Subtle differences in commercial heparins can have serious consequences for cardiopulmonary bypass patients: A randomized controlled trial

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Objective: To compare the potency, reversibility, and perioperative bleeding risk of Hepalean with those of PPC heparin.

Methods: Because in vitro testing failed to detect differences in the potency or protamine reversibility of the 2 heparin preparations, we conducted a parallel group, single-center, double-blind, randomized, controlled trial to compare the anticoagulant effects of Hepalean to those of PPC heparin in patients undergoing coronary artery bypass grafting with cardiopulmonary bypass.

Results: From June 1, 2011, to June 30, 2011, we randomly assigned 11 patients to receive PPC heparin and 10 to receive Hepalean. Despite similar initial doses of heparin, the median initial activated clotting time was numerically lower in the PPC heparin group than in the Hepalean group (median, 516.0 seconds; interquartile range, 481.0-633.0; vs median, 584.0 seconds, interquartile range, 520.0-629.0; $P = .418$). Those given PPC heparin required a greater total heparin dose (median, 46,000.0 U; interquartile range, 39,500.0-60,000.0 vs median, 34,500.0 U; interquartile range, 32,250.0-37,000.0; $P = .011$) and a greater dose of heparin per kilogram than those given Hepalean (median, 572.9 U/kg; interquartile range, 443.0-659.7 vs median, 401.1 U/kg; interquartile range, 400.0-419.4; $P = .003$). The key secondary results included an increased median total protamine dose (median, 600.0 mg; interquartile range, 550.0-700.0; vs median, 500.0 mg; interquartile range, 425.0-542.5; $P = .026$) and a trend toward increased chest tube output within 24 hours (median, 830.0 mL; interquartile range, 425.0-1135.0; vs median, 702.5 mL; interquartile range, 550.0-742.5; $P = .324$).

Conclusions: PPC heparin use was associated with greater heparin and protamine dose requirements than Hepalean. These findings indicate that heparin preparations are not interchangeable and suggest that a direct comparison of the potency with the brand in use is needed if a change is made to ensure that the agents exert similar anticoagulant effects in vivo. (*J Thorac Cardiovasc Surg* 2012;144:944-50)

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Most cardiac surgical operations are performed using cardiopulmonary bypass (CPB) that maintains the systemic blood circulation during cardioplegia. Heparin is routinely used to inhibit coagulation and prevent thrombus formation on the surfaces of the bypass circuitry and the oxygenator.

In patients undergoing cardiac surgery, the first dose of heparin is given before CPB, and the anticoagulant response is monitored using a point-of-care device that measures the activated clotting time (ACT).^{1,2} The usual target ACT before CPB is at least 480 seconds.^{3,4} The ACT is monitored throughout the procedure, and additional heparin doses are given to maintain this ACT target. At the end of the procedure, the anticoagulant effect of heparin is reversed with protamine sulfate.⁵

Heparin is most commonly derived from porcine intestinal mucosa, and several brands are commercially available. Hamilton Health Sciences recently switched from Hepalean (Organon Teknika, Toronto, Ontario, Canada) to Pharmaceutical Partners of Canada (PPC) heparin (PPC, Richmond Hill, Ontario, Canada) because the production of Hepalean

Abbreviations and Acronyms

ACT	= activated clotting time
aPTT	= activated partial thromboplastin time
CABG	= coronary artery bypass grafting
CPB	= cardiopulmonary bypass
PRP	= platelet-rich plasma

was discontinued. After switching heparin brands, cardiac surgeons, anesthesiologists, perfusionists, and intensive care unit staff had the impression that greater doses of PPC heparin were needed to achieve a minimum threshold ACT, more protamine was needed to reverse the effects of PPC heparin, and the incidence of bleeding complications was increased.

Differences in the potency between heparin brands have been previously described in *in vitro* studies⁶ and have been investigated clinically.⁷ In addition, investigators at several institutions have reported differences in heparin and protamine dosing when replacing Hepalean with PPC heparin.^{8,9} However, differences between heparin brands have not been rigorously evaluated in a randomized trial.

