

BACKGROUND

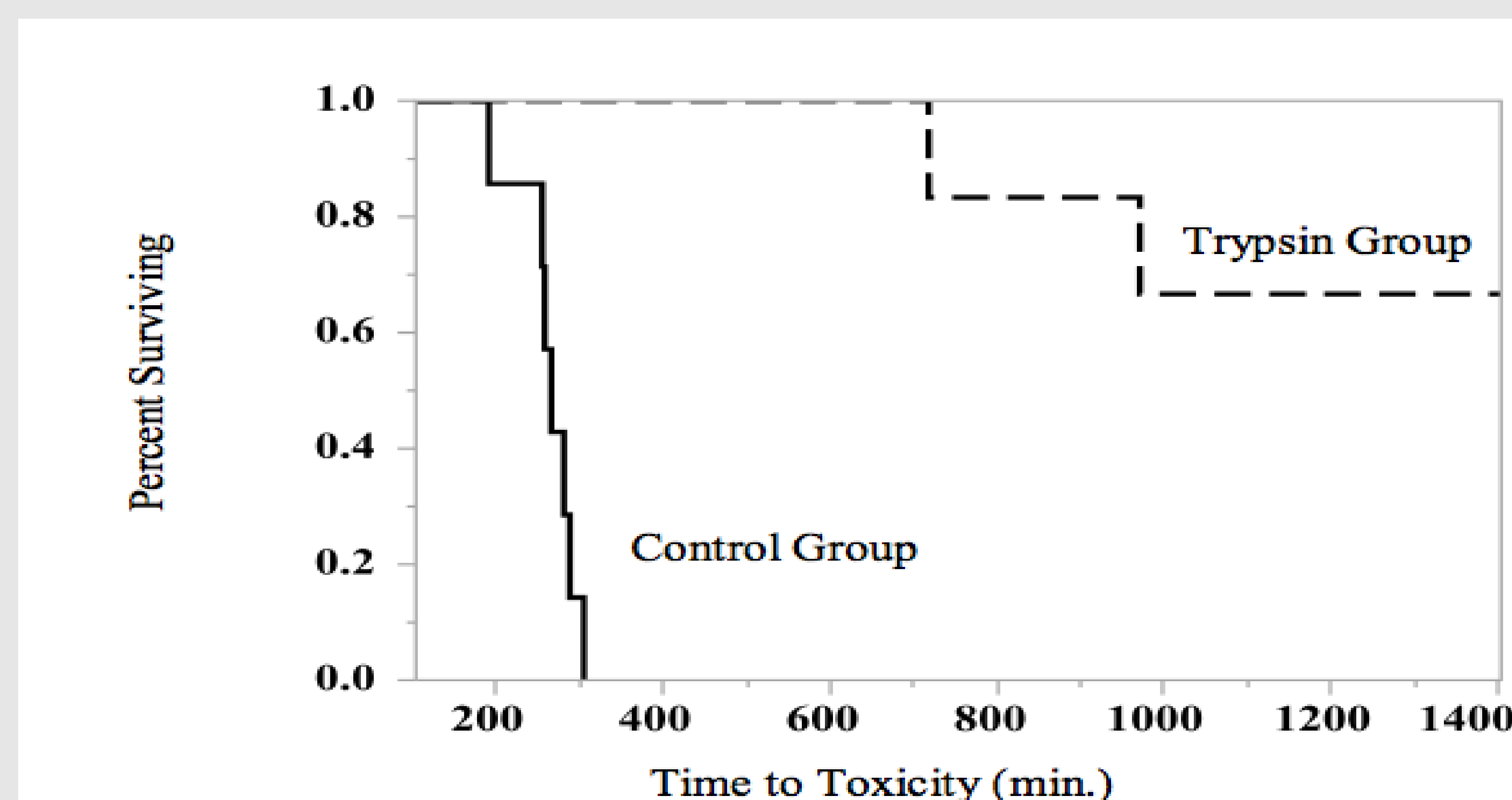
- ◆ Investigation into alternative treatments to Eastern coral snake (*Micrurus fulvius*) envenomation is beneficial because antivenom for coral snake is not immediately available.
- ◆ There is also the risk of potential adverse events such as anaphylaxis, anaphylactoid reaction, and delayed serum sickness, reported to occur in 18% of those receiving North American Coral Snake Antivenin.
- ◆ Trypsin is a serine protease digestive enzyme that has demonstrated efficacy in other *Elapidae* envenomations.
- ◆ A prior *in vitro* pilot study demonstrated that pre-incubation of trypsin with *M. fulvius* venom preceding intraperitoneal injection into mice prolonged survival significantly compared to venom only controls.
- ◆ **Objective:** The aim of this study is to assess the efficacy of trypsin in an *in vivo* porcine model by assessing survival after treatment of Eastern coral snake envenomation with the local injection of trypsin at the envenomation site.

MATERIALS & METHODS

- ◆ Six week-old Yorkshire, Duroc, Landrace mixed breed pigs were used.
- ◆ The animal use protocol was approved by the ECU Animal Care Committee.
- ◆ 13 females were randomized into 2 experimental groups: venom and saline control (n=7) vs. venom and trypsin (n=6).
- ◆ Animals were sedated and anesthetized with 2% isoflurane and 2% oxygen.
- ◆ Buprenorphine (0.02 mg/kg) was given pre-operatively and meloxicam (0.4 mg/kg) post-operatively for analgesia.
- ◆ Freeze-dried Eastern coral snake venom (MedToxin, Deland, FL) was dissolved in sterile water (10 mg/mL).
- ◆ Lyophilized trypsin powder (Worthington Biochemical Corporation, Lakewood, NJ) was dissolved in normal saline (100 mg/ml).
- ◆ Animals received 1 mL subcutaneous injection of Eastern coral snake venom in the right distal hind limb.
- ◆ One minute post injection animals were either injected with 1 mL of saline or 1 mL of trypsin solution at the site of venom injection.
- ◆ Animals were extubated after 4 hrs. Animals were monitored continuously the first four hours, and then every 8 hours for the next three days for signs of toxicity, defined as respiratory rate of <15 breaths/min, apnea, falling pulse oximetry, or agonal respirations.
- ◆ Animals displaying signs of respiratory distress, self-mutilation, or morbidity they were euthanized.

RESULTS

Figure 1: Survival curves for pigs injected with 10 mg Eastern Coral snake venom, followed by injection with 100 mg trypsin in 1 mL saline, or 1 mL of saline without trypsin.



SUMMARY

- ◆ All pigs in the control group expired before the end of the 3-day study period.
- ◆ Four pigs in the treatment group survived to the end of the study period.
- ◆ The 2 trypsin treatment animals that did not survive showed signs of toxicity at 844.5 ± 178.9 minutes vs. controls 263.42 ± 36.5 minutes.
- ◆ Using Fisher's Exact test, control pigs were more likely to develop signs of toxicity ($p=0.009$) and less likely to survive to 12 hours ($p=0.002$) or 24 hours ($p=0.009$) than treated pigs.
- ◆ Trypsin injection produced inflammation evolving to ulceration at the site of injection in all treated pigs that survived the 3 day study period, but these animals were walking on the extremity without evidence of significant pain

CONCLUSION

- ◆ Local injection of trypsin, a proteolytic enzyme, at the site of envenomation decreased the toxicity of eastern coral snake venom and increased survival significantly. Further investigation is required to determine the efficacy trypsin injection at later treatment injection times.

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