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A novel model of highly lethal uncontrolled torso hemorrhage in swine

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ABSTRACT

Introduction: A reproducible, lethal noncompressible torso hemorrhage model is important to civilian and military trauma research. Current large animal models balancing clinical applicability with standardization and internal validity. As such, large animal models of trauma vary widely in the surgical literature, limiting comparisons. Our aim was to create and validate a porcine model of uncontrolled hemorrhage that maximizes reproducibility and standardization.
Methods: Seven Yorkshire-cross swine were anesthetized, instrumented, and splenectomized. A simple liver tourniquet was applied before injury to prevent unregulated hemorrhage while creating a traumatic amputation of 30% of the liver. Release of the tourniquet and rapid abdominal closure following injury provided a standardized reference point for the onset and duration of uncontrolled hemorrhage. At the moment of death, the liver tourniquet was quickly reapplied to provide accurate quantification of intra-abdominal blood loss. Weight and volume of the resected and residual liver segments were measured. Hemodynamic parameters were recorded continuously throughout each experiment.
Results: This liver injury was rapidly and universally lethal (11.2 ± 4.9 min). The volume of hemorrhage (35.8% ± 6% of total blood volume) and severity of uncontrolled hemorrhage (100% of animals deteriorated to a sustained mean arterial pressure <35 mmHg for 5 min) were consistent across all animals. Use of the tourniquet effectively halted preprocedure and postprocedure blood loss allowing for accurate quantification of amount of hemorrhage over a defined period. In addition, the tourniquet facilitated the creation of a consistent liver resection weight (0.0043 ± 0.0003 liver resection weight: body weight) and as a percentage of total liver resection weight (27% ± 2.2%).

The animals involved in this study were procured, maintained, and used in accordance with the Laboratory Animal Welfare Act of 1966, as amended, and NIH 80-23, Guide for the Care and Use of Laboratory Animals, National Research Council.

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Conclusions: This novel tourniquet-assisted noncompressible torso hemorrhage model creates a standardized, reproducible, highly lethal, and clinically applicable injury in swine. Use of the tourniquet allowed for consistent liver injury and precise control over hemorrhage. Recorded blood loss was similar across all animals. Improving reproducibility and standardization has the potential to offer improvements in large animal translational models of hemorrhage.
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Introduction

Traumatic hemorrhage remains a leading cause of death globally, and is the most frequent cause of death from potentially survivable injuries.[1-3] While many trauma research insights have ultimately reduced deaths from extremity hemorrhage, there remains great potential to reduce lives lost from noncompressible torso hemorrhage (NCTH).[4-6]

Porcine models predominate in trauma research largely because of similarities to humans in size, anatomy, and physiological response to injury.[7-9] Despite these advantages for trauma related research, current models have significant limitations. Models that use controlled hemorrhage offer reduced variability in hemorrhage volume and duration. The result is tight control of these variables and enhanced internal validity of the study. However, the porcine physiological response to controlled hemorrhage is highly variable and differs greatly from the response to *un*controlled hemorrhage after traumatic injury.[10,11] The net result is that the benefits of controlled hemorrhage models are frequently offset by the decreased clinical applicability in the absence of direct tissue injury. Conversely, existing uncontrolled hemorrhage models provide direct tissue injury and a more realistic physiological response to injury, but these models are notoriously difficult to standardize due to inherent differences in each animal's anatomy and physiological response to injury. These inconsistencies are often difficult to reconcile and can result in large differences in hemorrhage volume and injury lethality. Consequently, controlling for this variation between animals often requires large sample sizes that incur more cost and increase the difficulty of reducing animal numbers for the experiment.[12,13]

This frequently requires an investigator to choose between the two types of hemorrhage models and accept their inherent limitations (Fig. 1). To combine the benefits and minimize the negative aspects of both approaches to modeling NCTH in swine, we have developed a simple liver tourniquet to assist in the creation of a reproducible and highly lethal model of injury. Our ultimate aim was to create a model that maximizes reproducibility and standardization in a highly lethal model of porcine uncontrolled hemorrhage. The purpose of this article is to describe the rationale, technique, and results of this novel approach to studying NCTH in a large animal model.

Methods

Animal preparation

This study was approved by the Institutional Animal Care and Use Committee at David Grant USAF Medical Center, Travis Air Force Base, California. All animal care and use was in strict compliance with the Guide for the Care and Use of Laboratory Animals in a facility accredited by the Association for the Assessment and Accreditation of Laboratory Animal Care International. Healthy adult, castrate male and nonpregnant female Yorkshire-cross swine (*Sus scrofa*) were obtained from a single United States Department of Agriculture-approved vendor through the University of California Davis and were acclimated for a minimum of 7 d before use. At the time of experimentation, animals weighed between 63 and 88 kg, with ages between 5 and 7 mo. Each animal was premedicated with 6.6 mg/kg tiletamine/zolazepam (Telazol; Fort Dodge Animal Health, Fort Dodge, IA) intramuscularly, followed by isoflurane induction and endotracheal intubation. Maintenance anesthesia consisted of 2% isoflurane in 100% oxygen. To overcome the vasodilatory effects of general anesthesia, an intravenous infusion of norepinephrine (0.01 mcg/kg/h) was initiated and then titrated to a mean arterial pressure (MAP) of at least 75 mmHg. Animals were mechanically ventilated with tidal volumes of 7-10 mL/kg and a respiratory rate of 10-15 breaths per minute sufficient to maintain end tidal CO_2 at 40 ± 5 mmHg. The pigs were placed on a warming blanket set at 39°C to minimize hypothermia. Arterial access was obtained to facilitate blood collection and hemodynamic monitoring. Normal saline was administered at a maintenance rate of 5 mL/kg/hr. A 25 units/kg bolus of heparin sulfate was administered to achieve an activated clotting time of 90-130 s.

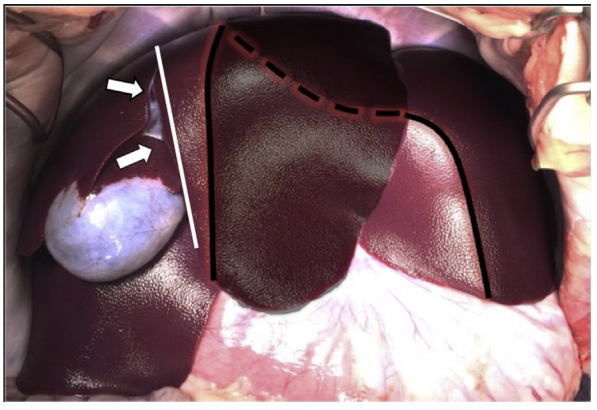

Fig. 1 – *In situ* porcine liver. Note the presence of four distinct hepatic lobes. The white line and white arrows denote Cantlie's line. The black lines along the left lateral and medial lobes indicate the resection margin. The shaded portions of these lobes indicate the resected area. This margin provides amputation of approximately 80% of the left lateral lobe of the liver and 40% of the left medial lobe of the liver. (Color version of figure is available online.)