In response to the concerns of the cardiovascular surgical team at our institution, we first conducted a retrospective audit of patients treated with PPC heparin compared with historical controls treated with Hepalean. Next, we performed *in vitro* studies to compare the potency of the 2 heparin brands. Because neither of these exploratory investigations was conclusive, we then undertook a randomized controlled trial to compare the anticoagulant effects and reversibility of PPC heparin and Hepalean in patients undergoing coronary artery bypass grafting (CABG) with CPB.

METHODS

All investigations were performed at the Hamilton General Hospital in Hamilton, Ontario, Canada. Ethics approval was not required for the retrospective audit, because it was performed as a part of routine clinical care. The Research Ethics Board of Hamilton Health Sciences approved the protocol for the randomized controlled trial. All participating patients provided written informed consent. The clinical trial registration number was NCT01343381 (www.clinicaltrials.gov).

Phase I: Retrospective Chart Audit

We reviewed the medical records of 10 consecutive patients who had undergone isolated CABG surgery by a single surgeon from January 12 to February 10, 2011, and received PPC heparin and compared them with 10 consecutive patients undergoing isolated CABG from March 26 to April 27, 2010, who had received Hepalean. We collected the baseline patient characteristics and operative data, including the amount of heparin administered intraoperatively, ACTs, and protamine dosages. We recorded the charted postoperative chest tube drainage, blood product requirements, and the results of routine coagulation tests.

Phase II: In Vitro Studies

Venous blood samples were collected from 5 healthy donors into 0.105-M sodium citrate anticoagulated tubes and into 2 sets of 1-mL syringes prefilled

with 40 μ L of differing concentrations of either PPC heparin or Hepalean. The final concentration of each type of heparin in the syringes was 0.5, 1, 2, and 4 USP U/mL. As each syringe was filled, it was detached from the butterfly set, and the contents were dispensed into plastic test tubes to enable thorough mixing of the anticoagulant. ACT testing was then performed using a Hemochron Junior Signature Plus instrument (International Technidyne Corporation, Edison, NJ) according to the manufacturer's instructions. A syringe containing 40 μ L of 0.15 M NaCl instead of heparin was collected last and tested immediately to obtain a control ACT measurement for each donor. Sodium citrate anticoagulated donor samples were centrifuged at 2000g for 10 minutes. Plasma was harvested and centrifuged again at 2000g for 10 minutes to minimize platelet contamination. The double-spun donor plasma samples were frozen in aliquots at -80°C within 2 hours of collection and thawed at 37°C at assay.

Hepalean or PPC heparin was added to the donor plasma to yield the final concentrations of 0.5, 1, 2, and 4 U/mL. An equal volume of 0.15 M NaCl was added to plasma instead of heparin as a baseline control. The activated partial thromboplastin times (aPTTs) and anti-factor Xa levels were determined. aPTT assays were performed using a Diagnostica Stago coagulation analyzer (Diagnostica Stago, Parsippany, NJ) and Dade Behring Actin FSL reagent (Dade-Behring, Marburg, Germany). Anti-factor Xa assays were performed using an Amelung Sigma Amax 190 coagulation analyzer (Sigma Diagnostics, St Louis, Mo) and a Diagnostica Stago Starchrom kit (Diagnostica Stago). The World Health Organization international standard heparin (97/578) was used to construct the anti-factor Xa standard curve. Plasma samples containing 1, 2, and 4 U/mL heparin were diluted in pooled normal plasma to obtain anti-factor Xa levels within the linear section of the standard curve. The pooled normal plasma was tested to verify that the anti-factor Xa activity of the diluent was negligible.

Whole blood samples containing PPC heparin or Hepalean at 4 U/mL were obtained from the same 5 donors in syringes using the previously described technique. The concentration of heparin was chosen to approximate that usually achieved during CABG. The effect of the addition of 1, 3, or 5 μ g of protamine sulfate on the ACT of the heparin anticoagulated blood samples was determined. An equivalent volume of 0.15 M NaCl was used instead of protamine sulfate as a control.

