Perioperative use of iloprost in cardiac surgery patients diagnosed with heparin-induced thrombocytopenia-reactive antibodies or with true HIT (HIT-reactive antibodies plus thrombocytopenia): An 11-year experience

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Thrombocytopenia and thromboembolism(s) may develop in heparin immune-mediated thrombocytopenia (HIT) patients after reexposure to heparin. At the Onassis Cardiac Surgery Center, 530 out of 17,000 patients requiring heart surgery over an 11-year period underwent preoperative HIT assessment by ELISA and a three-point heparin-induced platelet aggregation assay (HIPAG). The screening identified 110 patients with HIT-reactive antibodies, out of which 46 were also thrombocytopenic (true HIT). Cardiac surgery was performed in HIT-positive patients under heparin anticoagulation and iloprost infusion. A control group of 118 HIT-negative patients received heparin but no iloprost during surgery. For the first 20 patients, the dose of iloprost diminishing the HIPAG test to ≤5% was determined prior to surgery by in vitro titration using the patients’ own plasma and donor platelets. In parallel, the iloprost “target dose” was also established for each patient intraoperatively, but before heparin administration. Iloprost was infused initially at 3 ng/kg/mL and further adjusted intraoperatively, until ex vivo aggregation reached ≤5%. As a close correlation was observed between the “target dose” identified before surgery and that established intraoperatively, the remaining 90 patients were administrated iloprost starting at the presurgery identified “target dose.” This process significantly reduced the number of intraoperative HIPAG reassessments needed to determine the iloprost target dose, and reduced surgical time, while maintaining similar primary clinical outcomes to controls. Therefore, infusion of iloprost throughout surgery, under continuous titration, allows cardiac surgery to be undertaken safely using heparin, while avoiding life-threatening iloprost-induced hypotension in patients diagnosed with HIT-reactive antibodies or true HIT. Am. J. Hematol. 90:608–617, 2015. © 2015 Wiley Periodicals, Inc.

Introduction

Heparin-induced thrombocytopenia (HIT) is a severe complication of unfractionated heparin (UFH) or low-molecular-weight heparin (LMWH) and can result in potentially life-threatening thromboembolism (HIT/T) and even death [1–3]. The pathogenesis of HIT and HIT/T is owing to the development of mainly IgG antibodies that recognize the complex of PF4–heparin, forming a multimolecular complex of IgG-(PF4-heparin), which binds to FcγRII receptors on the platelet surface and crosslinks them. This binding/crosslinking induces intense platelet activation and aggregation, massive thrombin generation, and finally fibrin forms the skeleton of the thrombus [1–4]. Systemic anticoagulation is necessary during cardiac surgery. To date, the only well-established anticoagulation protocol involves the use of heparin, which owing to its predictability, rapid action and reversibility with protamine, becomes an integral part of cardiac operations performed with and without extracorporeal circulation. However, re-exposure of HIT or HIT/T patients to heparin should be avoided as the possibility of severe thromboembolic events and even risk of death is increased postoperatively [5–9] and on rare occasions during surgery (Melissari et al., unpublished data). In these patients, alternative anticoagulation must be used. Current HIT and HIT/T treatment approaches focus on the agents that rapidly control thrombin generation such as lepirudin, bivalirudin, and argatroban, or on the new thrombin generation-blocking agent danaparoid. However, lacking specific antidotes, all these agents are complicated with excessive blood loss during surgery and postoperatively [10–13]. Furthermore, danaparoid was a new drug in Greece

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when the study started and in the meantime both danaparoid and lepirudin have been withdrawn from the Greek market, whereas argatroban is not yet available in Greece, and bivalirudin was not available until more recently.

Early reports on anticoagulation for cardiopulmonary bypass (CPB) in patients diagnosed with HIT described the use of full heparinization combined with one or more antiplatelet agents including tirofiban [14]. In a number of small studies and case reports, iloprost, a stable analogue of prostacyclin, has been used [15–20]. However, the use of iloprost is associated with severe hypotension [20,21].

