UFH (RR 17; 95% CI 4.2-72).

Pathogenesis

HIT pathogenesis can be described as a three-step process: first, antibodies are generated (immune response); second, platelets are activated and thrombin generation is increased (the hypercoagulable state); third, in some patients, thrombosis occurs, elevating the condition to heparin-induced thrombocytopenia with thrombosis (HITT).

HIT pathogenesis involves the formation of multi-molecular complexes of the platelet alpha-granule PF4, heparin and anti-heparin antibodies. (21) PF4 is released by platelets when they are activated by agonists including thrombin, collagen, and heparin. HIT usually involves production of IgG anti-heparin antibodies, termed HIT IgGs, but IgM and IgA anti-heparin antibodies have also been implicated. (22,23) PF4:heparin:IgG-anti-heparin immune complexes interact with platelet FcγII receptors and thereby induce potent platelet activation, platelet aggregation, and a marked increase in thrombin levels. (22,24) The process can escalate as antibodies bind complexes on neighboring platelets, and consequently induce further PF4 release, platelet activation and antibody production. (25) In the presence of heparin, HIT sera have also been shown to generate pro-coagulant platelet microparticles of undetermined function that appear to emanate from activated platelet pseudopodia. (26,27)

HIT-IgG has been shown to activate endothelium in vitro, by interacting with heparan: PF4 complexes on the endothelial cell surface. (22) Heparan is an endogenous glycosaminoglycan thought to serve as an anticoagulant on vascular endothelium. This endothelial cell activation could augment tissue factor synthesis, and thereby further escalate excess thrombin generation, thrombus formation, and platelet activation. Furthermore endothelial cell hyperplasia caused by immune-mediated injury may also contribute to HIT-associated small vessel thrombosis and occlusive disease. (28)

Clinical Manifestations

The thrombocytopenia of HIT has a characteristic timing and severity as described below.

Timing

Figure 1 provides a summary of the temporal patterns of thrombocytopenia in HIT patients. The hallmark sign of HIT is a plunge in platelet count that becomes evident within 5-10 days of commencing heparin treatment. (2) In one study it was observed that none of 189 patients who had early thrombocytopenia (within 4 days of starting heparin) had HIT, while 9 of 11 patients who developed late thrombocytopenia (after 5 or more days) were serologically diagnosed with HIT. Thus late thrombocytopenia onset is a critical clinical harbinger of HIT. Rapid-onset thrombocytopenia can develop into HIT in rare cases. However, this generally occurs only in patients who received heparin within the previous few months, and who therefore had circulating HIT antibodies already present when heparin was reintroduced. (29,30) Recent studies indicate that an anamnestic HIT antibody response is unlikely. (31)

Delayed-onset HIT following heparin withdrawal has been described. (32,33) Delayed-onset HIT was reported in 12 patients after an average of 9 days (range: 5 to 19 days) post-heparin withdrawal. (34) These patients had high titer platelet-activating antibodies and exhibited elevated heparin-dependent and heparin-independent platelet activation. In a large retrospective study involving 260 patients from three hospitals, there were 14 cases of delayed-onset HIT (median onset 14 days, range: 9 to 40 days). (33)

Severity

Thrombocytopenia (a platelet count fall of ≥50%), the most common clinical manifestation of HIT, occurs in ≥95% of HIT patients. (3,5,34) However, thrombocytopenia is rarely severe (median nadir platelet count = 50-60x10^9/L) and is rarely associated with bleeding. (35) Many patients with confirmed HIT go on to develop concomitant thrombosis (HITT), even in cases in which the heparin treatment was used only for antithrombotic prophylaxis. (36,37)

In a large clinical trial (38), 8 of 9 post-operative orthopedic patients with HIT developed thrombosis, compared with only 117 of 656 patients without HIT (odds ratio, 37; 95% CI, 4.8-1638; $p < 0.001$). A retrospective review found that venous and arterial thrombosis occurred in 61% (78/127) and 14% (18/127) of serologically confirmed HIT patients. (37) Approximately half of these patients presented with a complicating thrombotic event before being diagnosed with HIT. And 53% of patients diagnosed with HIT in the absence of thrombosis went on to develop thrombosis within 30 days. Many prospective and case-controlled studies (5,29,39-41) have suggested that HIT patients have a 35-75% chance of developing thrombosis.