Platelet-rich plasma (PRP) was obtained from the 5 donors by centrifuging 0.105 M sodium citrate anticoagulated blood samples at 200g for 8 minutes at room temperature. Platelet poor plasma was then obtained by centrifuging the remainder of the sample at 2000g for 10 minutes. The platelet count of the PRP was adjusted to $200 \times 10^9/\text{L}$ by dilution with platelet poor plasma. PPC heparin or Hepalean was added to the PRP at a final concentration of 4 U/mL. An equal volume of 0.15 M NaCl was substituted for heparin as a baseline control. Platelet aggregation studies were performed on PRP using a Chrono-log 4 channel platelet aggregometer (Chronolog, Havertown, Pa) according to the manufacturer's instructions. Collagen (final concentration, 2 $\mu\text{g}/\text{mL}$) and adenosine diphosphate (final concentration, 5 μM) were used as agonists, and 0.15 M NaCl was used as a negative control.

Phase III: Randomized Controlled Trial

Study design. We conducted a parallel-group, single-center, randomized, controlled trial. Patients undergoing CABG surgery with CPB were stratified by urgent and elective status in blocks of 2 and 4 and randomly assigned (1:1) by a computer-generated sequence to PPC heparin or Hepalean.

Study participants. All adult patients (age, ≥ 18 years) undergoing an isolated CABG procedure with CPB from June 1 to June 30, 2011, were approached for recruitment into the present study by our research nurse. Patients with a history of allergy or intolerance to heparin and those who had received unfractionated or low-molecular-weight heparin less than 24 hours before surgery were excluded. The size of the study was restricted by the limited supply of Hepalean.

Study procedures. The participants were randomized preoperatively to receive either PPC heparin or Hepalean during surgery. An unblinded research pharmacist prepared the blinded study drug kits. The participants, research nurse, surgeons, bedside nurses, outcome assessors, and statisticians were all unaware of the treatment allocation; 10,000 U of study heparin was used to prime the CPB machine, and the initial heparin dose given to each patient was 400 U/kg. If an appropriate ACT for CPB (defined for the present study as ≥ 480 seconds) was not achieved, additional heparin boluses were administered in increments of 5000 U until the target ACT was achieved. Once achieved, CPB was initiated, and surgery proceeded according to the usual practice. If needed, additional heparin (in increments of 5000 U) was given during the surgery according to usual practice. At the end of surgery, protamine was administered at an initial dosage of 1 mg/100 U of heparin given during the surgery. Additional doses of protamine were given as needed according to usual care. The postoperative patient care followed standard best practice guidelines. The patients were followed up postoperatively for 24 hours. The chest tube output, protamine and blood product administration, and occurrence of reoperation for bleeding or cardiac tamponade were recorded during this period.

Blood samples were collected into sodium citrate tubes at 4 points during the study: preoperatively, after administration of the initial dose of heparin, after 45 minutes of CPB, and after the initial protamine administration. The plasma was separated using a double-centrifugation technique, spinning the sample twice at 2000g for 10 minutes. The separated plasma was distributed into 5 aliquots at each point and frozen in a -80°C freezer within 2 hours of collection. The anti-factor Xa levels and aPTTs were obtained for each of the blood samples using the methods described.

Study outcomes. The primary outcomes were the ACT measured immediately after the initial weight-adjusted dose of heparin and the total weight-adjusted heparin dose administered during CPB.

The secondary outcomes were the ACT measured immediately after the initial weight-adjusted protamine dose at the end of the procedure; total protamine dose administered to neutralize heparin anticoagulation; blood loss, which was evaluated by the total chest tube output, number of units transfused, and the need for reoperation for bleeding or cardiac tamponade; and the results of the coagulation blood tests, including anti-factor Xa level and aPTT measured after administration of the initial dose of heparin, after 45 minutes of CPB, and after initial protamine administration.

