Effectiveness of a Kaolin-Based Hemostatic Dressing* in an Anticoagulated Porcine Model

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ABSTRACT:

Thousands of patients receive anticoagulation therapy such as warfarin and clopidogrel. Warfarin works as a vitamin K agonist[1] whereas clopidogrel inhibits an ADP receptor on platelets which prevents aggregation.[2] Patients needing common surgical procedures while on these drugs are at an increased risk for hemorrhagic complication.[3] Kaolin activates factor XII or release of platelet factor 3 causing clot formation which accelerates hemostasis.[4] Because kaolin activates the clotting cascade, it is hypothesized that a kaolin-based hemostatic dressing will be effective in patients receiving either warfarin or clopidogrel.[4]

The in vivo bleeding control response of the kaolin-based hemostat compared to standard gauze was tested in an anticoagulated porcine model. For a minimum of 5 days preoperatively, animals were dosed daily either with 75 mg/day of clopidogrel or with 5-7.5 mg/day of warfarin until an INR > 2.5 was reached. Liver, spleen, and mesenteric root lacerations were treated with the kaolin dressing and standard gauze. Success was defined as no visible bleeding after 5 minutes of manual compression. The kaolin-based dressing was found to be more effective than standard gauze (chi-square test $p<0.05$, 95% confidence) at controlling bleeding following abdominal injuries in anticoagulated swine.

INTRODUCTION:

Warfarin is the most commonly prescribed oral anticoagulant in the United States. Over 3 million Americans take Coumadin or other generic forms of warfarin each year.[5] A vitamin K agonist, Warfarin affects the intrinsic coagulation pathway. Plavix (Clopidogrel) is another commonly used antiplatelet agent. It is estimated that between 2.5 and 3 million prescriptions were written for Plavix each month in the US in 2010.[6] Plavix, an antiplatelet agent, affects the extrinsic coagulation pathway.

The use of anticoagulants and antiplatelet agents increases each year. The probability of a patient needing emergency surgery while taking one of these agents is also higher. Complications of hemorrhage can be severe and will likely be higher in an anticoagulated patient.[7] It can often be either difficult or time consuming to reverse the effect of these drugs when necessary.[3] There is a need to have available novel means to control bleeding in patients who are treated with these drugs.

Kaolin has been known for decades to promote clotting via the intrinsic pathway. Contact activation of the coagulation cascade is mainly based on the unique property of Factor XII, a circulating serine-protease produced by the liver, to undergo "autoactivation" and a change in shape when exposed to negatively charged artificial or biological surfaces.[8,9] Factor XII is converted to factor XIIa and the cascade proceeds to clot formation. Kaolin has been shown to cause aggregated platelets to release platelet factor 3.[4,10,11,12] Because kaolin activates the coagulation cascade, it was hypothesized that a kaolin-based dressing (QC)* would be effective in anticoagulated animals treated with Coumadin or Plavix.

MATERIALS AND METHODS:

Adult Yorkshire swine weighing between 45–60 kg were used in this experiment. All animals in this study were used according to a protocol approved by Institutional Animal Care and Use Committee.

Five animals received Coumadin (Bristol-Myers Squibb Princeton, New Jersey 08543 USA) via regular food. Dosage was adjusted according to human use and pertinent literature. A loading dose of 7.5 mg/day were given for 3 days followed by 5 mg/day for the remainder of the study. An INR > 2.5 was the goal and PT testing was repeated at regular intervals until the goal was reached. As soon as INR of 2.5 or greater was reached, surgery was scheduled.

An additional 5 animals were treated with Plavix (Bristol-Myers Squibb/Sanofi Pharmaceuticals Partnership, Bridgewater, NJ, USA) also according to human dosing and pertinent literature. The animals were treated with a dose of 75 mg/day. As indicated above, Plavix was administered mixed with food or according to Veterinary recommendation. As per literature (in both humans and pigs), Plavix-treated animals reached steady activity in altering platelet aggregation in 5 days from starting the treatment. Once this steady state of platelet dysfunction was reached, animals were taken to surgery.

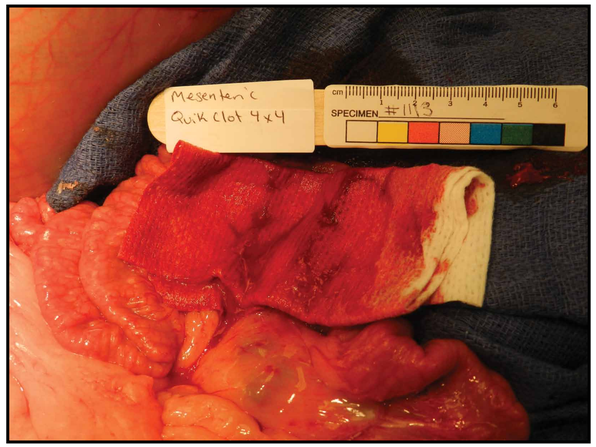

Figure 1: 4x4 QC Dressing Applied to Mesentery

The animals were fasted overnight with water allowed ad libitum before surgery. They were then sedated using cocktail of telazol-ketamine-xylazine (TKX), intubated, and placed on mechanical ventilation with 100 % Oxygen. Anesthesia was maintained throughout the surgery at 1-2% isoflurane. Ventilatory parameters were adjusted to attain the end-tidal pCO_2 of 40 mm Hg ± 5 mm Hg. Intravenous Lactated Ringer's solution via 20 or 24 gauze auricular catheter at 5 mL/kg/hr was administered to maintain body fluid of animals.

