

Delayed-Onset Heparin-Induced Thrombocytopenia and Thrombosis

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Background: Heparin-induced thrombocytopenia is a prothrombotic drug reaction caused by platelet-activating antibodies that recognize complexes of platelet factor 4 and heparin.

Objective: To describe a syndrome termed *delayed-onset heparin-induced thrombocytopenia*, in which thrombocytopenia and thrombotic events begin 5 or more days after withdrawal of heparin.

Design: Case series.

Setting: Secondary and tertiary care hospitals.

Patients: 12 patients who presented with serologically confirmed, delayed-onset heparin-induced thrombocytopenia, including 6 outpatients presenting after hospital discharge.

Measurements: The platelet serotonin-release assay was used to measure IgG-induced heparin-dependent and heparin-independent platelet activation; an enzyme immunoassay that detects IgG against platelet factor 4–heparin complexes was also used.

Results: Patients with delayed-onset heparin-induced thrombocytopenia presented with thrombocytopenia and associated thrombosis a mean of 9.2 days (range, 5 to 19 days) after stopping heparin therapy. Nine patients received additional heparin, with further decrease in platelet counts. Compared with controls, patients with delayed-onset heparin-induced thrombocytopenia had higher titers of IgG antibodies to platelet factor 4–heparin and greater IgG-induced heparin-dependent and heparin-independent platelet activation.

Conclusions: Delayed-onset heparin-induced thrombocytopenia should be suspected when patients present with thrombocytopenia and thrombosis up to 3 weeks after exposure to heparin. This syndrome could be caused by high titers of platelet-activating IgG induced by heparin.

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Heparin-induced thrombocytopenia is an immunologic drug reaction characterized by paradoxical association with venous and arterial thrombosis (1, 2). The syndrome is usually caused by IgG antibodies that are reactive against complexes of platelet factor 4 and heparin (3). Patients typically develop thrombocytopenia while receiving heparin; the peak onset is 5 to 8 days after starting heparin therapy (2). In this report, we describe 12 patients who experienced thrombocytopenia or symptoms of thrombosis an average of 9 days after all heparin was withdrawn. We refer to this phenomenon as *delayed-onset heparin-induced thrombocytopenia*.

REPRESENTATIVE CASE REPORT

Patient 1

A 68-year-old woman underwent laparoscopic cholecystectomy. She received one preoperative and two postoperative subcutaneous 5000-U injections of unfractionated heparin before discharge on postoperative day 1. Eight days later, she returned to the hospital because of sudden onset of severe anterograde and retrograde amnesia; her platelet count was 50×10^9 cells/L. The anterograde amnesia cleared within 6 hours, consistent with transient global amnesia. Although no heparin was given, the platelet count decreased further to 14×10^9 cells/L (nadir); the patient developed isch-

emia of the left great toe and disseminated intravascular coagulation. The fibrinogen level was 1.1 g/L (normal, 1.5 to 4.5 g/L), the D-dimer level exceeded 3000 $\mu\text{g/L}$ (normal, $<500 \mu\text{g/L}$), and thrombin–antithrombin complexes were greater than 60 $\mu\text{g/L}$ (normal, $<4 \mu\text{g/L}$). Heparin-induced thrombocytopenia was diagnosed, and an 11-day course of danaparoid was associated with resolution of the ischemic symptoms and signs. Twenty-four days after withdrawal of danaparoid, and during persisting thrombocytopenia (platelet count, 94×10^9 cells/L), the patient developed proximal deep venous thrombosis. She received a 40-day course of danaparoid, with clinical resolution. However, 100 days after stopping the second course of danaparoid, and during persisting thrombocytopenia (platelet count, 109×10^9 cells/L), symptomatic deep venous thrombosis recurred. The patient received additional danaparoid for 8 days, with overlapping warfarin for 6 months. Six months after heparin-induced thrombocytopenia began, the platelet count remained above 150×10^9 cells/L. The patient was well at 1-year follow-up.

Results of laboratory tests were strongly positive for heparin-dependent antibodies. The patient's serum specimen caused 94% serotonin release at 0 U of heparin per mL, 100% serotonin release at 0.2 U of heparin per mL, and 0% serotonin release at 100 U of heparin

per mL. In addition, platelet factor 4–heparin IgG antibodies were detected by enzyme immunoassay (1.95 optical density units [positive result, >0.45 optical density units]). Antibodies could be detected in blood specimens obtained up to 9 months after the onset of heparin-induced thrombocytopenia; subsequent specimens yielded negative results.

Serology for antiphospholipid antibodies (nonspecific inhibitor and anticardiolipin antibodies), as well as levels

of antithrombin III, protein C, and protein S (measured after discontinuation of warfarin therapy), had normal results. Factor V Leiden mutation was not present.