Statistical analysis. All analyses were conducted in a blinded fashion. Continuous data are presented as the median and interquartile range or mean $\pm$ standard deviation, and categorical data are presented as numbers and proportions. The baseline, intraoperative, and postoperative data were compared using the Wilcoxon-Mann-Whitney rank sum test for nonparametric variables, except for age, weight, height, and body surface area, which were compared using Student *t* tests. Categorical variables were compared using Fisher's exact test. All *P* values were 2 sided.

RESULTS

Phase I: Audit

The results of our retrospective audit are listed in Table 1. Compared with the 10 historical controls who received Hepalean, the 10 PPC heparin recipients required a significantly greater initial dose of heparin by weight to achieve an ACT of 480 seconds (median, 440.7 U/kg; interquartile range, 421.8-523.8; vs median, 407.4 U/kg; interquartile range, 401.7-422.5; *P* = .021). The total protamine dose given in the operating room and intensive care unit was significantly greater in the PPC heparin group than in the Hepalean group (median, 425.0 mg; interquartile range, 400.0-500.0; vs median, 350.0 mg; interquartile range,

TABLE 1. Results of retrospective audit of 20 patients undergoing coronary artery bypass grafting

Variable	PPC heparin (n = 10)	Hepalean (n = 10)	<i>P</i> value*
Initial heparin dose (U)			.111
Median	40,000.0	36,500.0	
Interquartile range	36,500.0-48,750.0	28,500.0-41,500.0	
Initial heparin dose by weight (U/kg)			.021
Median	440.7	407.4	
Interquartile range	421.8-523.8	401.7-422.5	
Total perioperative heparin dose (U)			.495
Median	44,000.0	43,500.0	
Interquartile range	40,250.0-53,750.0	35,750.0-48,750.0	
Pre-CPB ACT (s)			.570
Median	609.5	659.5	
Interquartile range	545.8-659.5	555.5-928.3	
Total protamine dose (mg)			.038
Median	425.0	350.0	
Interquartile range	400.0-500.0	323.8-400.0	
Total protamine/100 U heparin (mg/100 U)			.082
Median	1.00	0.89	
Interquartile range	0.27-0.85	0.78-0.94	

ACT, Activated clotting time; CPB, cardiopulmonary bypass; PPC, Pharmaceutical Partners of Canada. *For Wilcoxon-Mann-Whitney rank-sum test comparing Hepalean and PPC groups.

323.8-400.0; *P* = .038). The protamine dose remained numerically greater after adjusting for the total amount of heparin administered, although the difference was no longer statistically significant (median, 1.00 mg/100 U heparin; interquartile range, 0.27-0.85; vs median, 0.89 mg/100 U heparin, interquartile range, 0.78-0.94; *P* = .082). No significant difference was found between the 2 groups in the bleeding outcomes.

Phase II: In Vitro Studies

According to the ACT, aPTT, and anti-factor Xa levels, PPC heparin and Hepalean had similar anticoagulant activity (Figures E1 and E2). Both brands of heparin were reversed with protamine in a similar manner (Figure E3). Platelet aggregation in response to collagen or ADP was similarly affected by PPC heparin and Hepalean (Figure 1).

Phase III: Randomized Controlled Trial

The patient flow is shown in Figure E4. A total of 60 patients were screened, and 23 patients agreed to participate. Of the 23 patients, 2 were not randomized; the first patient underwent emergency surgery during off hours and the second patient received 10,000 U of nonstudy heparin at CPB prime. The remaining 21 patients were randomized to PPC heparin (*n* = 11) or Hepalean (*n* = 10).

FIGURE 1. Molecular weight distribution of Hepalean and PPC Heparin. PPC, Pharmaceutical Partners of Canada.

The baseline and surgical characteristics are displayed in Table 2. The 2 groups were similar in all baseline characteristics, except for the preoperative platelet count, which was slightly greater in the PPC heparin group. All patients underwent isolated CABG surgery with CPB and received 2 to 7 grafts. All procedures used the left internal mammary artery. Two procedures, 1 in each group, also used the right internal mammary artery.