Here, we report that over an 11-year period we screened 530 patients from a total of 17,000 patients, who underwent cardiac surgery at the Onassis Cardiac Surgery Center (OCSC) to diagnose the patients with HIT-reactive antibodies, who we refer to as “HIT-positive” and patients with HIT-reactive antibodies who were also thrombocytopenic just prior to surgery, who we refer to as “true HIT” patients. Inclusion of patients for screening was based on a panel of criteria adopted by the OCSC protocol, which permitted the preoperative as well as postoperative identification of all HIT-positive or true HIT patients. Screening identified 110 HIT-positive patients out of which 46 also had thrombocytopenia just prior to cardiac surgery and were therefore “true HIT.” We describe our management of the 110 HIT-positive patients resulting in a safe and full anticoagulation with UFH during heart surgery using iloprost under continuous titration as well as patient outcome. One hundred and eighteen out of the remaining 420 patients who had no previous history of HIT (HIT negative), and had no detectable antibodies just before surgery, were selected as controls for the study. These patients had only been exposed to heparin once during their cardiac catheterization. Furthermore, the control group had similar demographic and preoperative risk factors as the HIT-positive group and underwent heart surgery of similar magnitude and complexity but received no iloprost.

Study limitations

This study has the limitation of not using a true control group. The patients in the control HIT-negative group had similar demographic, preoperative risk factors, and underwent heart surgery of similar magnitude and complexity as the HIT-positive group. However, these patients did not receive iloprost as it was not ethical to expose them to an unnecessary treatment. Similarly, it was not also ethical to include a control group of patients who tested positive for HIT-reactive antibodies (or had true HIT) but were not administered iloprost infusion during surgery. In these patients, the possibility of life-threatening thromboembolic events would be increased during the postoperative period owing to re-exposure to heparin during heart surgery.

Materials and Methods

At the OCSC, our policy of avoiding devastating postoperative consequences after re-exposure to heparin during heart surgery in patients found just prior to surgery to have HIT-reactive antibodies (with or without thrombocytopenia) is based first, on an initial screening of all patients at risk. If patients are identified as HIT positive only, or with true HIT (HIT-reactive antibodies + thrombocytopenia), then an established management procedure is followed using iloprost in combination with heparin during surgery. Finally, the administration of heparin or LMWH postoperatively is avoided and patients are managed using alternative anticoagulation.

Patients. Five hundred and thirty out of 17,000 patients, who underwent heart surgery over an 11-year period from January 2000 to December 2011, were screened for HIT preoperatively according to the OCSC protocol. One hundred and ten patients were found to have HIT-reactive antibodies, 46 out of them had also thrombocytopenia (HIT) or true HIT at the time of cardiac surgery. These patients received iloprost during heart surgery (treated group). One hundred and eighteen of the remaining 420 HIT-negative patients who had no previous history of HIT (HIT negative) and had been exposed to heparin only once during their cardiac catheterization neither had detectable HIT antibodies when the assessment of HIT was carried out just before surgery (O.D. <0.300 in the EIA test) but who nevertheless had similar demographic, preoperative risk factors and type of heart surgery to the HIT-positive patients group were included in the study as a control group and received no iloprost during surgery (Tables 1 and II). All 228 participants to the study were patients requiring urgent or semi-urgent heart surgery and all received systemic anticoagulation with heparin during the procedure. Iloprost was infused perioperatively under continuous titration in the 110 HIT-positive patients. All patients signed prospectively an informed consent in accordance with the Declaration of Helsinki for enrollment into the study. Approval was obtained from the Ethics Board Committee of the OCSC Hospital. All authors had access to the primary data of the study.

