Aprotinin Shows Both Hemostatic and Antithrombotic Effects During Off-Pump Coronary Artery Bypass Grafting

Robert S. Poston, MD, Charles White, MD, Junyan Gu, MD, PhD, James Brown, MD, James Gammie, MD, Richard N. Pierson, MD, Andrew Lee, Ingrid Connerney, RN, DrPH, Thrity Avari, MS, Robert Christenson, PhD, Udaya Tandry, PhD, and Bartley P. Griffith, MD

Departments of Surgery, Radiology, Clinical Effectiveness, and Pathology, University of Maryland School of Medicine, Baltimore, Maryland; Chronolog Corp, Havertown, Pennsylvania; and Sinai Center for Thrombosis Research, Baltimore, Maryland

Background. **Hemostatic drugs are widely thought to be unnecessary and potentially detrimental in off-pump coronary artery bypass graft surgery (OPCABG), despite well-established use in on-pump surgery. In a randomized, prospective OPCABG trial, we assessed efficacy and safety of aprotinin through a comprehensive assessment of graft patency and hematologic function.**

Methods. **Sixty patients were randomly assigned to full-dose aprotinin or placebo. Heparin was titrated to a kaolin-based activated clotting time of greater than 300 seconds. Exclusionary criteria included creatinine greater than 2 mg/dL, conversion to on-pump CABG, and preoperative GPIIb/IIIa inhibition. Hematologic assessments were obtained preoperatively, at the end of surgery, and on days 1 and 3: mean platelet volume, thrombin generation (prothrombin fragment 1.2 assay), and aspirin resistance using a modified thrombelastography, whole blood aggregometry, 11-dehydro-thromboxane B_2 levels, and flow cytometry. Thrombotic events were defined as postoperative myocardial infarction by electrocardiography or elevated troponin I, clinical stroke by examination and head computed tomography, and bypass graft failure by multichannel computed tomography angiography on day 5.**

Results. **Aprotinin was associated with a significant reduction in intraoperative and postoperative blood loss compared with placebo but had no effect on transfusion rates. Patients treated with aprotinin had significantly fewer thrombotic events (3% versus 23%, $p < 0.05$, Fisher's exact test) and less postoperative aspirin resistance (20% versus 46%, respectively, $p < 0.05$, Fisher's exact test). Postoperative prothrombin fragment 1.2 level was reduced by aprotinin use.**

Conclusions. **Aprotinin reduced perioperative bleeding after OPCABG. Preserved aspirin sensitivity in the aprotinin group may explain the observed reduction in thrombotic events and might be related to the suppression of perioperative and transmyocardial thrombin formation.**

(Ann Thorac Surg 2006;81:104–11)

Aprotinin (Trasylol; Bayer Pharmaceuticals Corp, West Haven, CT) reduces the need for blood transfusions and preserves platelet function in patients undergoing cardiopulmonary bypass (CBP) [1, 2]. Similarly, off-pump coronary artery bypass graft surgery (OPCABG) is also associated with reduced transfusions and improved hemostatic function as compared with on-pump CABG [3–5]. Aprotinin use has been widely avoided during OPCABG based on the concern of provoking hypercoagulability [6]. The lack of an established in vitro assay of hypercoagulability hinders critical evaluation of OPCABG, aprotinin therapy, and the risk of de novo thrombotic events. Despite the known benefit of aspirin therapy for early saphenous vein graft (SVG) failure [7], insufficient inhibition of in vitro platelet function, or acetylsalicylic acid (aspirin) resistance (ASA-R), is seen in nearly 50% of patients on an aspirin regimen after cardiac surgery [8, 9]. As ASA-R is associated with an increased risk of SVG failure after OPCABG [8], this endpoint could provide a surrogate for monitoring for hypercoagulability in this setting.

A recent randomized trial showed a favorable effect of aprotinin on bleeding after OPCABG [10]. Unfortunately, anatomic SVG patency was not assessed in this cohort, as early graft failure, often clinically silent [2], can have long-term consequences. In the current study, postoper-

Accepted for publication May 10, 2005.

Presented at the Forty-first Annual Meeting of The Society of Thoracic Surgeons, Tampa, FL, Jan 24–26, 2005.

Address correspondence to Dr Poston, Division of Cardiac Surgery, N4W94 22 S Greene St, Baltimore, MD 21201; e-mail: rposton@smail.umaryland.edu.

Ms Avari discloses a financial relationship with Chronolog, Corp.