METHODS

Patients

Our study included patients who had thrombocytopenia and thrombotic events 5 or more days after with-

Table. Twelve Patients with Heparin-Induced Thrombocytopenia and Thrombosis Beginning after Discontinuation of Heparin Therapy

| Patient | Sex | Age | Inpatient or Outpatient | Heparin Use* | Platelet Count | | Time from Last Heparin Use to Onset of Thrombocytopenia† | Further Heparin Given | Complications of Heparin-Induced Thrombocytopenia | Day of Completion* | Clinical Course after Diagnosis of Heparin-Induced Thrombocytopenia |
|---------|-----|-----|-------------------------|--------------|-----------------------|-------|--|-----------------------|--|--------------------|---|
| | | | | | Initial Presentation | Nadir | | | | | |
| | | y | | d | $\times 10^9$ cells/L | | d | | | | |
| 1 | F | 68 | Outpatient | 1 | 50 | 14 | 8 | No | Transient global amnesia Hypofibrinogenemia Right hallux ischemia Proximal deep venous thrombosis | 9 9 11 40 | Recovery during danaparoid therapy; subsequent relapses requiring further danaparoid and warfarin (see text) |
| 2 | M | 67 | Outpatient | 6 | 30 | 18‡ | 11 | Yes | Proximal deep venous thrombosis Hypofibrinogenemia Cardiac arrest after 10 000-U intravenous heparin bolus | 18 18 18 | Limb gangrene and fatal intracranial hemorrhage with anicrod and warfarin |
| 3 | F | 70 | Outpatient | 7 | 36 | 23‡ | 6 | Yes | Aorto–bifemoral graft occlusion | 13 | Progressive bilateral foot ischemia during anicrod therapy, with recovery after substituting danaparoid for anicrod |
| 4 | M | 69 | Outpatient | <1 | 17 | 17 | 8 | No | Bilateral lower limb arterial thrombosis Hypofibrinogenemia | 8 8 | Bilateral limb gangrene, fatal myocardial infarction during treatment with anicrod |
| 5 | F | 75 | Outpatient | 6 | 52 | 38‡ | 8 | Yes | Proximal deep venous thrombosis | 14 | Recovery with vena cava filter and warfarin |
| 6 | F | 80 | Outpatient | 2 | 135 | 59‡ | 19 | Yes | Pulmonary embolism | 21 | Recovery with argatroban and warfarin |
| 7 | M | 61 | Inpatient | 2 | 130 | 65 | 6 | No | Adrenal hemorrhagic infarction | 8 | Recovery with continuing warfarin |
| 8 | F | 65 | Inpatient | 3§ | 50 | 19‡ | 12 | Yes | Proximal deep venous thrombosis | 15 | Recovery with anicrod and warfarin |
| 9 | M | 73 | Inpatient | 2§ | 95 | 10‡ | 5 | Yes | Pulmonary embolism | 7 | Recovery with danaparoid and warfarin |
| 10 | M | 72 | Inpatient | 6 | 150 | 73‡ | 8 | Yes | Bilateral proximal deep venous thrombosis Pulmonary embolism | 14 14 | Recovery with danaparoid and warfarin |
| 11 | M | 89 | Inpatient | 2 | 92 | 50‡ | 14 | Yes | Proximal deep venous thrombosis | 16 | Recovery with danaparoid and warfarin |
| 12 | F | 72 | Inpatient | 3 | 205 | 20‡ | 5 | Yes | Proximal deep venous thrombosis Warfarin-associated venous gangrene | 8 20 | Recovery with danaparoid and warfarin |

* First day of heparin use, day 0. Heparin was given in whole or in part by the subcutaneous route for patients 1, 3, 5, and 10.