Primary outcomes. The data regarding heparin and protamine dosing are listed in Table 3. Despite similar median initial doses of PPC heparin and Hepalean (median, 404.5 and 400.5 U/kg, respectively), according to the study protocol, the patients in the PPC heparin group had a numerically lower initial ACT than the Hepalean group (median, 516.0 seconds; interquartile range, 481.0-633.0; vs median, 584.0 seconds; interquartile range, 520.0-629.0; $P = .418$).

The median total amount of heparin administered during CPB was significantly greater in the PPC heparin group than in the Hepalean group (median, 46,000.0 U; interquartile range, 39,500.0-60000.0; vs median, 34,500.0 U; interquartile range, 32,250.0-37,000.0; $P = .011$). This difference remained evident after adjustment for body weight (median, 572.9 U/kg; interquartile range, 443.0-659.7; vs 401.1 U/kg; interquartile range, 400.0-419.4; $P = .003$). Two patients, both in the PPC heparin group, failed to achieve the minimum target ACT of 480 seconds after the first dose of heparin and required additional doses. Also, 8 patients in the PPC heparin group and 2 in the Hepalean group ($P = .030$) required additional doses of heparin during surgery; an average of 211.3 U/kg in the PPC heparin group and 21.2 U/kg in the Hepalean group. The heparin dose by weight and the ACT values after the first dose of heparin and after subsequent heparin doses are shown in Figure E5.

Secondary outcomes. The results of coagulation testing are listed in Table 4.

The initial protamine dosage was determined from the total heparin dosage administered during surgery. No significant difference was found between the 2 groups in the ACTs achieved after the initial protamine dose (median, 112.0

TABLE 2. Baseline patient characteristics and details of surgery according to randomized treatment allocation

Characteristic	PPC heparin (n = 11)	Hepalean (n = 10)	P value
Gender (n)			—
Male	11	9	
Female	0	1	
Age (y)	63.9 ± 10.9	68.7 ± 14.2	.394*
Weight (kg)	89.3 ± 16.9	82.0 ± 16.0	.327*
Height (cm)	172.5 ± 7.1	170.0 ± 7.9	.446*
Body surface area (m ²)	2.06 ± 0.22	1.95 ± 0.22	.277*
Creatinine (μmol/L)			.217†
Median	91.0	78.0	
Interquartile range	79.0-110.0	73.5-91.0	
Hemoglobin (g/L)			.944†
Median	142.0	146.0	
Interquartile range	128.5-148.5	132.3-149.3	
Platelet count (×10 ⁹ /L)			.024†
Median	222.0	181.5	
Interquartile range	205.5-255.0	162.0-207.8	
International normalized ratio			.079†
Median	1.0	1.1	
Interquartile range	1.0-1.0	1.0-1.1	
PTT (s)			.287†
Median	33.0	31.5	
Interquartile range	32.0-35.0	31.0-33.8	
TCT 2 unit (s)			.337†
Median	23.0	24.0	
Interquartile range	22.0-24.0	23.8-24.3	
Surgery type (n)			—
Urgent	6	5	
Elective	5	5	
Grafts (n)			.454†
Median	4.0	4.0	
Interquartile range	3.0-4.5	3.0-4.0	
CPB duration (h)			.503†
Median	1.32	1.52	
Interquartile range	1.14-1.64	1.33-1.71	

Data presented as mean ± standard deviation, median and interquartile range, or numbers of patients. CPB, Cardiopulmonary bypass; PPC, Pharmaceutical Partners of Canada; TCT, thrombin clotting time; PTT, partial thromboplastin time. *Two-sided Student's *t*-test. †Wilcoxon-Mann-Whitney rank sum test.