Criteria for HIT antibody screening. Cardiac surgery patients have often been exposed to heparin, for example during an earlier cardiac catheterisation(s), coronary angioplasty for the treatment of acute coronary syndrome, or during previous vascular surgery. Therefore, at the OCSC we developed a local protocol for the identification and management of HIT-positive patients. This involves: (a) a preoperative HIT screening which excludes all the opportunity for the possible HIT-reactive antibodies or with true HIT undergoing heart surgery to be recognised/identified preoperatively, (b) management of cardiac surgery HIT patients, and (c) postoperative HIT antibody screening. Patients were selected for HIT antibody screening provided they (i) had a marginal platelet count (~150 × 10^9/L) or thrombocytopenia possibly related to the previous use of UFH or LMWH for more than three consecutive days; (ii) developed thrombocytopenia owing to HIT or HIT/T during a previous exposure to UFH or LMWH; (iii) developed thrombocytopenia + thrombosis during current anticoagulation with UFH, or LMWH or danaparoid; (iv) were exposed to UFH or LMWH in the preceding 12 months associated with a platelet count reduction of more than 30% irrespectively of platelet count, for more than three consecutive days. HIT antibody screening was also carried out postoperatively in patients with: (i) a persistent, and (ii) a significant drop in the platelet count (more than 30%) associated or not with thromboembolic events. In addition, the assessment of HIT was carried out postoperatively: (a) in the control group (118 patients assessed by EIA) to detect possible HIT seroconversion owing to the use of heparin during heart surgery and (b) in the HIT-positive patients (110 patients assessed both by EIA and heparin-induced platelet aggregation assay [HIPAG]) to detect any rise in the strength of HIT-reactive antibodies following the administration of heparin during heart surgery.

Hematological measurements and HIT antibody evaluation. For the purposes of this study, blood samples were withdrawn preoperatively, postoperatively (admitted to ICU), on the fourth postoperative day for the assessment of HIT and on discharge. Blood samples were withdrawn in 3.2% of trisodium citrate (at a ratio of 9:1) for platelet aggregation tests or EDTA for the assessment of whole-blood count.

Whole-blood count was carried out on an LH 780 Coulter analyzer (Beckman-Coulter, USA) and platelet aggregation was performed on a PAP-4 aggregometer (Biodata, USA). We used HIT plasma, collagen, ADP, and arachidonic acid to stimulate the platelets of donors used for the assessment of HIPAG to confirm that platelet receptors were functional [22,23]. Ristocetin was also included as a fourth platelet agonist. Overall, these platelet agonists were used to ensure the quality of donor platelets for the evaluation of HIPAG. The quality of the platelet was ascertained each time a functional test for the assessment of HIT antibodies was performed. Platelet-rich plasma (PRP) was obtained by centrifugation at 800 rpm for 8 min and platelet-poor plasma (PPP) by subsequent centrifugation for 10 min at 3,000 rpm.

Two diagnostic tests for HIT antibodies detection were performed. First, an immunomassay where antibodies against PF4–heparin complexes was detected using a commercial solid-phase enzyme immunoassay (EIA) kit (Asserachrom HPIA, Diagnostica Stago, Asnières, France) according to the manufacturer’s instructions. A test was considered strong positive if an optical density (OD) of ≥1.200 was recorded. Plasma samples that gave OD values between 0.900 and 1.100 were considered as weakly positive. HIT was also assessed by a functional test for the detection of HIT-reactive antibodies. The test was carried out through monitoring donor’s platelet activation by antibodies to PF4–heparin in patient’s plasma and stimulated by heparin according to the method described by Choug [24,25] modified slightly by us adding a third point (heparin, 2.5 IU/mL) instead of two (HIPAG) to increase the sensitivity of the test. The assessment was carried out on a standard platelet aggregometer. Donor PRP was resuspended in patient PPP to a final platelet count of 200 × 10^9/L to achieve reliable and reproducible results, and aggregation was evaluated after the addition of heparin. HIT antibodies produce platelet activation with 0.1, 0.5, and 2.5 IU/mL heparin (equivalent to heparin concentration used during extracorporeal circulation), which is inhibited in the presence of excess heparin (100 IU/mL). Platelet aggregation was the end point of such the assessment of platelet activation (HIPAG test). A positive response was characterised by a lag phase of several minutes after the addition of heparin, followed by irreversible aggregation. In HIPAG-positive results, all tested concentrations of heparin produced aggregation.