Venous thromboembolism, such as deep vein thrombosis (DVT) and pulmonary embolism are the most common thrombotic events observed, especially in post-operative patients. Rare clinical events such as gradually progressive neurologic dysfunction can sometimes be attributed to venous thrombosis (i.e. cerebral venous thrombosis). (36) Arterial thrombotic events associated with HIT most often manifest as acute ischemia of the leg due to occlusion of the distal aorta or a large limb artery. (36) Microvascular thrombosis in the presence of DVT has also been reported to produce limb ischemia in HIT patients undergoing warfarin anticoagulation therapy who develop severe protein C depletion. (42) Arterial thrombotic complications of HIT can also result in stroke and myocardial infarction.

In patients with HIT, DVT occurs more commonly in the legs than in the arms. (43) In a retrospective study of 260 antibody-positive HIT patients, only 14 episodes of upper extremity DVT were observed (5.4% of the cases). In all 14 cases, the DVT occurred at the site of a central venous catheter. About 10 to 20% of patients who develop HIT while receiving subcutaneous injections of heparin develop injection-site skin lesions, ranging from painful erythematous plaques to skin necrosis. (36,44) Some patients develop an acute systemic reaction within 5 to 30 min of receiving an intra-venous heparin bolus. (36,45,46) A minority of HIT patients suffer from additional complications including disseminated intravascular coagulation, (3,36) adrenal hemorrhage and transient global amnesia. (47,30)

Diagnosis

Clinical diagnosis

The HIT diagnostic assays with the greatest sensitivity and specificity are costly and time consuming, and are not available at many institutions. (47,48) Thus a prudent awareness of the clinical signs of HIT is crucial so that its diagnosis can be promptly considered in cases of unexplained thrombocytopenia and/or thrombosis during or following heparin exposure. Clinical scoring rubrics have been developed to discriminate between patients that are unlikely, likely, or highly likely to have HIT. (3) The scoring is based on multiple parameters including absolute platelet count, rate of platelet decrease, platelet recovery after heparin withdrawal, presence of cutaneous reactions, and occurrence of thrombosis.

Laboratory assay diagnosis

HIT diagnosis can be confirmed by serotonin release assay (SRA), heparin-induced platelet aggregation assay (HIPA), or solid phase immunoassay. (49,50) The platelet SRA, which is the detection of radiolabeled serotonin release from washed platelets, (51) is considered the gold standard assay, both in term of sensitivity and specificity, which are in the range of 65% to 90%. Consequently, the SRA is often used as the reference standard in the testing of new assays. (29,52-54) SRA has the practical drawback of being quite technically demanding. (55) In contrast to the SRA, the solid phase ELISA immunoassay is not a functional assay. For the solid phase immunoassay, patient serum samples are simply added to microtiter plates that have been pre-coated with heparin-PF4 complexes. If heparin antibodies are present in the serum, antibody-bound complexes are detected by incubation with a specific secondary antibody. (56) Although this is a very sensitive assay, detecting 91 to >97% of HIT cases, many patients that will not develop HIT also have a positive result (low specificity: 74 to 86%). (57,58) Thus the solid phase immunoassay has a favorable negative predictive value, >95%, but only a moderate positive predictive value of 50 to 93%, depending on the study population and timing of thrombocytopenia onset. (2,58-60) As 10-20% of immunoassay results may be inconsistent with findings of other assays, it should be used in combination with another assay. (2) The sensitivity and specificity of some immunological assays are summarized in **Table 3.**

Treatment

HIT differs from most other acquired hypercoagulability syndromes (61) in that it is transient: platelet count deficits generally resolve within days or weeks, and the pathogenic HIT antibodies are no longer detectable within weeks to a few months. (62) Upon diagnosis of HIT, patients should be immediately withdrawn from exposure to heparin, including heparin-bound catheters and heparin flushes. (2,63) LMWH should not be considered safe in patients that developed HIT as a result of UFH exposure since LMWH may cross-react with UFH-induced antibodies (2,29) and induce further heparin-dependent IgG antibody formation. Given that HIT patients develop thrombotic complications within 30 days in as many as 53% of cases, (37) further interventions beyond heparin withdrawal, such as administration of agents that reduce thrombin generation (e.g. danaproid) or inhibit thrombin activity (e.g. lepirudin), (64-66) should be initiated to prevent severe complications.