Animals treated with Plavix were checked for efficacy of treatment at time of surgery. In the operating room, under general anesthesia before the surgical procedure begins, 50 mL of blood was removed to be tested for platelet aggregation to assure treatment efficacy. This test was done only for internal verification; it is accepted standard of care in humans to

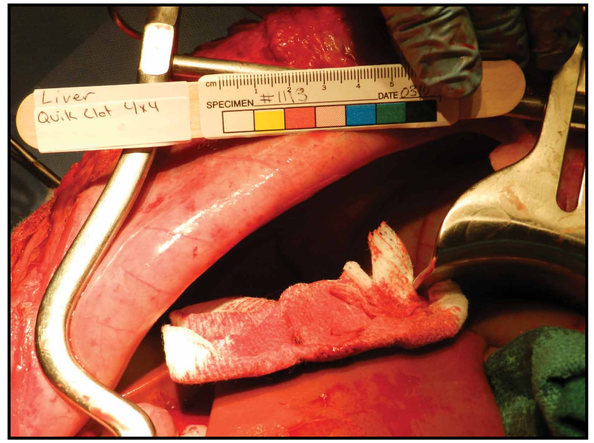

Figure 2: 4x4 QC Dressing Applied to Liver Injury

presume that platelets function is affected by taking Plavix for at least 5 days at 75 mg/day. This concept is also confirmed by studies in pigs.

Under general anesthesia, a midline laparotomy was performed and the liver, spleen and the root of the mesentery were inspected grossly for obvious abnormalities. Each organ was lacerated using a scalpel to cause bleeding as follows: 5 cm long x 0.5 cm deep in the medial lobe of the liver, 5 cm long x 0.5 cm deep in the transverse spleen, and 5 cm long x 0.3 cm deep at the root of the mesentery. The efficacy of QC or the control standard woven cotton surgical gauze (SG) was then tested. QC or SG was applied to the wound and held until bleeding stopped or up to five minutes (Figure 1, 2). A "pass" was recorded if bleeding ceased within 5 minutes of applying the gauze. Multiple injuries were created in each organ to repeat testing of QC or SG.

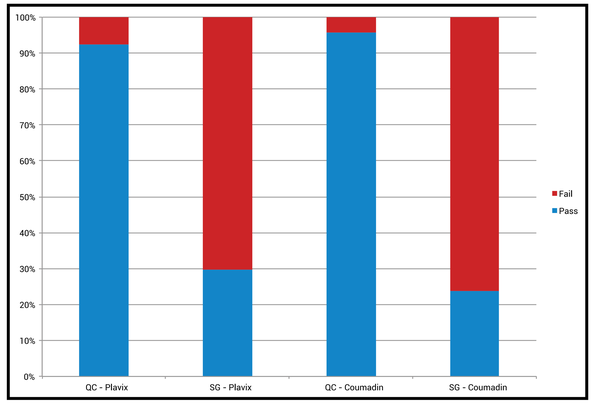

Figure 3: % Tests Passed/Failed by Treatment Group

RESULTS:

A total of 86 tests were performed in Coumadin treated animals: 16 liver, 37 mesentery, and 33 spleen lacerations. All 9 QC treated liver lacerations passed while 4/7 SG failed. In 19/20 mesentery lacerations QC stopped bleeding within 5 minutes while only 3/17 SG passed. 18/19 QC treated spleen lacerations passed while 3/14 SG passed. Overall, 95.83% of QC tests passed while only 23.68% of SG passed (Figure 3, $p=0.000$).

Ninety total tests were performed in Plavix treated animals. All 10 QC treated liver injuries passed while 3/10 SG failed. 19/21 QC and 3/14 SG treated spleen injuries passed. In the mesentery, 20/22 injuries treated with QC passed while only 1/13 treated with SG passed. Overall 92.45% of QC tests passed while only 29.72% of SG passed (Figure 3, $p=0.000$).

CONCLUSION:

In this study, the kaolin-based hemostatic dressing controlled bleeding in liver, mesentery, and spleen injuries more effectively in Plavix (clopidogrel) and Coumadin (warfarin) treated animals than SG. In previous studies, QC was also proven to be more effective than SG in life-threatening hemorrhage models.[13]

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