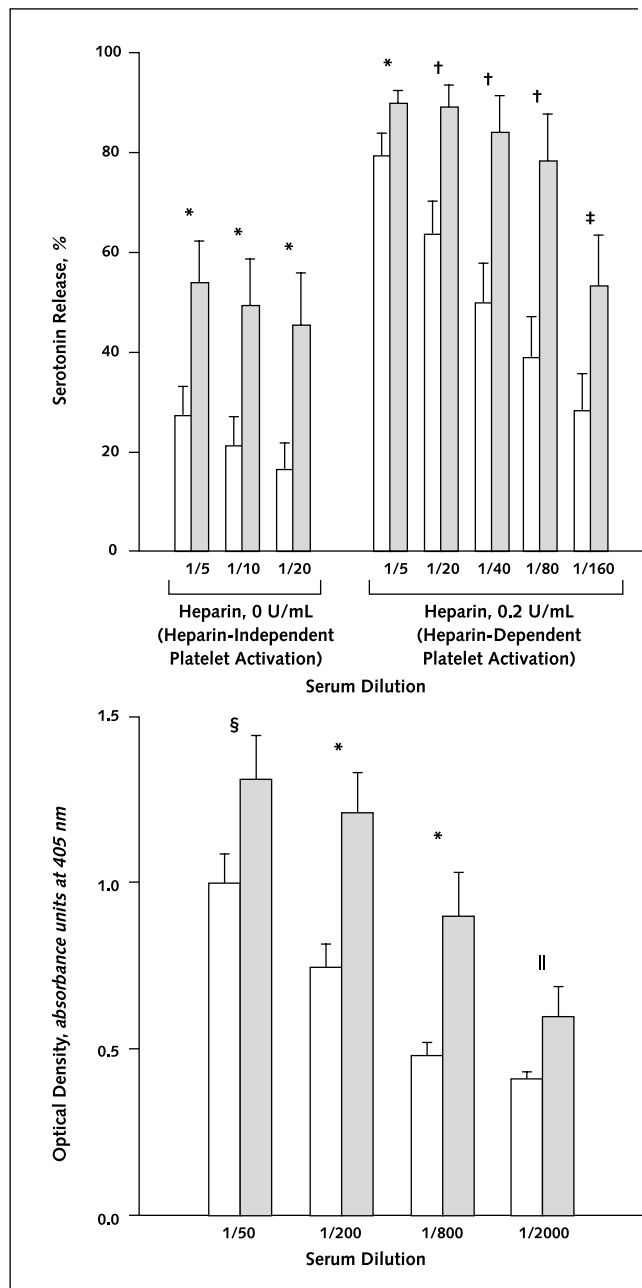
† Interval between last use of heparin and beginning of platelet count decrease (inpatient) or return to hospital with thrombosis (outpatient).

‡ Platelet nadir after further use of heparin.

§ Heparin exposure limited to flushes to maintain intravascular catheter patency.

|| The platelet count represents a 60% decrease from baseline in a patient with myeloproliferative disease.

Figure. Serum specimens from 12 patients with delayed-onset heparin-induced thrombocytopenia.



Top. Heparin-dependent and heparin-independent platelet activation produced by serum specimens from 12 patients with delayed-onset heparin-induced thrombocytopenia (gray bars) and 24 control patients with typical heparin-induced thrombocytopenia (white bars). Serum dilutions (final) ranged from 1/5 (standard assay) to 1/160. Addition of Fc receptor–blocking monoclonal antibodies consistently reduced the amount of serotonin release to less than 5% (not shown), confirming that the platelet activation was caused by IgG antibodies interacting with the platelet Fc receptors. Normal control specimens consistently produced less than 5% serotonin release under all reaction conditions (not shown).

drawal of heparin. We chose a minimum 5-day interval between the last heparin use and onset of thrombocytopenia (for inpatients) or onset of symptoms of thrombosis (for outpatients) because we expected that almost all heparin, whether given intravenously or subcutaneously, would have cleared by this time.

Six patients were identified in university-affiliated hospitals in Hamilton, Ontario, Canada, among approximately 180 consecutive patients with serologically confirmed heparin-induced thrombocytopenia. In addition, 3 patients were identified in secondary care hospitals and were then referred for further management at a Hamilton hospital, and 3 patients were identified in other secondary care hospitals during discussions with physicians who referred blood samples for diagnostic testing because of atypical presentations of heparin-induced thrombocytopenia.

Laboratory Testing

We performed two tests for heparin-dependent antibodies: the platelet serotonin-release assay (4) and an enzyme immunoassay that detects IgG antibodies that are reactive against platelet factor 4–heparin complexes (5, 6). We compared the 12 patient serum specimens with 24 randomly chosen control specimens from patients with heparin-induced thrombocytopenia that were matched for year of diagnosis and had been stored at -70°C . Heparin-dependent platelet activation was measured at an unfractionated heparin concentration of 0.2 U/mL (final), whereas heparin-independent platelet activation was measured without addition of heparin. Statistical comparison of the serologic results between patients with delayed-onset and those with typical heparin-induced thrombocytopenia was made by using the *t*-test (Quattro Pro 8 [1997], Corel Corp., Ottawa, Canada). The institutional review board at McMaster University approved the investigational protocol.