0003-4975/06/$32.00
doi:10.1016/j.athoracsur.2005.05.085

Abbrevations and Acronyms

ASA-R = aspirin resistance
AUC = area under the curve
CABG = coronary artery bypass grafting
CT = computed tomography
F1.2 = prothrombin fragment 1.2
KIU = kallikrein inhibiting units
MA = maximum amplitude
OPCABG = off-pump coronary artery bypass grafting
SVG = saphenous vein graft

ative computed tomography (CT) imaging of bypass SVG patency and serial assessment of hematologic function were performed. We hypothesized that this comprehensive study design would allow us to better assess any possible discrete consequences of aprotinin therapy in OPCABG.

Patients and Methods

Patient Selection and Enrollment

A randomized, double-blind, placebo-controlled, prospective study of aprotinin was conducted in 60 OPCABG patients. The protocol was approved by the Institutional Review Board (protocol #0902312) and Food and Drug Administration (Investigational New Drug application #67,890), and each patient provided informed consent. Exclusion criteria included nonambulatory patients, chronic renal insufficiency (creatinine $>$ 2.0 mg/dL), active hepatitis or cirrhosis, allergy to radiographic contrast media, prior exposure to aprotinin, and use of GPIIb/IIIa inhibitors or clopidogrel within 3 days of surgery. Randomization was based on permuted blocks of size 4 to preserve approximate balance between groups based on anticipated enrollment.

Treatments

A modified full-dose regimen was used: 10,000 kallikrein inhibiting units (KIU) intravenous test dose (or saline placebo), 2 million KIU aprotinin through a central line before sternotomy, and 500,000 KIU/h until the end of the operation. Study drug or saline placebo was delivered to operating room in unlabeled bottles to maintain the blinding. Heparin dose was calculated by protamine titration using the HMS heparin assay (0.0 to 2.5 mg/kg [cat. no. 30407; Medtronic, Minneapolis, Minnesota]) to maintain levels greater than 2 μg/mL and a kaolin-based activated clotting time greater than 300 seconds. The heparin effect was partially reversed by administering half the dose of protamine calculated by the HMS device. The algorithm for intraoperative and postoperative blood product transfusions was based on thrombelastography analysis, as described [11]. All patients received preoperative and postoperative aspirin (325 mg orally each morning and within 6 hours after intensive care unit arrival) as the sole platelet inhibitor. Compliance was documented by patient questioning and examining medication administration records. Glucose levels were maintained at less than 150 mg/dL in the intensive care unit using insulin infusion. Given an association with ASA-R and direct effects on thromboxane production, nonsteroidal anti-inflammatory drugs were prohibited until hospital discharge.

Surgery and Perioperative Management

Four surgeons experienced in OPCABG enrolled patients. Internal mammary conduits were used in all patients. Saphenous veins were harvested using an endoscopic or open approach and stored in heparinized blood until use. The distal anastomosis was facilitated with suction-based devices (Medtronic). Enrolled patients converted to on-pump CABG (n = 9) were excluded from analysis. The volume retrieved intraoperatively by a cell salvage device (Haemonetics, Braintree, Massachusetts) and amount of postoperative shed blood after 24 hours was measured. Chest drain red blood cell volume was calculated at 24 hours by the hematocrit of contents in the chest drainage system (Atrium Medical Corp, Hudson, New Hampshire) multiplied by total volume [12]. Patients were excluded from the study if reoperation for bleeding due to a "surgical" source was required (n = 1). Intensive care unit and hospital discharge criteria followed closely monitored protocols.

Saphenous Vein Graft Quality Assurance

Intraoperatively, flow and pulsatility index (maximum-minimum/mean blood flow) were assessed in each SVG using a transit-time techniques (Medistim, Minneapolis, Minnesota). Saphenous vein grafts with flow that remained less than 10 cc/min and pulsatility index greater than 5 despite revision were excluded from analysis (n = 2). Endothelial integrity was analyzed in surplus portions of each SVG. A segment was frozen in cutting compound, and 5-μm sections were stained with monoclonal antibody against the endothelial marker, CD31 (R&D System, Minneapolis, MN). Percentage endothelial integrity was calculated by image analysis software (Bioquant Nova Prime; BIOQUANT Image Analysis Corp, Nashville, TN).

Assays for Coagulation

All coagulation and platelet function testing were performed on citrated blood samples drawn at four time points: before skin incision, postoperatively after protamine, and the mornings of postoperative days 1 and 3. International normalized ratio, partial thromboplastin time, fibrinogen, and quantitative d-dimer were performed by the clinical laboratory. Prothrombin fragment F1.2, released when thrombin is generated from prothrombin, was assessed in platelet-poor plasma using a commercially available enzyme-linked immunosorbent assay kit (Enzygnost F1.2 Micro; Dade-Behring, Deerfield, IL). The percent perioperative difference was calculated as follows: (preoperative F1.2 – postoperative F1.2) / preoperative F1.2 $\times$ 100. In a subset of patients (n = 20), coronary sinus samples were