Lepirudin

The FDA has approved lepirudin, a recombinant hirudin, for treatment of HIT associated thrombosis. This agent can also prevent the de novo development of thromboses in patients with isolated HIT. (67) Two prospective, multi-center, historically controlled trials (HAT-1 and HAT-2) evaluated the safety and efficacy of lepirudin in HIT patients. (68,69) A third prospective multi-center study (HAT-3) evaluated the safety and efficacy of lepirudin in patients confirmed to have HIT by the very sensitive functional heparin-induced platelet activation (HIPA) test. (70) The HAT studies demonstrated that lepirudin treatment significantly reduced the combined incidence of amputations, thromboses and death. When incidences of these clinical complications were considered separately, only the incidence of new thrombotic events was significantly reduced in the lepirudin treated groups. It is worth noting that about one-third of the clinical events in

these trials occurred during the pre-treatment delay. (70,71) This underscores the importance of prompt commencement of HIT treatment. Indeed, patients presenting with clinical signs of HIT should be withdrawn immediately from heparin administration and then given an alternative anticoagulant without waiting for laboratory results.

Argatroban

Argatroban is a non-immunogenic small-molecule direct thrombin inhibitor approved to treat both isolated HIT and HIT-associated thrombosis. The usual argatroban dose is 2 μg/kg/min adjusted by aPTT (usual target 1.5 to 3 times baseline). Argatroban has been shown to reduce the risk of thrombosis. (2,72) Because argatroban metabolism involves hepatobiliary excretion, patients with liver dysfunction should be administered argatroban at no more than 25% of the normal dose. Because argatroban is associated with a relatively prolonged elevated clotting tendency, as reflected by the international normalized ratio (INR) index, (73) warfarin treatment should be postponed in patients receiving argatroban in order to prevent microvascular thrombosis. (74)

Danaproid

Danaproid is a heparinoid which consists predominantly of dermatan sulfate and low-sulfated heparan sulfate. Experimental danaproid treatment was associated with a favorable outcome in 93% (215/230) of HIT patients. (75) Complications among patients who did not have a favorable outcome included bleeding (n =2), recurrent thrombocytopenia (n =4), persistent thrombocytopenia (n =5), and thromboembolic events (n =4). Overall, five patients demonstrated danaproid cross-reactivity with HIT-IgGs (one of these patients developed a new thrombosis). Mortality possibly or probably due to danaproid treatment was reported in 3% of the patients (n =7); complications in these cases included bleeding, thrombosis, and septic shock. (75)

Fondaparinux

Fondaparinux is a synthetic highly sulfated pentasaccharide whose sequence is derived from the minimal antithrombin binding region of heparin. (2,76) Similarly to danaproid, fondaparinux has anti-factor Xa activity with a long half-life and no or negligible in vitro cross-reactivity with HIT antibodies. Thus fondaparinux is a potential agent for HIT treatment, but there is not yet sufficient evidence upon which to judge its safety and efficacy.

Warfarin

Warfarin is a naturally derived anticoagulant that is generally used prophylactically to prevent the occurrence or reoccurrence of thrombosis and embolism. Because of warfarin's inhibitory action on regulatory factors, (42) its initial effect can paradoxically be clot promoting. It should therefore only be administered after a patient has been stably anticoagulated with a thrombin-specific inhibitor and the patient's platelet count has recovered to at least $100x10^9$/L. (2) There should be at least 5 days of overlapping therapy before the thrombin-specific inhibitor is discontinued; and the initial doses of warfarin should be modest (<10 mg/d). The target range for anticoagulation with warfarin should be an INR in the range of 2.0 to 3.0. Given the high risk of thrombosis within the first month after HIT diagnosis, (41) warfarin anticoagulation treatment should be continued for at least several months. (77)

Prevention

In light of the complexity of HIT diagnosis and treatment, preventive measures should be taken. Platelet count should be monitored relative to a baseline value at least every third day between days 5-14 of heparin exposure. Prophylactic oral warfarin in patients prescribed heparin therapy for acute thrombosis or atrial fibrillation should be used to minimize the duration of heparin exposure, which preferably should be no more than 5 days. It is vital that occurrences of HIT be properly documented to prevent risky heparin re-exposures. Patients with a history of HIT should not be re-exposed to heparin for at least 3 months, and subsequent exposures should be of minimal duration or preferably avoided altogether.

