Abstract

Extracorporeal circulation is associated with extensive alterations in platelet number and function. In certain individuals these adverse platelet changes are exacerbated by heparin. ZK36374 (ZK), a stable analogue of prostacyclin, reversibly inhibits all phases of platelet activation. A patient was presented with heparin induced platelet activation and paradoxical thrombosis who required cardiopulmonary bypass to correct severe tricuspid stenosis and ASD. We employed full heparinization and ZK to prevent surface and heparin induced thrombocytopenia. Prior to heparinization, a constant infusion of ZK was begun at 10 nanograms/kilogram/minute, tapered to 2 nanograms/kilogram/minute during bypass, and maintained until heparin was reversed with protamine. Prior to bypass, platelets demonstrated 50% reactivity and 2% reactivity when challenged by ADP and epinephrine, respectively, indicating inhibition of platelet function by ZK. Two hours after arrival in recovery 92% prebypass circulating platelet count, 90% preservation of platelet reactivity to ADP, and a template bleeding time near control were achieved. ZK was used safely to prevent both heparin and surface induced platelet changes during cardiopulmonary bypass in this patient.

Introduction

Contact between blood and the synthetic surfaces of an extracorporeal circuit results in profound alterations in platelet number and function. Additionally, heparin induced thrombocytopenia has been reported and may be associated with bleeding and/or thrombosis. These adverse platelet changes have been associated with prolonged postoperative bleeding times and extensive blood loss. Investigators have indicated that while circulation of blood through an extracorporeal circuit affects several hemostatic processes, "acquired platelet dysfunction appears to be the most significant abnormality."

In vitro simulated extracorporeal circulation with a membrane oxygenator and human blood shows a decline to approximately 20% of the initial levels of platelets within two minutes but then a slow increase during the next several hours. In vitro circuits containing a bubble oxygenator rather than a membrane oxygenator also showed a decrease in platelet levels but did so more slowly. In bubble oxygenators there appears to be no return of platelets to circulation as occurs with a membrane oxygenator with time. Experiments to characterize platelet behavior in the extracorporeal circuit have established the site of platelet loss as the membrane oxygenator. The surface area of a spiral coil membrane oxygenator, more specifically the blood volume surface area ratio, is known to influence platelet surface interaction. Using an in vitro extracorporeal
circuit which consists of a spiral coil membrane oxygenator, standard silicone tubing, and a roller pump, a new platelet inhibitor, a prostacycline analogue, was investigated.

Experiments were performed using ZK36374 (ZK), to effectively preserve circulating platelet count, prevent platelet granular release and preserve platelet functional and morphological integrity during in vitro extracorporeal circulation. The in vitro recirculation trials indicated that ZK maintained platelets counts of approximately 80 to 85% of the initial levels while controls fell to 30% of the initial level. Electron microscopy demonstrated the platelet structure also appeared to be preserved during recirculation studies. The test used to determine functional integrity of the platelets was measurement of plasma levels of platelet factor IV, a platelet specific release product, and ZK prevented release of platelet granule contents. Finally, platelet aggregation studies indicated that platelets recirculated in the extracorporeal circuit and protected by ZK, maintained their ability to react to both ADP and epinephrine after discontinuation of the infusion of ZK. A study involving the use of ZK in humans undergoing cardiopulmonary bypass was the next step in the evaluation of this prostacyclin analogue.

**Case Report**

During the course of the clinical trials of ZK, a patient presented with severe tricuspid stenosis and atrial septal defect. The patient was a 41-year-old woman presented with a short history of chest pain, some dizziness, and headaches. Two months prior, she had been phlebotomized having a hemoglobin of 19.2 grams and a hematocrit of 56%. Her platelet count was 121,000. In the past, the patient had been a three pack per day (60 pack years) smoker. Part of the problem was thought to have been polycythemia, secondary to heavy smoking. She had facial weakness and weakness of the right upper extremity.

After extensive work-up, the impression was hypochromic anemia, secondary to phlebotomy, cerebralvascular disease, possible pulmonary hypertension, and possible intra or exocardiac shunt. The patient was placed on heparin at which time she developed a heparin associated thrombocytopenia and right arm thrombosis. Platelet counts increased off heparin then decreased again to 50,000 off heparin in the absence of heparin associated antibodies.

