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Centhaquin improves survival in a swine model of hemorrhagic shock

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ARTICLE INFO

Article history:
Received 28 April 2015
Received in revised form
19 June 2015
Accepted 23 June 2015
Available online 29 June 2015

Keywords:
Hemorrhagic shock
Centhaquin
Resuscitation
Fluids
Survival

ABSTRACT

Background: Hemorrhage is a frequent event in hospital and prehospital settings. The aim of the present study was to investigate whether centhaquin improves 24-h survival and reduces the total volume of required fluids in an established model of swine hemorrhagic shock.

Material and methods: Twenty-five pigs were instrumented and subjected to hemorrhagic shock. The animals were randomly allocated in two experimental groups, the control (vehicle) ($n = 10$) and the centhaquin groups (0.015 mg/kg, $n = 10$); all animals received lactated Ringer solution in the resuscitation phase until their mean arterial pressure reached 90% of the baseline. A sham group ($n = 5$) was added *a posteriori* to mimic the hemodynamic profile of the centhaquin group.

Results: A statistically significant difference was observed in the time required for the three groups to reach their target mean aortic pressure, 36.88 ± 3.26 min for the control group *versus* 9.40 ± 1.01 min for the sham group and 7.10 ± 0.97 min for the centhaquin group ($P < 0.001$). The total amount of fluids in the control and the sham groups was significantly higher when compared with that of the centhaquin-treated animals ($P < 0.001$). All 10 animals in the centhaquin group survived for 24 h, whereas only three animals survived in the control group and one animal in the sham group ($P = 0.002$).

Conclusions: Centhaquin 0.015 mg/kg administered in the fluid resuscitation phase resulted in lower volume of fluids and better survival compared with control and sham-operated animals.

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http://dx.doi.org/10.1016/j.jss.2015.06.056

1. Introduction

Hemorrhage, which accounts for a large proportion of early deaths, is a frequent event in various hospital and prehospital settings. Marked loss of intravascular volume may lead to severe hemodynamic instability, decreased tissue perfusion, impaired oxygen delivery, cellular hypoxia, organ damage, and eventually death [1]. The current recommendations of the American College of Surgeons for the treatment of shock due to acute hemorrhage call for fluid resuscitation with crystalloids preferably with lactated Ringer solution (R-L), transfusion of compatible blood, and surgical repair of the reversible causes of hemorrhage [2–4]. Although the aim of the first-line treatment with fluids was to increase the circulating volume of blood and improve the systolic and diastolic pressures [5], large amounts of crystalloids have been associated with various side-effects [6]. As a result, an on-going debate is whether the same beneficial effects that are derived from aggressive fluid therapy can be achieved with reduced amount of fluids. To this end, centhaquin has been shown in various studies to have direct and indirect positive inotropic effects, without increasing the excitability of the myocardium [7]. Moreover, centhaquin can cause peripheral vasodilation, reduction in mean arterial pressure and, as a result, reduction in systemic vascular resistance (SVR) [8]. These combined actions of restoring cardiac output (CO) and reduction of SVR may be of importance in resuscitation in hemorrhagic shock.

Studies showed that centhaquin improves tissue oxygenation and short-term survival in rats [9]. Whether centhaquin has any effect on the 24-h survival and on total fluid requirements and resuscitation times remains unknown. The primary aim was to investigate whether centhaquin improves the 24-h survival, and the secondary aim was to assess whether this substance may reduce the total required fluids and achieve quicker resuscitation times in an established model of swine hemorrhagic shock.

2. Materials and methods

The protocol was approved by the Hellenic Veterinary Services (license No 7157/30-11-2012). Twenty-five female pigs of conventional microbiological status with an average weight of 20 ± 1 kg (aged 10–12 wk) were the study subjects. All animals received anesthetic and surgical procedures in compliance with the Guide for the Care and Use of Laboratory Animals.