LMWH is less likely than UFH to produce HIT. (78) In a prospective study of postoperative orthopedic patients, heparin-dependent IgG antibodies were found less often in patients given LMWH (2.2%) than in patients given UFH (7.8%). Moreover, while 2.7% of patients given UFH developed HIT, no cases were reported in the LMHW group. (33) Furthermore, among 499 patients given LMHW who underwent hip replacement surgeries at multiple institutions, only one developed HIT. (79)

In summary, HIT may be prevented or reduced by minimizing heparin re-exposure and utilization of LMWH rather than UFH. If clinical signs of HIT are present, heparin exposure should be halted immediately and an alternative anticoagulant treatment course should be initiated. The currently available data suggest that the direct thrombin inhibitors lepirudin and argatroban, when used with caution, are the best available treatment options for HIT (41).

Table 1. Main Features of the Two Different Forms of Heparin-Induced Thrombocytopenia

Feature	Type I	Type II
Frequency	Up to 20%	Up to 5%
Onset	1-4 d	5-14 d
Pathogenesis	Heparin-induced aggregation	Antibody to PF4-heparin complex
Thrombosis	No	Yes
Management	No heparin withdrawal required	Heparin withdrawal mandatory-alternative anticoagulation

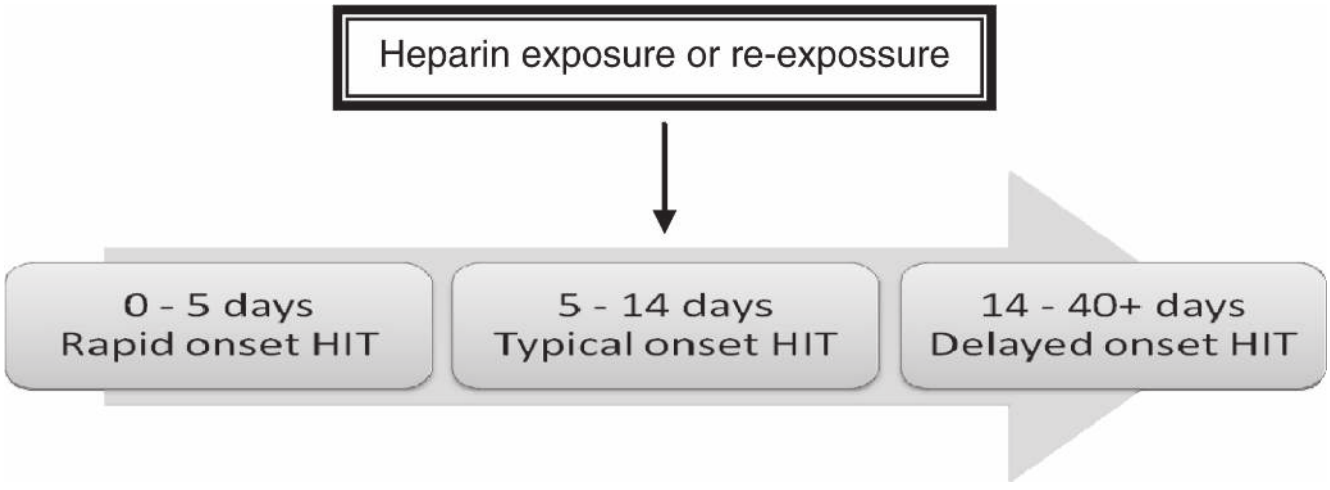
Table 2. Summary of Studies Examining the Incidence of Immune HIT Type Il in Different Patient Populations, Receiving UFH or LMWH

Category	Incidence (%)	References
UFH		
After orthopedic surgery	2-6.5	(5,8,12)
After heart surgery	1	(11)
After general or vascular surgery	0	(13)
Medical patients	0.3-0.9	(14-17)
LMWH		
After orthopedic surgery	0-0.6	(5,8,12,79)
After general or vascular surgery	0	(13)
Medical patients	0.5-0.8	(16,18)

Table 3. Sensitivity and Specificity of Some Immunological Assays Used for the Diagnosis of HIT Syndrome

Diagnostic assay	Sensitivity (%)	Specificity (%)	
		Early platelet decrease	Late platelet decrease
Serotonin release assay (SRA)	90-98	>95	80-97
Heparin-induced platelet activation assay (HIPA)	90-98	>95	80-97
PF4/heparin EIA	>90	>95	50-93
Combination of platelet activation assay & PF4 antigen EIA	100	>95	80-97