The patient was taken to the cath lab on two occasions. The first, for a right heart catherization for hemodynamics and right atrial and ventricular angiography. This showed a mean RA pressure of 9, with a mean LA of 6, and a large right to left shunt through a patent foramen ovale. The right ventricle (RV) appeared to contract normally. The tricuspid valve bowed slightly into the RV before opening. Also, a question of a mass or filling defect on the atrial surface of the tricuspid leaflets was noted. Oxygen saturation showed a step-up at the left atrium with pulmonary venous blood being 97% saturated. The PA pressures were low normal. The shunt calculated was a systemic flow of 4.8 liters per minute with a pulmonary flow of 3.0 liters per minute and a shunt flow in the right to left direction of 1.8 liters per minute. The QP/QS was 62%. The second cath was performed using a balloon occlusion of the patent foramen ovale. The findings were consistent with right ventricular inflow obstruction with the balloon occlusion of the patent foramen ovale. The RA pressures were: A-wave = 18, and V-wave = 10. The right ventricular pressures at the same time were 17/5 with a 15 mm gradient between the RA and RV during atrial systole. The balloon occlusion of the patent foramen ovale resulted in a left atrial blood saturation of 97%.

Admitted for surgery, the patient persistently had platelet counts below 70,000 while on steroids. Surgery had to be postponed. With platelet transfusions and weaning of the steroids, the platelet count was elevated to 165,000.

Because of the proven ability of ZK to protect platelets in vitro and the encouraging results from the clinical trials, it was decided that its use in the clinical setting may be appropriate for a patient with a predisposition to platelet aggregation. Special permission was granted for the use of ZK on this patient.

The patient was brought to the operating room for correction of the tricuspid stenosis and ASD. The baseline ACT was 164 seconds and the platelet count was 100,000. Prior to heparinization, a constant infusion of ZK was begun at 10 nanograms/kilogram/minute, and was tapered to 5 nanograms/kilogram/minute after five minutes of bypass and to 2 nanograms/kilogram/minute during closure of the atriotomy. This concentration was maintained until heparin was reversed with protamine. The patient was fully heparinized (300 units/kilogram). The post heparin ACT was greater than 1000 seconds. Prior to bypass the platelets demonstrated a 50% reactivity and a 2%
reactivity when challenged with ADP and epinephrine, respectively, indicating that the platelets were inhibited by ZK.

The cardiopulmonary bypass circuit included: a spiral coil membrane oxygenator, a centrifugal pump, standard medical grade PVC tubing, a 40 micron arterial filter, and a filtered cardiomyotomy reservoir. The system was primed with 2000 ml Normosol R, 50 Grams of 25% Albumin and 5000 units Sodium Heparin. Aortic Arch cannulation was accomplished with a 24 Fr flexible Aortic cannula. The superior and inferior vena cava were cannulated with 32 Fr venous catheters. Myocardial protection consisted of moderate systemic hypothermia (26 degrees Centigrade), cold topical solution, and crystalloid cardioplegia (30 mEq KCl/L).

Cardiopulmonary bypass was initiated. Perfusion flows ranged from 2.2 to 2.4 liters per minute per square meter of body surface area, and pressures, blood gases, and electrolytes were all within normal limits. The platelet count fell to 60,000. No heparin was added during CPB and all ACTs were greater than 1,000 seconds.

A moderately sized patent foramen ovale was present. The tricuspid valve was quite stenotic with a fair amount of clot on the valve. The clot appeared not to be fresh but several weeks old. The tricuspid valve appeared to be congenitally stenotic. The valve was replaced and the ASD closed. Cardiopulmonary bypass was discontinued with ease. Protamine (250 mg) was given and the patient was hemostatically stable. The aortic cross-clamp time was 55 minutes and the bypass time was 78 minutes. The post protamine ACT was 164 seconds. Two hours after arrival in recovery 92% of the pre-bypass circulating platelet count and 90% preservation of platelet activity to ADP, and a template bleeding time near control was achieved. One week after operation the patient was discharged from the hospital. One month after discharge the patient was doing well and was intending to return to work.

Discussion

Heparin induced thrombocytopenia is well reported in the medical literature. Cooper and Reed from Texas Heart have also reported in the perfusion literature a case study of heparin induced thrombocytopenia in a patient receiving a coronary angioplasty. The percentage of patients with heparin induced thrombocytopenia is low but significant.

A decision was made to use a platelet inhibitor, ZK, in conjunction with the routine heparin anticoagulation protocol for cardiac surgery. The surgery was performed for a tricuspid valve replacement and closure of the ASD. The platelet count and function returned to pre-surgery levels despite full heparinization and cardiopulmonary bypass.

The ability of a drug such as ZK, to prevent the surface induced platelet changes in extracorporeal circulation, as well as heparin induced thrombocytopenia, will be invaluable. Continued in vivo evaluation of ZK is planned using extracorporeal circuits with membrane and bubble oxygenators. We expect that ZK will become a valuable tool in an effort to help control another variable in such an invasive procedure as cardiopulmonary bypass.

References