The examples of aggregation plots are shown in Fig. 1A, B.
In all 110 patients with confirmed clinically relevant HIT antibodies, the HIPAG test was repeated 1 day before surgery, and in all 110 patients HIT-reactive antibodies were detected the day before surgery. HIT plasma samples prepared from donors with clinical history of HIT were used each time to assess donors’ platelets bodies were detected the day before surgery. HIT plasma samples prepared from test was repeated 1 day before surgery, and in all 110 patients HIT-reactive anti-

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was transiently ceased as no heparin was administered to the patient until after the establishment of the iloprost target dose. If aggregation developed upon the addition of heparin in vitro, then the iloprost infusion rate was doubled and continued for a further 10 min under constant monitoring of blood pressure and heart rate. A second blood sample was then withdrawn and a further HIPAG assessment was performed. The process of increasing the rate of iloprost infusion was continued until a negative HIPAG test was achieved (the absence of aggregation, i.e., target dose). After a negative HIPAG test, the intravenous infusion of iloprost was restarted and continued at the rate which was identified by the last HIPAG test (target dose), and then heparin was administered as per usual hospital practice. After heparinization and just before CPB a new HIPAG test was carried out (without heparin addition in the test in this case) to ensure that it remained negative. At the completion of surgery, heparin was neutralized with protamine and the dose of iloprost was halved. At this point, a further HIPAG test was carried out to exclude that any circulating heparin remained that could potentially cause platelet activation/aggregation. Shortly after the admission to ICU, the dose of iloprost was halved again but continued for approximately a further 60 min before being stopped to anticipate any possible early heparin rebound phenomenon.

For the first 20 patients, a close correlation between the actual target dose of iloprost established intraoperative and the dose that was identified before surgery in vitro was observed. Therefore, for the subsequent 90 patients, infusion of iloprost was commenced using the “target dose,” which was determined in vitro prior to surgery as described above. Target dose was established as the amount of iloprost that had to be added in vitro to prevent platelet aggregation with the addition of heparin and was based on the assumptions that (i) blood volume remained constant before surgery and (ii) plasma volume for both men and women is 40–50 mL/kg and (iii) total blood volume is 60–80 mL/kg. As for the first 20 patients, 10 min after administering the target dose of iloprost after the commencement of anesthesia, blood was withdrawn and a HIPAG test was performed to establish whether platelet aggregation occurred. If so, then the dose of iloprost was increased until a negative HIPAG test was achieved (intraoperative titration) as described above. Importantly, the process of establishing the target dose in vitro prior to surgery significantly reduced the number of the assessments of HIPAG that were required during anesthesia, and thus saving critical surgical time. In addition, it ensured that all the patients received the exact amount of iloprost that was required and not more.

Statistical analysis. The values (mean ± SD) were compared between these two groups (treated and controls) using parametrical (t-test) or nonparametrical procedures (Mann–Whitney tests [median, quartiles] and Wilcoxon signed-rank test) accordingly. Repeated-measures one-way ANOVA was used for the comparison of blood pressure changes over time between both groups. Pearson’s correlation was used to assess the association between the target dose of iloprost in vitro and in vivo.

### Results

How HIT-reactive antibodies were diagnosed preoperatively in 110 patients requiring heart surgery

Five hundred and thirty out of 17,000 patients requiring heart surgery met the criteria for HIT antibody screening preoperatively.

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**Figure 1.** The assessment of HIPAG before neutralization by iloprost in two patients with HIT-reactive antibodies requiring a target dose of iloprost of 12 ng/kg/min on top (A), and the highest one (24 ng/kg/min) on the bottom (B) to diminish HIPAG response to <5%. The lag phase was 10 min in the first patient compared to 3 min in the second. Arrow indicates the addition of heparin. (C) The effects of iloprost at various concentrations and corresponding HIPAG at various concentrations of UFH in vitro. (●, No iloprost; ▲, iloprost 60 pg/mL (=3 ng/kg/min); ■, iloprost 120 pg/mL (=6 ng/kg/min); asterisk represents iloprost of 240 pg/mL (=12 ng/kg/min).
Intraoperative and in vitro identified iloprost target dose in HIT-positive patients

Overall, the mean target dose of iloprost established intraoperatively (7.63 ng/kg/min) was not significantly different than the in vitro mean target dose of 6.9 ng/kg (±3.24) (paired t-test t = 2.96, P = 0.004) for all 110 treated patients. Furthermore, in approximately 80% of the cases the target dose identified in vitro was identical with the one specified intraoperatively. Interestingly, the target dose of iloprost identified in vitro before the operation showed excellent correlation with that identified intraoperatively (correlation coefficient, 0.768; P < 0.001). The distribution of the target dose of iloprost identified in vitro and in vivo is shown in Fig. 3. By initially applying in vivo the in vitro established target dose of iloprost for HIPAG neutralization and then by increasing the rate of iloprost infusion, fewer assessments of HIPAG were required intraoperatively, and thus saving critical surgery time. In fact, for 88 HIT patients the required number of intraoperative assessments of HIPAG was limited to one and for the remaining 22 patients no more than three assessments were needed.