Figure 1. Temporal Patterns of Thrombocytopenia in HIT



References

1. Chong BH, Castaldi PA. Platelet proaggregating effect of heparin: possible mechanism for non-immune heparin-associated thrombocytopenia. Aust N Z J Med 1986;16:715-6.
2. Warkentin TE, Greinacher A. Heparin-induced thrombocytopenia: recognition, treatment, and prevention: the Seventh ACCP Conference on Antithrombotic and Thrombolytic Therapy. Chest 2004;126:311S-37S.
3. Warkentin TE. Heparin-induced thrombocytopenia: pathogenesis and management. Br J Haematol 2003;121:535-55.
4. Stanton PE Jr, Evans JR, Lefemine AA, Vo NM, Rannick GA, Morgan CV Jr, et al. White clot syndrome. South Med J 1988;81:616-20.
5. Warkentin TE, Roberts RS, Hirsh J, Kelton JG. An improved definition of immune heparin-induced thrombocytopenia in postoperative orthopedic patients. Arch Intern Med 2003;163:2518-24.
6. Sheridan D, Carter C, Kelton JG. A diagnostic test for heparin-induced thrombocytopenia. Blood 1986;67:27-30.
7. Chong BH. Heparin induced thrombocytopenia. Aust NZJ Med 1992;22:145-52.
8. Ganzer D, Gutezeit A, Mayer G, Greinacher A, Eichler P. Prevention of thromboembolism as a cause of thromboembolic complications. A study of the incidence of heparin-induced thrombocytopenia type II. Z Orthop Ihre Grenzgeb 1997;135:543-9.
9. Heeger, PS, Backstrom, JT. Heparin flushes and thrombocytopenia. Ann Intern Med 1986;105:143.
10. Laster J, Silver D. Heparin-coated catheters and heparin-induced thrombocytopenia. J Vasc Surg 1988;7:667-72.
11. Warkentin TE, Sheppard JA, Horsewood P, Simpson PJ, Moore JC, Kelton JG. Impact of the patient population on the risk for heparin-induced thrombocytopenia. Blood 2000;96:1703-8.
12. Mahlfeld K, Franke J, Schaeper O, Kayser R, Grasshoff H. Heparin-induced thrombocytopenia as a complication of postoperative prevention of thromboembolism with unfractionated heparin/low molecular weight heparin after hip and knee prosthesis implantation. Unfallchirurg 2002;105:327-31.
13. Lindhoff-Last E, Eichler P, Stein M, Plagemann J, Gerdsen F, Wagner R, et al. A prospective study on the incidence and clinical relevance of heparin-induced antibodies in patients after vascular surgery. Thromb Res 2000;97:387-93.
14. Kappers-Klunne MC, Boon DM, Hop WC, Michiels JJ, Stibbe J, van der Zwaan C, et al. Heparin-induced thrombocytopenia and thrombosis: a prospective analysis of the incidence in patients with heart and cerebrovascular diseases. Br J Haematol 1997;96:442-6.
15. Lozano M, Perez S, Cid J, et al. Is the incidence of heparin induced thrombocytopenia the same everywhere? The case of Spain. Thromb Haemost 2001(suppl); XVIII Congress ISTH (abst P2718).
16. Lindhoff-Last E, Nakov R, Misselwitz F, Breddin HK, Bauersachs R. Incidence and clinical relevance of heparin-induced antibodies in patients with deep vein thrombosis treated with unfractionated or low-molecular-weight heparin. Br J Haematol 2002;118:1137-42.
17. Girolami B, Prandoni P, Stefani PM, Tanduo C, Sabbion P, Eichler P, et al. The incidence of heparin-induced thrombocytopenia in hospitalized medical patients treated with subcutaneous unfractionated heparin: a prospective cohort study. Blood 2003;101:2955-9.
18. Prandoni P, Siragusa S, Girolami B, Fabris F; BELZONI Investigators Group. The incidence of heparin-induced thrombocytopenia in medical patients treated with low-molecular-weight heparin: a prospective cohort study. Blood 2005;106:3049-54.
19. Martel N, Lee J, Wells PS. Risk for heparin-induced thrombocytopenia with unfractionated and low-molecular-weight heparin thromboprophylaxis: a meta-analysis. Blood 2005;106:2710-5.
20. Warkentin TE, Sheppard JA, Sigouin CS, Kohlmann T, Eichler P, Greinacher A. Gender imbalance and risk factor interactions in heparin-induced thrombocytopenia. Blood 2006;108:2937-41.
21. Greinacher A. Antigen generation in heparin-associated thrombocytopenia: the nonimmunologic type and the immunologic type are closely linked in their pathogenesis. Semin Thromb Hemost 1995;21:106-16.
22. Visentin GP, 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. J Clin Invest 1994;93:81-8.
23. Amiral J, Wolf M, Fischer A, Boyer-Neumann C, Vissac A, Meyer D. Pathogenicity of IgA and/or IgM antibodies to heparin-PF4 complexes in patients with heparin-induced thrombocytopenia. Br J Haematol 1996;92:954-9.
24. Greinacher A, Pötzsch B, Amiral J, Dummel V, Eichner A, Mueller-Eckhardt C. Heparin-associated thrombocytopenia: isolation of the antibody and characterization of a multimolecular PF4-heparin complex as the major antigen. Thromb Haemost 1994;71:247-51.
25. Newman PM, Chong BH. Heparin-induced thrombocytopenia: new evidence for the dynamic binding of purified anti-PF4-heparin antibodies to platelets and the resultant platelet activation. Blood 2000;96:182-7.
26. Warkentin TE, Hayward CP, Boshkov LK, Santos AV, Sheppard JA, Bode AP, et al. Sera from patients with heparin-induced thrombocytopenia generate platelet-derived microparticles with procoagulant activity: an explanation for the thrombotic complications of heparin-induced thrombocytopenia. Blood 1994;84:3691-9.
27. Hughes M, Hayward CP, Warkentin TE, Horsewood P, Chorneyko KA, Kelton JG. Morphological analysis of microparticle generation in heparin-induced thrombocytopenia. Blood 2000;96:188-94.
28. Kwaan HC, Sakurai S. Endothelial cell hyperplasia contributes to thrombosis in heparin-induced thrombocytopenia. Semin Thromb Hemost 1999;25:23-7.
29. Warkentin TE, Levine MN, Hirsh J, Horsewood P, Roberts RS, Gent M, et al. Heparin-induced thrombocytopenia in patients treated with low-molecular-weight heparin or unfractionated heparin. N Engl J Med 1995;332:1330-5.
30. Warkentin TE, Hirte HW, Anderson DR, Wilson WE, O'Connell GJ, Lo RC. Transient global amnesia associated with acute heparin-induced thrombocytopenia. Am. J. Med. 1994;97:489-91.
31. Ling E, Warkentin TE. Intraoperative heparin flushes and subsequent acute heparin-induced thrombocytopenia. Anesthesiology 1998;89:1567-9.
32. Warkentin TE, Kelton JG. Timing of heparin-induced thrombocytopenia (HIT) in relation to previous heparin use: absence of an anamnestic immune response, and implications for repeat heparin use in patients with a history of HIT. Blood 1998;92:182a.
33. Warkentin, TE, Kelton, JG. Delayed-onset heparin-induced thrombocytopenia and thrombosis. Ann Intern Med 2001;135:502-6.