RESULTS

Clinical Outcomes

The Table summarizes the characteristics of 12 patients with delayed-onset heparin-induced thrombocyto-

Bottom. Platelet factor 4–heparin IgG antibodies detected by enzyme immunoassay. Serum dilutions (final) ranged from 1/50 (standard assay) to 1/2000. Normal control specimens yielded negative results (not shown). Error bars indicate the standard error of the mean. * $0.01 < P < 0.05$; † $P < 0.01$; ‡ $P = 0.063$; § $P > 0.2$; || $P = 0.193$.

penia. Thrombocytopenia and associated clinical sequelae began an average of 9.2 days (range, 5 to 19 days) after the last use of unfractionated heparin (no patients had received low-molecular-weight heparin). For all 6 inpatients, serial platelet counts decreased beginning at least 5 days after heparin withdrawal. Associated thrombotic events began an average of 8.3 days (range, 5 to 14 days) after heparin was withdrawn. Six outpatients developed thrombosis symptoms an average of 10.0 days (range, 6 to 19 days) after hospital discharge. For 8 of the 12 patients, the preceding period of heparin use that led to immune sensitization was 3 or fewer days. Four patients received heparin by subcutaneous injections.

All 12 patients developed one or more thrombotic complications. Venous thromboembolism occurred in 9 patients (3 with pulmonary embolism). Other complications (in 1 patient each) were bilateral lower limb artery thrombosis, postoperative occlusion of a vascular graft, digital ischemia, warfarin-associated venous gangrene, and adrenal infarction. Nine patients received additional heparin for the thrombosis, and all had further decreases in platelet count. One patient developed cardiac arrest 15 minutes after receiving the heparin bolus.

Serologic Studies

On both assays, serum specimens from the 12 patients with delayed-onset heparin-induced thrombocytopenia yielded strongly positive results for heparin-dependent antibodies (Figure). The patient specimens produced significantly greater heparin-dependent platelet activation at serial dilutions compared with control specimens. In addition, the 12 patient specimens produced significantly greater heparin-independent platelet activation than control specimens.

DISCUSSION

We discuss 12 patients with an unusual presentation of heparin-induced thrombocytopenia, characterized by delayed onset of thrombocytopenia and thrombosis, that began a mean of 9.2 days (range, 5 to 19 days) after withdrawal of all heparin. Many thrombotic and other complications occurred. Venous thromboembolism ($n = 9$) and thrombosis of arteries or arterial grafts ($n = 2$) were the most common; other unusual sequelae included adrenal hemorrhagic infarction, warfarin-associated venous limb gangrene, and transient global amnesia (2).

Three of the 12 patients (25%) had disseminated intravascular coagulation severe enough to be associated

with depletion of fibrinogen. This is a rare complication of heparin-induced thrombocytopenia (2, 7), but we believe it may be more common in patients with delayed-onset heparin-induced thrombocytopenia. In such patients, the heparin has left the circulation, allowing unopposed thrombin generation (8, 9) due to effects of procoagulant platelet-derived microparticles (10–12).

Our serologic studies showed that patients with delayed-onset heparin-induced thrombocytopenia have high-titer platelet-activating antibodies that exhibit increased heparin-dependent and heparin-independent platelet activation. It has been established that heparin-dependent antibodies recognize one or more sites on platelet factor 4 (5, 13) that are conformationally modified by pharmacologic heparin or endogenous glycosaminoglycans, such as endothelial-bound heparan sulfate (14, 15). Therefore, delayed-onset heparin-induced thrombocytopenia could be caused by high titers of heparin-dependent antibodies that recognize platelet factor 4 bound to the platelet surface in the absence of residual pharmacologic heparin, platelet factor 4 bound to endothelial glycosaminoglycans, or both.

Because our study is small, we cannot draw definitive conclusions about optimal treatment. Recent consensus recommendations state that heparin-induced thrombocytopenia should be treated with an agent that reduces thrombin generation (16, 17). In our study, patients who received an agent that inhibits either coagulation factor Xa (danaparoid) or thrombin (argatroban) seemed to have better outcomes than patients treated with ancrod, a defibrinogenating snake venom (18) that does not reduce thrombin generation (19) (Table).

It is generally assumed that heparin-induced thrombocytopenia can be avoided by limiting heparin use to fewer than 4 days (20). However, 8 of our patients received heparin for only 3 or fewer days. Thus, even if heparin use is restricted, at least a few patients will develop delayed-onset heparin-induced thrombocytopenia with life-threatening consequences. Low-molecular-weight heparin causes immune thrombocytopenia less often than unfractionated heparin (1, 6), so this complication of delayed-onset heparin-induced thrombocytopenia may occur less often in patients who receive low-molecular-weight heparin.

Physicians should consider delayed-onset heparin-induced thrombocytopenia when a patient presents with thrombosis and unexplained thrombocytopenia up to 3

weeks after the end of heparin therapy. Our study shows the importance of recognizing this syndrome. Nine of our 12 patients were re-treated with heparin, which caused platelet count to further decrease. In addition, the repeat use of heparin was associated with cardiac arrest in 1 patient. Early recognition of this syndrome is especially relevant because alternative, rapidly effective nonheparin anticoagulants (16, 17) are available.

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