Patients before surgery requiring very high doses of iloprost for HIPAG neutralization and their management

Five patients (three with true HIT) in the treated group had an initial highly positive response (EIA OD, >2.200 and HIPAG >80%
and a lag phase about 3 min) (Fig. 1B) in association with a very high target dose of iloprost in vitro (3.4–3.8 \( \text{ng/mL} \)) for a HIPAG of <5%, which corresponded to an in vivo target dose of \( \geq 48 \text{ ng/kg/min} \). Because such a high dose of iloprost would most probably have led to uncontrollable hypotension in these five patients, their heart surgery was postponed (their condition allowed it) for between 2 weeks and 2 months in order for their HIT antibody titer to decline (HIPAG = 50–70%). Afterward, these patients were included in the study. During this holding back surgery time, all five patients were under close cardiac observation (two patients remained in hospital). It appears, therefore, that the target iloprost dose identification in vitro indicates in advance whether heart surgery in patients with HIT-reactive antibodies or HIT patients is safe under full anticoagulation with UFH. Obviously, neither LMWHs nor heparin (in any form) were given postoperatively.

Hematological measurements and blood transfusion requirements in patients and controls

The mean preoperative platelet count was much higher in the control group (190.1 ± 79.3 \times 10^9/L) than in the iloprost-treated group (153.5 ± 28.5 \times 10^9/L) (\( P = 0.001 \)). In both groups, a significant percentage drop in platelet counts from preoperative evaluation to ICU admission was observed (36% for the control group and 23% for the treated group \( [P < 0.001 \text{ for both groups}] \)). At discharge, platelet counts in both groups were more than 200 \times 10^9/L. During hospitalization, a platelet count for all patients included in the study was carried out on a daily basis. None of the patients showed a decrease in platelet count of >20% after admission to the ICU and until discharge. Furthermore, before surgery both Hb and Ht were significantly higher in the control group than in the iloprost group: (13.5 g/dL ± 1.7 over 12.9 g/dL ± 1.6, \( P = 0.009 \) and 40.8% ± 4.7 over 39.1% ± 4.4 \( P = 0.007 \), respectively). Differences postoperatively were not statistically significant (\( P = 0.12 \) for Hb; \( P = 0.06 \) for Ht) (Tables IV and V). No significant difference was noted between the groups in the volume of postoperative blood loss (median, quartiles: 655, 490–970 for the control group and 705, 515–1,190 for the treated group, respectively) seen mainly as mediastinal bleeding and excessive chest–tube drainage (Table II). Re-exploitation owing to excessive bleeding was undertaken in both groups: nine patients in the treated group and eight patients in the control group, this difference being not significant. No significant difference was noted in the transfused units of red cell (median: 5 U, 90%; 4 U, 82% for the treated and control groups, respectively); of fresh frozen plasma (4 U, 79%; 4 U, 69.5%); of platelet concentrates (5 U, 32.7%; 5 U, 28.8%) or in the apheresis units of platelets (1 U, 22.7%; 1 U, 30.5%) (Tables IV and V).

Blood pressure in patients and controls

In the control group, the mean systolic blood pressure decreased by 26.5 ± 11.1 mmHg before CPB, whereas in the iloprost group the mean systolic pressure decreased by 37 ± 10.3 mmHg before CPB (\( P = 0.01 \), Wilcoxon test). Serious hypotension was seen in 17 patients in the treated group with an overall incidence of 15.5% as opposed to 12 patients (10.2%) in the control group (\( P = 0.003 \)). In all 17 patients in the treated group, hypotension was attributed to iloprost and all 17 patients received a target dose of iloprost \( \geq 12 \text{ ng/kg/min} \). However, hypotension was successfully treated using nor-epinephrine infusion (1–4 \( \mu \text{g/kg/min} \)) or a 100 \( \mu \text{g} \) phenylephrine bolus.