34.Rice L, Attisha WK, Drexler A, Francis JL. Delayed-onset heparin-induced thrombocytopenia. Ann Intern Med 2002;136:210-5.
35.Warkentin TE, Kelton JG. Interaction of heparin with platelets, including heparin-induced thrombocytopenia. In: Bounameaux H, editor. Low-Molecular Weight Heparins in Prophylaxis and Therapy of Thromboembolic Diseases. Fundamental and Clinical Cardiology. New York: Marcel Dekker; 1994. p. 75.
36.Warkentin TE. Clinical picture of heparin-induced thrombocytopenia. In: Warkentin TE, Greinacher A, editors. Heparin induced thrombocytopenia. 3rd ed. New York (NY): Marcel Dekker; 2004. p. 53-106.
37.Warkentin TE, Kelton JG. A 14-year study of heparin-induced thrombocytopenia. Am J Med 1996;101:502-7.
38.Wallis DE, Workman DL, Lewis BE, Steen L, Pifarre R, Moran JF. Failure of early heparin cessation as treatment for heparin-induced thrombocytopenia. Am J Med 1999;106:629-35.
39.Hong AP, Cook DJ, Sigouin CS, Warkentin TE. Central venous catheters and upper-extremity deep-vein thrombosis complicating immune heparin-induced thrombocytopenia. Blood 2003;101:3049-51.
40.Girolami B, Prandoni P, Stefani PM, Tanduo C, Sabbion P, Eichler P, et al. The incidence of heparin-induced thrombocytopenia in hospitalized medical patients treated with subcutaneous unfractionated heparin: a prospective cohort study. Blood 2003;101:2955-9.
41.Warkentin TE. Management of heparin-induced thrombocytopenia: a critical comparison of lepirudin and argatroban. Thromb Res 2003;110:73-82.
42.Warkentin TE, Elavathil LJ, Hayward CP, Johnston MA, Russett JI, Kelton JG. The pathogenesis of venous limb gangrene associated with heparin-induced thrombocytopenia. Ann Intern Med 1997;127: 804-12.
43.Hong AP, Cook DJ, Sigouin CS, Warkentin TE. Central venous catheters and upper-extremity deep-vein thrombosis complicating immune heparin-induced thrombocytopenia. Blood 2003;101:3049-51.
44.Warkentin TE. Heparin-induced skin lesions. Br J Haematol 1996;92:494-7.
45.Ansell JE, Clark WP Jr, Compton CC. Fatal reactions associated with intravenous heparin. Drug Intell Clin Pharm 1986;20:74-5.
46.Popov D, Zarrabi MH, Foda H, Graber M. Pseudopulmonary embolism: acute respiratory distress in the syndrome of heparin-induced thrombocytopenia. Am J Kidney Dis 1997;29:449-52.
47.Arthur CK, Grant SJ, Murray WK, Isbister JP, Stiel JN, Lauer CS. Heparin-associated acute adrenal insufficiency. Aust N Z J Med 1985;15:454-5.
48.Sheridan D, Carter C, Kelton JG. A diagnostic test for heparin-induced thrombocytopenia. Blood 1986;67:27-30.
49.Greinacher A, Amiral J, Dummel V, Vissac A, Kiefel V, Mueller-Eckhardt C. Laboratory diagnosis of heparin-associated thrombocytopenia and comparison of platelet aggregation test, heparin-induced platelet activation test, and platelet factor 4/heparin enzyme-linked immunosorbent assay. Transfusion 1994;34:381-5.
50.Kelton JG, Warkentin TE. Diagnosis of heparin-induced thrombocytopenia. Still a journey, not yet a destination. Am J Clin Pathol 1995;104:611-3.
51.Leo A , Winteroll S. Laboratory diagnosis of heparin-induced thrombocytopenia and monitoring of alternative anticoagulants. Clin Diagn Lab Immunol 2003;10:731-40.
52.Chong BH, Burgess J, Ismail F. The clinical usefulness of the platelet aggregation test for the diagnosis of heparin-induced thrombocytopenia. Thromb Haemost 1993;69:344-50.