seconds; interquartile range, 107.5-117.3; vs median, 113.5 seconds; interquartile range, 112.0-114.0; $P = .721$). However, the patients receiving PPC heparin had significantly greater aPTT values than those receiving Hepalean (median, 38.20 seconds; interquartile range, 36.25-50.90; vs median, 35.30 seconds; interquartile range, 34.0-41.03; $P = .049$). In addition, 9 patients in the PPC heparin group compared with 4 patients in the Hepalean group received additional protamine in the intensive care unit ($P = .081$). The decision to administer additional protamine to these patients was determined by clinical considerations (eg, continued oozing, excessive chest tube drainage) rather than the laboratory test results. The median total protamine dose was significantly greater in the

TABLE 3. Heparin and protamine dosing according to randomized treatment allocation

Dosing	PPC heparin (n = 11)	Hepalean (n = 10)	P value*
Heparin			
Initial dose/weight (U/kg)			.285
Median	404.5	400.5	
Interquartile range	401.4-413.3	400.0-402.8	
ACT after initial dose† (s)			.418
Median	516.0	584.0	
Interquartile range	481.0-633.0	520.0-629.0	
Total administered (U)			.011
Median	46,000.0	34,500.0	
Interquartile range	39,500.0-60,000.0	32,250.0-37,000.0	
Total dose by weight (U/kg)			.003
Median	572.9	401.1	
Interquartile range	443.0-659.7	400.0-419.4	
Protamine			
Initial dose (mg)			.222
Median	500.0	435.0	
Interquartile range	425.0-525.0	362.5-492.5	
ACT after initial dose (s)			.721
Median	112.0	113.5	
Interquartile range	107.5-117.3	112.0-114.0	
Total intraoperative dose (mg)			.110
Median	550.0	475.0	
Interquartile range	500.0-675.0	412.5-515.0	
Administered in ICU (mg)			.048
Median	50.0	0.0	
Interquartile range	50.0-50.0	0.0-50.0	
Total administered (mg)			.026
Median	600.0	500.0	
Interquartile range	550.0-700.0	425.0-542.5	
Total dose/100 U heparin (mg/100 U)			.132
Median	1.3	1.4	
Interquartile range	1.1-1.4	1.3-1.5	

ACT, Activated clotting time; ICU, intensive care unit; PPC, Pharmaceutical Partners of Canada. *Wilcoxon-Mann-Whitney rank sum test. †Two patients (one in each group) had an initial ACT > 1000-s limit of measurement; these values were considered to be 1000 for this analysis.

patients receiving PPC heparin than in those receiving Hepalean (median, 600.0 mg; interquartile range, 550.0-700.0; vs median, 500.0 mg; interquartile range, 425.0-542.5; $P = .026$).

Bleeding outcomes and the amount of blood products administered are listed in Table E1. A trend was seen toward increased postoperative bleeding in the group receiving PPC heparin compared with the Hepalean group, as determined by the total chest tube output (median, 830.0; interquartile range, 425.0-1135.0; vs median, 702.5; interquartile range, 550.0-742.5; $P = .3240$). The bleeding

TABLE 4. Results of coagulation testing according to randomized treatment allocation

Sample	PPC heparin (n = 11)	Hepalean (n = 10)	P value*
UFH anti-factor Xa (U/mL)			
Baseline			.639
Median	0.05	0.05	
Interquartile range	0.05-0.19	0.05-0.10	
After initial heparin			.698
Median	8.10	7.65	
Interquartile range	7.05-8.78	6.68-8.25	
After 45 min on CPB			.098
Median	7.50	6.47	
Interquartile range	6.53-8.70	5.66-6.86	
After initial protamine			.246
Median	0.09	0.06	
Interquartile range	0.06-0.18	0.05-0.11	
PTT (s)†			
Baseline			.472
Median	37.60	36.95	
Interquartile range	35.60-40.70	34.95-38.85	
After initial heparin			—
Median	300.00	300.00	
Interquartile range	300.00-300.00	300.00-300.00	
After 45 min on CPB			—
Median	300.00	300.00	
Interquartile range	300.00-300.00	300.00-300.00	
After initial protamine			.049
Median	38.20	35.30	
Interquartile range	36.25-50.90	34.0-41.03	
AT (U/mL)			
Baseline			.910
Median	0.97	0.90	
Interquartile range	0.79-1.04	0.84-1.03	
After initial heparin			.698
Median	1.08	1.00	
Interquartile range	0.94-1.15	0.93-1.15	