Postoperative complications/adverse events in iloprost-treated patients and controls

The duration of ICU stay and length of postoperative hospitalization were similar in both groups (Table II). On the 4th postoperative day, all 228 patients who were included in the study were assessed for the presence for HIT-reactive antibodies by EIA and HIPAG, the latter assessed only when EIA was found to be positive. Seroconversion was not observed in any of the 118 control group patients (range of OD in the EIA assay, 0.080–0.280). Furthermore, the titer of postoperative HIT antibodies did not increase in any of the 110 HIT-positive iloprost-treated patients. In this latter group, the OD range of the EIA assay was 0.700–1.100 for the 22 patients who required a low target dose of infused iloprost intraoperatively (3 ng/kg/min), whereas for the remaining 88 patients, who required higher doses of iloprost, the corresponding range of OD in the postoperative EIA assay remained unchanged (\( \geq 1.200 \)). In addition, on the 4th postoperative day, the HIPAG test in 105 of the HIT patients was found unchanged, whereas in five patients with HIT the HIPAG test was negative. These latter five patients also had a low OD value of <1.100 measured by EIA assay. Postoperative complications for both treated and control groups are summarized in Table VI. The incidence of thrombotic or thrombembolic events was similar in both groups: 5.4% (six patients EIA: OD, 0.080–0.200) in the control group and 5.1% (six patients EIA OD, 1.300–1.800) in the iloprost-treated group. These events included stroke, peripheral vascular occlusion, deep vein thrombosis, and pulmonary embolism. Operative (30-day) mortality
was similar in both groups: 8.2% in the treated group (nine patients) compared to 8.5% in the controls (10 patients). The cases of infection (10 vs. 4) and mediastinitis (2 vs. 0) were higher in the iloprost-treated group. It is not clear whether these differences were related to the longer surgical times of the treated group. Overall, fewer patients (49, 44.5%) in the treated group experienced adverse events ($X^2 = 6.85, P = 0.009$) compared to the controls (74, 62.7%).

### Discussion

At the OCSC, which is a Reference Center for heart disorders in Greece, from January 2000 to over an 11-year period, 530 patients out of 17,000 requiring heart surgery were screened for the presence of HIT antibodies preoperatively according to an established HIT screening protocol. Criteria selection of patients for preoperative HIT screening included any previous history of HIT. In contrast, the ACCP guidelines published in 2008 regarding HIT screening criteria indicate that screening should be extended only up to 100 days post-HIT [29]. However, Warkentin and Sheppard [30] recently detected HIT-reactive (platelet activating) antibodies that were evident approximately 1 week postcardiac surgery in 47% of former patients with HIT who were deliberately rechallenged with heparin between 8 weeks and 13.5 years (mean, 4.4 years) after their initial exposure. The authors attributed this HIT immune response to a secondary one involving long-lived B-lymphocytes. Therefore, the OCSC protocol is likely to minimize the risk that HIT-positive patients undergoing heart surgery may not be identified pre- and postoperatively. Furthermore, the risk of overdiagnosis of HIT is reduced as in the study only patients tested and found positive for HIT by both PF4-UFH dependent EIA and functional assay (HIPAG) were included.

One hundred and ten patients were found to be HIT positive despite having a relatively high mean platelet count of 153.5 × 10^9/L (±28.5/L, SD). This confirms that HIT antibodies could exist without the presence of true thrombocytopenia [5,31]. In addition, similar to the previously published reports [32], we found that approximately one-fifth of the screened patients (110 out of 530) undergoing CPB surgery had detectable HIT antibodies before the procedure as a
result of prior heparin or LMWH heparin exposure, 46 of whom suffered true HIT preoperatively (mean platelet count 126 × 10^9/L [±15.0, SD]). Therefore, the incidence of preoperative HIT-reactive antibodies in patients scheduled for cardiac surgery was 0.65% (110 out of 17,000 patients), which is similar to the frequency of HIT previously described in medical patients exposed to heparin or LMWH (0.63–0.84%) [33,34]. In addition, we found that the incidence of true HIT preoperatively was 0.27%.