53.Walenga JM, Jeske WP, Wood JJ, Ahmad S, Lewis BE, Bakhos M. Laboratory tests for heparin-induced thrombocytopenia: a multicenter study. Semin Hematol 1999;36:22-8.
54.Warkentin TE, Hayward CP, Smith CA, Kelly PM, Kelton JG. Determinants of donor platelet variability when testing for heparin-induced thrombocytopenia. J Lab Clin Med 1992;120:371-9.
55.Eichler P, Budde U, Haas S, Kroll H, Loreth RM, Meyer O, et al. First workshop for detection of heparin-induced antibodies: validation of the heparin-induced platelet-activation test (HIPA) in comparison with a PF4/heparin ELISA. Thromb Haemost 1999;81:625-9.
56.Martinuzzo ME, Forastiero RR, Adamczuk Y, Pombo G, Carreras LO. Antiplatelet factor 4--heparin antibodies in patients with antiphospholipid antibodies. Thromb Res 1999;95:271-9.
57.de Larrañaga G, Martinuzzo M, Bocassi A, Fressart MM, Forastiero R. Heparin-platelet factor 4 induced antibodies in patients with either autoimmune or alloimmune antiphospholipid antibodies. Thromb Haemost 2002;88:371-3.
58.Arepally GM, Ortel TL. Clinical practice. Heparin-induced thrombocytopenia. N Engl J Med 2006;355:809-17.
59.Pouplard C, Amiral J, Borg JY, Laporte-Simitsidis S, Delahousse B, Gruel Y. Decision analysis for use of platelet aggregation test, carbon 14-serotonin release assay, and heparin-platelet factor 4 enzyme-linked immunosorbent assay for diagnosis of heparin-induced thrombocytopenia. Am J Clin Pathol 1999;111:700-6.
60.Verma AK, Levine M, Shalansky SJ, Carter CJ, Kelton JG. Frequency of heparin-induced thrombocytopenia in critical care patients. Pharmacotherapy 2003;23:745-53.
61.Warkentin TE. Platelet count monitoring and laboratory testing for heparin-induced thrombocytopenia. Arch Pathol Lab Med 2002;126:1415-23.
62.Warkentin TE, Kelton JG. Temporal aspects of heparin-induced thrombocytopenia. N Engl J Med 2001;344:1286-92.
63.Napolitano LM, Warkentin TE, Almahameed A, Nasraway SA. Heparin-induced thrombocytopenia in the critical care setting: diagnosis and management. Crit Care Med 2006;34:2898-911.
64.Warkentin TE. Heparin-induced thrombocytopenia: yet another treatment paradox?. Thromb Haemost 2001;85:947-9.
65.Warkentin TE. Limitations of conventional treatment options for heparin-induced thrombocytopenia. Semin Hematol 1998;35:17-25.
66.Warkentin TE, Chong BH, Greinacher A. Heparin-induced thrombocytopenia: towards consensus. Thromb Haemost 1998;79:1-7.
67.Lubenow N, Eichler P, Lietz T, Farner B, Greinacher A. Lepirudin for prophylaxis of thrombosis in patients with acute isolated heparin-induced thrombocytopenia: an analysis of 3 prospective studies. Blood 2004;104:3072-7.
68.Greinacher A, Völpel H, Janssens U, Hach-Wunderle V, Kemkes-Matthes B, Eichler P, et al. Recombinant hirudin (lepirudin) provides safe and effective anticoagulation in patients with heparin-induced thrombocytopenia: a prospective study. Circulation 1999;99:73-80.
69.Greinacher A, Janssens U, Berg G, Böck M, Kwasny H, Kemkes-Matthes B, et al. Lepirudin (recombinant hirudin) for parenteral anticoagulation in patients with heparin-induced thrombocytopenia. Heparin-Associated Thrombocytopenia Study (HAT) investigators. Circulation 1999;100:587-93.
70.Lubenow N, Eichler P, Lietz T, Greinacher A; Hit Investigators Group. Lepirudin in patients with heparin-induced thrombocytopenia - results of the third prospective study (HAT-3) and a combined analysis of HAT-1, HAT-2, and HAT-3. J Thromb Haemost 2005;3:2428-36.
71.Greinacher A, Eichler P, Lubenow N,