The animals were transported 1 wk before experimentation [10] to the research facility (Experimental-Research Center ELPEN, European Ref Number EL 09 BIO 03). All pigs were purchased from the same breeder (Validakis, Koropi, Greece). The animals were fasted the day before the experimentation, but they had free access to water.

Centhaquin citrate (Lot # PMZ-2010/2012/09A) was synthesized at Pharmazz India Private Limited, Greater Noida, India (courtesy Dr Manish Lavhale). The experimental protocol has been previously described [11]. In brief, the animals were premedicated with intramuscular ketamine hydrochloride (Merial, Lyon, France) 10 mg/kg, midazolam (Roche, Athens, Greece) 0.5 mg/kg, and atropine sulfate (Demo, Athens, Greece) 0.05 mg/kg. The animals were subsequently transported to the operation research facility and intravascular access (iv) was obtained through the auricular veins. Induction of anesthesia was achieved with an intravenous bolus dose of propofol (Diprivan 1% w/v; AstraZeneca, Luton, United Kingdom) (2.0 mg/kg) and fentanyl (Janssen Pharmaceutica, Beerse, Belgium) (2 μg/kg). The same researcher performed the intubation, while the animals were breathing spontaneously, with a size 6.0-mm cuffed endotracheal tube. The endotracheal tube was secured on the lower jaw, and successful intubation was ascertained by auscultation of both lungs while ventilated with a self-inflating bag.

The animals were then immobilized in the supine position on the operating table. Additional propofol 1 mg/kg, cisatracurium (Nimbex 2 mg/mL GlaxoSmithKline, Athens, Greece) 0.15 mg/kg, and fentanyl 0.01 mg/kg were administered to ascertain synchrony with the ventilator. The animals were mechanically ventilated (Siare Alpha-Delta Lung Ventilator; Siare s.r.l. Hospital Supplies, Bologna, Italy) with a gas mixture of 40% oxygen. Anesthesia was maintained with infusion of propofol 150 μg/kg/min (propofol MCT/LCT 1%; Fresenius Kabi Hellas A.E., Athens, Greece). Normocapnia was achieved using continuous monitoring of end-tidal CO_2 (Tonocap TC-200-22-01; Engstrom Division, Instrumentarium Corp, Helsinki, Finland), and the respiratory rate was adjusted to maintain end-tidal CO_2 35–40 mm Hg. Pulse oximetry (SpO_2) was monitored throughout the experiment. Body temperature was monitored by a rectal temperature probe and was maintained between 38.5°C and 39.5°C with a heating blanket.

Electrocardiographic monitoring was used, using leads I, II, III, aVR, aVL, and aVF, which were connected to a monitor (Mennen Medical, Envoy; Papapostolou, Athens, Greece). The monitor electronically calculated the heart rate. For measurement of the aortic pressure, an arterial catheter (model 6523, USCI CR, Bart; Papapostolou) was inserted and forwarded into the descending aorta after surgical preparation of the right internal carotid artery. The systolic and diastolic pressures were recorded, whereas mean aortic pressure (MAP) was determined by the electronic integration of the aortic blood pressure waveform. The right internal jugular vein was surgically prepared, and a catheter was inserted for fluid administration. The right internal jugular vein was also cannulated with a catheter to measure central venous pressure (CVP). Intravascular catheters were attached to pressure transducers that were aligned to the level of the right atrium and were calibrated before their use. This allowed the recording of CVP and systolic, diastolic, and mean pressures of the aorta. CO was measured as the product of time-velocity integral of Doppler transaortic flow, the diameter of the aortic valve, and heart rate, as previously described [12]. SVR was calculated using the formula $SVR = (MAP - CVP)/CO \times 80$, as previously described [9], measured in mm Hg × min/L. Arterial blood gases were measured on a blood-gas analyzer (IRMA SL Blood Analysis System, Part 436301; Diametrics Medical Inc, Roseville, MN 55113, pH, pO_2, pCO_2).

The protocol simulated a civilian trauma scenario and has been previously described [13]; it was divided into six distinct phases: stabilization, hemorrhagic, maintenance,