AT, Antithrombin; CPB, cardiopulmonary bypass; PPC, Pharmaceutical Partners of Canada; PTT, partial thromboplastin time; UFH, unfractionated heparin. *Wilcoxon-Mann-Whitney rank sum test. †Maximum measureable PTT, 300 s.

indexes were similar in the 2 groups, and no patients returned to the operating room for re-exploration for bleeding.

DISCUSSION

The results of our retrospective chart review suggested an increase in heparin requirements for PPC heparin compared with Hepalean, but our in vitro studies provided no evidence of differences in the potency of the 2 heparins. Therefore, we performed a randomized trial, which, although restricted in size by the lack of availability of Hepalean, showed that in patients undergoing CABG surgery with CPB, those patients receiving PPC heparin had numerically lower ACT levels after initial heparin administration, required significantly greater doses of heparin during surgery, and displayed a trend toward increased postoperative bleeding. These results have confirmed the anecdotal observations

made by the cardiovascular surgery team and the results of our retrospective audit.

The small sample size of our study was dictated by the lack of Hepalean. Despite this limitation, a significant and substantial difference was found in the total dose of heparin required to reach and maintain the target ACT in the PPC heparin group compared with the Hepalean group. Also, correspondingly, the total dose of protamine required to reverse the anticoagulant effect was also greater in the PPC heparin group.

Our results are consistent with previous reports of differences in the anticoagulant response when switching from 1 heparin to another in patients undergoing bypass surgery. In 2005, the London Health Sciences Centre in London, Ontario, Canada noted in a retrospective study that when they replaced Hepalean with PPC heparin, the number of blood transfusions were greater in the patients who received PPC heparin (51.3% vs 47.7%; $P = .09$) and that reoperation for bleeding occurred more frequently (4.3% vs 2.7%; $P = .057$). After switching back to Hepalean, a reversal of these trends occurred.⁸ Prompted by the concerns raised by other centers about the potential for increased bleeding with PPC heparin, the investigators at the Hospital for Sick Children (Toronto, Ontario, Canada) undertook a retrospective cohort study involving 903 pediatric cardiac surgery patients.^{8,9} Compared with Hepalean, these investigators found that PPC heparin was associated with increased red blood cell transfusions in the first 24 hours and an increased risk of hemorrhagic complications (odds ratio, 7.0; 95% confidence interval, 2.0-24.2) and early reoperation (odds ratio, 5.3; 95% confidence interval, 1.0-28.0).

Our randomized study mandated that both groups received the same initial dose of heparin, and our primary outcomes were the ACT measured immediately after the initial weight-adjusted dose of heparin and the total weight-adjusted heparin dose administered during CPB. A nonsignificant trend was seen for the initial postheparin ACT to be lower in the PPC heparin group than in the Hepalean group. Furthermore, the ACT of many patients decreased during surgery, leading to the administration of additional doses of heparin to maintain the ACT within the target range. In the PPC heparin group, an additional 211.3 U/kg was required during the procedure, but in the Hepalean group, only an additional 21.2 U/kg was required. The reason for this marked difference in heparin dosage requirements to maintain the ACT in the target range remains to be determined but might be explained by differences in clearance or protein binding of the 2 heparin preparations. The subsequent trend toward increased bleeding and a significant increase in protamine requirement is consistent with the findings of the observational studies reviewed and the results of our retrospective chart review and can be explained by the heparin rebound resulting from the greater heparin

dose given to the PPC heparin group. Heparin rebound is well recognized after CPB and occurs as a result of the re-entry of heparin from the extravascular to the intravascular space and the release of protein-bound heparin after routine heparin neutralization by protamine. Because the heparin-protamine complex is rapidly cleared in the urine, the heparin that re-enters the circulation has the potential to cause postoperative bleeding.^{10,11} In our study, despite the initial postprocedure protamine dose used for neutralization, the aPTT and subsequent protamine doses were significantly greater in the PPC heparin group than in the Hepalean group. Both these observations can be plausibly explained by greater heparin rebound in the PPC heparin group. Studies are currently underway to explore the mechanism of the pharmacologic differences of the 2 heparins.