Kwasny H, Luz M. Heparin-induced thrombocytopenia with thromboembolic complications: meta-analysis of 2 prospective trials to assess the value of parenteral treatment with lepirudin and its therapeutic aPTT range. Blood 2000;96:846-51.
72. Lewis BE, Wallis DE, Leya F, Hursting MJ, Kelton JG; Argatroban-915 Investigators. Argatroban anticoagulation in patients with heparin-induced thrombocytopenia. Arch Intern Med 2003;163:1849-56.
73. Warkentin TE. Bivalent direct thrombin inhibitors: hirudin and bivalirudin. Best Pract Res Clin Haematol 2004;17:105-25.
74. Smythe MA, Warkentin TE, Stephens JL, Zakalik D, Mattson JC. Venous limb gangrene during overlapping therapy with warfarin and a direct thrombin inhibitor for immune heparin-induced thrombocytopenia. Am J Hematol 2002;71:50-2.
75. Magnani HN. Heparin-induced thrombocytopenia (HIT): an overview of 230 patients treated with orgaran (Org 10172)). Thromb Haemost 1993;70: 554-61.
76. Chong BH, Magnani HN. Danaparoid for the treatment of heparin-induced thrombocytopenia. In: Warkentin TE, Greinacher A, editors. Heparin-induced thrombocytopenia. 3rd ed. New York (NY): Marcel Dekker; 2004. p 371-96.
77. Watson HG, Keeling DM; BCSH Taskforce in Haemostasis and Thrombosis. The management of heparin-induced thrombocytopenia. Br J Haematol 2006;135:269.
78. Gruel Y, Pouplard C, Nguyen P, Borg JY, Derlon A, Juhan-Vague I, et al. Biological and clinical features of low-molecular-weight heparin-induced thrombocytopenia. Br J Haematol 2003;121:786-92.
79. Planès A, Samama MM, Lensing AW, Büller HR, Barre J, Vochelle N, et al. Prevention of deep vein thrombosis after hip replacement--comparison between two low-molecular heparins, tinzaparin and enoxaparin. Thromb Haemost 1999;81:22-5.