Investigating a Chandler Loop System for Thrombogenicity Testing of Biomaterials

Jessica Yau, Megan Jamiolkowski, Richard Malinauskas, Qijin Lu
US Food and Drug Administration, University of Maryland College Park

Statement of Purpose: Thrombosis related complications of blood-contacting cardiovascular devices remain major concerns within the medical community. A robust in vitro method to evaluate biomaterial thrombogenicity would be a useful tool in improving the design and safety evaluation of medical devices, while reducing the need for expensive animal studies. The Chandler loop model, a rotating blood flow loop partially filled with air, is a commonly used in vitro method for testing biomaterial hemocompatibility. However, little is known about the influence of various test parameters on the effectiveness of this technique. The aim of this study was to optimize the Chandler loop test protocol for biomaterial thrombogenicity evaluation by determining the effects of anticoagulant concentration, air volume, and blood flow rate on the test results.

Methods: Two materials with different thrombosis potentials were tested, a thromboresistant polytetrafluoroethylene (PTFE) cord (0.1" Diameter) and a thrombogenic latex tube (1/8" OD, lumen sealed with a PTFE plug). Polyvinyl chloride (PVC) tubing (45 cm long, 1/4" ID, 14 ml total loop volume) was used to form the Chandler loop by connecting the two ends with a silicon sleeve. Test samples (12 cm long) were inserted into two separate Chandler loops through a small cut in the PVC tubing and sealed with Parafilm. Porcine blood from live donors was drawn into Anticoagulant Citrate Dextrose Solution A (ACDA) for overnight shipping and used within 36 hours of blood drawing. Immediately before starting the loop tests, the blood was re-calcified (final calcium chloride concentration 13 mM) and heparinized. The heparin concentrations used for each blood donor were determined using a static latex pre-test, in which latex tubes were incubated in re-calcified blood with a series of heparin concentrations (3.0 to 7.0 U/ml, 0.5 U/ml increments) for 20 min at 37°C. The minimum heparin concentration that resulted in a thrombus surface coverage <10% was selected as the baseline concentration for the Chandler loop testing. Two lower heparin concentrations (0.25 and 0.5 U/ml below the baseline concentration) were also used to investigate the effects of heparin concentration. Different air volumes (1, 2, 3.5, and 7 mL) and blood flow rates (200 and 400 mL/min) were also evaluated. The loops were rotated for 1hour at 37°C. Any thrombus that deposited on the test materials was photographed and weighed. Blood platelet count and plasma free hemoglobin concentration were measured before and after the 1 hour test period.

Results: Per the static latex pre-test, the baseline heparin concentrations selected for the Chandler loop ranged from 5 to 6.5 U/ml, depending on the reactivity of the donor blood. Using the baseline heparin doses, the Chandler loop test was able to distinguish between the latex and the PTFE materials in both thrombus weight and platelet count reduction. A reduction in the heparin concentration by 0.25 and 0.5 U/ml resulted in increased thrombus weights and greater reductions in platelet count (Fig 1). These results suggest that the test is sensitive to the level of heparinization, and that the optimal heparin concentration is blood donor dependent. On the other hand, tests using different air volumes and blood flow rates did not substantially affect thrombus formation or platelet count reduction (Fig. 2). Hemolysis caused by the Chandler loop was minimal (<10 mg/dL change in plasma free hemoglobin concentration).

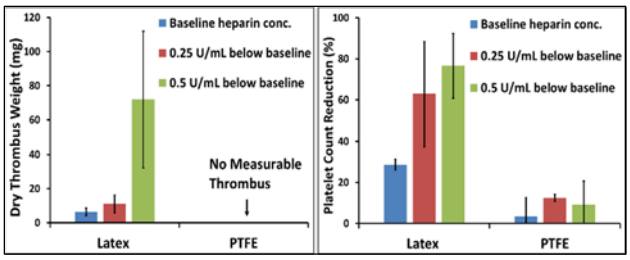

Fig. 1. Effects of heparin concentration. Test conditions: Air volume 1 ml, flow rate 200 ml/min. (N=3 donors).

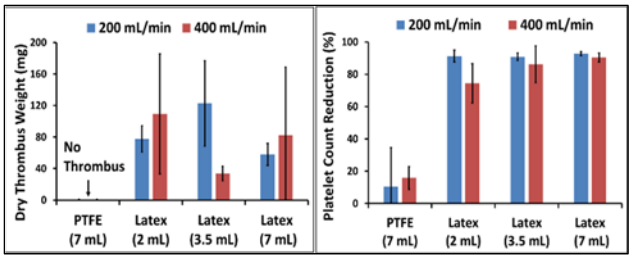

Fig. 2. Effects of flow rate (200 and 400 ml/min) and air volume (2, 3.5, 7 ml). Heparin concentration: 0.5 U/ml below baseline. (N=3 donors)

Conclusions: Preliminary results from this study demonstrate that the Chandler loop model can be used to effectively compare the thrombogenicity of biomaterials, and the sensitivity of the test can be increased by using donor-specific heparin concentrations. Future studies employing more test materials are needed to fully validate this method.