Within the medical community, it has long been assumed that all unfractionated heparin preparations are the same. PPC heparin and Hepalean are both marketed under the U.S. Pharmacopeia unfractionated heparin monograph standards. Under this regulation, they are required to have similar efficacy, potency, and reversibility. However, the findings of previous work, combined with our results, suggest that important clinical difference exist between PPC heparin and Hepalean that cannot be predicted with the usual in vitro tests.

Our results indicate that heparin preparations might not interchangeable despite identical anticoagulant profiles in vitro. We therefore suggest that if a change in heparins is made for CPB surgery, the anticoagulant profile of the new heparin should be compared with the brand in use by performing a chart review similar to the one we performed. If differences are found, our studies could provide a template for their additional investigation.

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FIGURE E1. Activated clotting times (ACTs) for various concentrations of Hepalean and PPC heparin. *PPC*, Pharmaceutical Partners of Canada.

FIGURE E3. In vitro study of protamine reversal of Hepalean and PPC heparin. *PPC*, Pharmaceutical Partners of Canada; *ACT*, activated clotting time.

FIGURE E2. Anti-factor Xa activity for various concentrations of Hepalean and PPC heparin. *PPC*, Pharmaceutical Partners of Canada.

FIGURE E4. Consort flow diagram. PPC, Pharmaceutical Partners of Canada.

FIGURE E5. Heparin dose by weight at each dose and resulting activated clotting time (ACT). PPC, Pharmaceutical Partners of Canada; CPB, cardiopulmonary bypass.

TABLE E1. Blood product use and bleeding outcomes according to randomized treatment allocation

Blood product use and bleeding outcomes	PPC heparin (n = 11)	Hepalean (n = 10)	P value*
Intraoperative blood products			
Intraoperative PRBCs (U)			.801
Median	0.0	0.0	
Interquartile range	0.0-1.5	0.0-1.5	
Intraoperative FFP (mL)			.189
Median	0.0	0.0	
Interquartile range	0.0-0.0	0.0-0.0	
Intraoperative platelets (mL)			.189
Median	0.0	0.0	
Interquartile range	0.0-0.0	0.0-0.0	
Intraoperative cryoprecipitate (mL)			—
Median	0.0	0.0	
Interquartile range	0.0-0.0	0.0-0.0	
Postoperative blood products			
Postoperative PRBCs (U)			.304
Median	0.0	0.0	
Interquartile range	0.0-0.5	0.0-0.0	
Postoperative FFP (mL)			.146
Median	0.0	0.0	
Interquartile range	0.0-0.0	0.0-0.0	
Postoperative platelets (mL)			.968
Median	0.0	0.0	
Interquartile range	0.0-335.0	0.0-340.0	
Postoperative cryoprecipitate (mL)			—
Median	0.0	0.0	
Interquartile range	0.0-0.0	0.0-0.0	
Bleeding outcomes			
Total chest tube output (mL)			.324
Median	830.0	702.5	
Interquartile range	425.0-1135.0	550.0-742.5	
24-h Postoperative hemoglobin (g/L)			.397
Median	96.0	87.5	
Interquartile range	86.0-100.5	82.3-96.3	
Total change in hemoglobin (g/L)			.672
Median	−43.0	−46.0	
Interquartile range	−37.0 to −47.5	−24.5 to −60.5	

FFP, Fresh frozen plasma; PRBCs, packed red blood cells; PPC, Pharmaceutical Partners of Canada. *Wilcoxon-Mann-Whitney rank sum test.