It appears that a significant number of patients with HIT platelet-activating antibodies develop thromboembolic events after re-exposure to heparin. According to Prandoni et al. [33], this occurs in up to 36% of cases. In addition, Kress et al. [5] state that "the presence of HIT antibodies even without reactivity toward platelet [HPF4] before surgical heparin administration is an independent and clinically significant risk factor for postoperative adverse events after cardiac surgery and that the presence of anti-[heparin-PF4] contraindicates further heparin exposure during cardiac surgical procedures.”

Therefore, we aimed to avoid such serious postoperative complications caused by surgical heparin re-exposure in 110 patients undergoing heart surgery who were diagnosed with HIT-reactive antibodies only, or with true HIT, just prior to surgery by using intraoperative infusion of iloprost. Of the remaining 420 patients who tested negative for HIT antibodies, 118 were included in the study as a control group and received no iloprost. It was observed that in the majority of patients with true HIT (characterized by the presence of HIT-reactive antibodies and thrombocytopenia just prior to cardiac surgery), a higher mean target dose of iloprost infusion was required (>10 ng/kg/min) to prohibit platelet activation during re-exposure to heparin intraoperatively, than in the majority of patients having HIT-reactive antibodies only (infused iloprost, ≤6 ng/kg/min).

Thrombocytopenia was evaluated by checking platelet counts on a daily basis for all the 228 patients who participated in the study regardless of their HIT status. In our study, HIT seroconversion was not observed postoperatively in the control patients and the titer of HIT antibodies did not increase in the iloprost-treated group. In fact, antibody titer decreased in a number (five) of iloprost-treated patients. This was possibly owing to drawing the specimen <5 days after surgery, and our policy of limiting the exposure of control patients to UFH or LMWH to no more than 3 days; for longer periods, alternative anticoagulation was given such as danaparoid or fondaparinux. Naturally, postoperative thromboprophylaxis in all HIT-positive diagnosed patients was carried out with danaparoid (after the exclusion of immunologic crossreactivity of the HIT antibodies with danaparoid or fondaparinux, which has recently replaced it). In the article by Bauer et al. [32], the authors attributed the increased incidence of HIT antibodies detected on the 5th postoperative day in 111 patients who underwent heart surgery (5% before surgery, 13% postoperative with SRA; 19% before and 51% postoperative with ELISA) to an anamnestic response in the previously sensitized individuals. In addition, all these 111 patients were exposed to heparin within 24 hr post surgery (1st postoperative day). However, apart from the fact that the study design and purpose of the two studies were different, in our study postoperatively the titer of HIT-reactive antibodies was unchanged or reduced. In addition, the HIT antibody reactivity intraoperatively had temporarily vanished with iloprost and until 60 min after heparin neutralization by protamine, in all 110 patients. Furthermore, no further heparin was given postoperatively and instead alternative anticoagulation was administrated. Therefore, intraoperative heparin most likely had no opportunity to perturbate further the immune system in all these 110 treated patients. In addition, as these 110 patients had already HIT-reactive antibodies on the day of the surgery it should be expected that if an increase of HIT antibodies would develop owing to intraoperative UFH exposure, then it should be obvious by the 4th postoperative day or even earlier as the need of “at least 5 days” to form antibodies would not exist owing to the fact that these antibodies were already present in other words seroconversion existed. Further support to our results is provided by the fact that none of the patients showed a decrease in platelet count of >20% after admission to the ICU and until discharge and that all of our patients had normal platelet counts on the day of their discharge form the hospital and remained free from thromboembolic complications approximately 3 weeks later when assessed during their first postsurgery appointment.

Furthermore, all our 118 control patients had no previous exposure to heparin, except during catheterization (HIT negative), and had no previous history of HIT or detectable HIT antibodies when the assessment of HIT was carried out just before surgery. In addition, postoperative exposure of these control patients to UFH or LMWH was limited up to 3 days; for longer periods, alternative anticoagulation was given, which diminished the possibility of HIT seroconversion.

The detection of HIT antibodies was carried out with EIA followed by the detection of their reactivity using the HIPAG test with the application of a two-point definition for a positive assay according to Chong [24,25], and by adding a third point, namely 2.5 U/mL of heparin (HIPAG) to increase the sensitivity of the test. This concentration possibly correlates with one of the lower concentrations of heparin used during CPB [35]. Hence, heparin administration in vivo intraoperatively was carried out when the infused iloprost had diminished the performed HIPAG to <5% for all the used concentrations of heparin in vitro.

Iloprost is a stable analogue of prostacyclin; thus, it stimulates adenylylcyclase, resulting in increased platelet c-AMP, which prevents in dose-dependent manner platelet activation by various platelet agonists including HIT antibodies. Because of its short half-life of 15–30 min, platelet reactivity returns approximately 3 hr after cessation. Olinger et al. [15] in 1984 were the first to identify that the use of iloprost could inhibit heparin-dependent platelet activation in the presence of HIT serum. Since then, small studies referring to the UFH’s deleterious effects being offset by iloprost administration in patients with HIT requiring cardiac surgery have been published [16–20,36,37]. The major drawback of iloprost is its effect on vaso-motor tone, particularly the marked vasodilatation and life-threatening hypotension that may result. However, in 2004, we reported [38] that in contrast to what was published by Krause and Krais [21] and Kraenzler and Starr [20], severe hypotension owing to iloprost could be anticipated with increased doses of phenylephrine. In this study, by titrating the dose, and by finding that the mean target dose of iloprost established intraoperatively was not significantly different from the in vitro mean target dose of iloprost possibly excess iloprost was not administered and hence life-threatening hypotension was avoided. Profound hypotension was seen when the concentration of infused iloprost exceeded 12 ng/kg/min (data not shown), and this was controlled using norepinephrine infusion (1–4 µg/kg/min) or a phenylephrine bolus (100 µg). In addition, heart surgery was postponed, whenever possible, when a high concentration of iloprost (>480 pg/mL) was required to diminish the HIPAG test in vitro. This was the case for 5 out of 110 patients with HIT (three with true HIT) who underwent heart surgery safely when the HIPAG test in vitro was about 50–70%. Furthermore, we did not observe increased postoperative bleeding over the control patients, probably owing to the quick platelet reactivity recovery after cessation of iloprost. In addition, there were no differences in transfusion requirements (P = 0.13). The incidence of thromboembolic complications and ischemic cerebrovascular adverse events was similar in both groups: 5.4% in the iloprost (six patients) and 5.0% in the control...
group (six patients) as well as the 30-day surgical mortality (9 patients in the treated group and 10 patients in the control group). The operative mortality for both groups was zero. Iloprost acts immediately on infusion and therefore its administration in all 110 patients started after the induction of anesthesia and approximately 60 min before heparinization. This was well-spent time allowing for the performance of HIPAG test neutralization, ascertaining that the patient's platelets would not aggregate by heparin. Doses between 3 and 24 ng/kg min were needed to achieve complete inhibition of heparin-induced platelet aggregation (HIPAG, <5%). The identification of the target iloprost dose is a time-consuming procedure as each concentration being tested takes at least 40 min to complete. In all 110 patients, initially the target dose of iloprost was determined in vitro and prior to surgery. In addition, in 80% of the cases the target dose identified in vitro is identical with the one specified in intraoperatively. Therefore, by initially applying in vivo (for HIPAG neutralization) the target dose of iloprost established prior to surgery in vitro, critical surgery time was saved by performing fewer assessments of HIPAG intraoperatively. The overall iloprost infusion was halved after protamine administration and again on the entrance to ICU. It was continued in this reduced concentration for approximately another 60 min before being stopped. In all 110 cases, all HIPAG tests remained negative after each reduction of iloprost infusion.

Conclusions

Based on our 11-year experience at the OCSC, we conclude that infused iloprost throughout surgery under continuous titration allows cardiac surgery to be undertaken safely, despite re-exposure to heparin in patients diagnosed with HIT-reactive antibodies or true HIT. Life-threatening hypotension is avoided and blood pressure is controlled, if needed, with no-repinephrine infusion. It is advisable, however, to postpone cardiac surgery, whenever possible, when the required dose of iloprost to neutralize the HIPAG in vitro is significantly high, and hence the possibility of a life-threatening hypotension cannot be excluded.

References


