

ORIGINAL ARTICLE

The effect of an electrical current on snake venom toxicity*

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An electric current (twenty 11 A, 7000 V spikes s⁻¹ for 90 s) from a commercial stun gun was applied directly to a rattlesnake venom solution in an electrolysis cell with 2 electrode compartments and a central compartment, in order to evaluate the effect that high voltage electroshock might have on the lethality of the venom. The venom was electroshocked for 18 times longer than recommended by stun gun manufacturers. There was no measurable inactivation of the venom using LD₅₀ determinations in mice. A venom sample was electrolyzed at a voltage lower than that from the stun gun, but with 4-5 times the total charge delivered from the stun gun. This inactivated the venom at the electrodes, but not within the central compartment, demonstrating that there was no direct effect of the electric field on the activity of the snake venom.

Key words: snake venom, envenomation, stun gun, electrical current

Introduction

Following publication of a letter [1] concerning the use of electroshock for snake venom poisoning, a number of articles appeared in the popular press exalting its value [2]. For the most part, these accounts have been anecdotal and unsupported by controlled experimental or clinical evidence. Both the logic and safety of the electroshock technique have been questioned [3,4], and subsequent experiments in animals have shown that electroshock is of questionable efficacy in snake venom poisoning [5-7]. Despite these data, electroshock therapy is still extolled in lay literature and first aid programs as an efficacious treatment modality.

Since some have claimed that the electroshock method inactivates venom proteins by a direct effect of the high voltage, the present study was initiated to test this hypothesis. Because high voltage electrophoresis is a routine laboratory tool for separating and isolating active proteins from complex biological mixtures, it appeared unlikely that a direct current (DC) electric field applied from a stun gun to a solution of venom proteins would have any extraordinary effect not seen with electrophoresis. In fact, high voltage

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electrophoresis has been used successfully to fractionate snake venom for the purpose of isolating active, single components [8]. The apparent confusion of whether an electric field inactivates snake venom can arise from differences in experimental designs. That is, one might expect contrary results between electrolysis performed by submerging electrodes directly into a venom solution and electrophoresis, which is similar to electroshock treatment in that the venom proteins are compartmented and shielded from direct contact with the electrodes. The present investigation was designed to determine whether an electric current, passing through a solution of snake venom shielded from contact with the electrodes, would have an effect different from that it would have on a solution in direct contact with the electrodes. It was also designed to determine whether electric currents, at voltages delivered from a commercially available 'stun gun,' would inactivate snake venom.

Materials and methods

An electrolysis cell was designed to mimic the compartmentation of venom following an envenomation, only to the extent that the membranes prevented the snake venom proteins from direct contact with the electrodes. The three-compartment cell consisted of two electrode compartments, separated by dialysis membranes from a central compartment. The dialysis membranes were separated by approximately 5 cm, equal to the distance between the electrodes of the stun gun. Each compartment was loaded with identical venom solution.

Figure 1 is a photograph of the electrolysis cell used in the experiments. Each of the three cell compartments held 2 ml. An access port leading into each reservoir was not included in the cell volume. The central compartment was separated from the electrode compartments by Spectropore dialysis membranes of molecular weight cutoff 6000–8000 Da to retain smaller, potentially toxic peptides.

Two similar electrolysis experiments were performed, differing only in the power supply, applied voltage and time of electrolysis. Both experiments used the same electrolysis cell, with 2.0 ml of 2.0 mg ml⁻¹ venom solution in each of the three cell compartments. At the end of each electrolysis, the electrode-containing solutions were pooled and the LD₅₀ was determined separately for the pooled electrode-containing compartments and for the central compartment. The pH of each solution was determined.

One experiment was designed to determine the effects of an electric field from a stun gun on snake venom. For this experiment, the electrolysis cell was powered with a Nova XR-5000 Stun Gun (Nova Technologies, Austin, TX, USA). The stun gun is normally powered with a 9 volt battery. For this investigation, a constant voltage power supply was substituted for the battery, since it would otherwise become drained during the course of an experiment. According to the manufacturer, the stun gun delivers a spike of 7000 V peak output over 5 μs, and 20 spikes s⁻¹. As the resistance of the electrolysis cell was approximately 625 ohms, a peak current of 11 A was produced. After filling the cell with the stock venom solution, the power was applied for 90 s. The voltage gradient and electric current applied to the electrolysis cell were identical to those recommended by Nova Technologies for the stun gun treatment.

A separate experiment was designed to compare the effects of the electric field to those of the electrode reactions on the lethality of snake venom. In this experiment, the electrolysis cell was powered with a BioRad model 1000/500 power supply. This

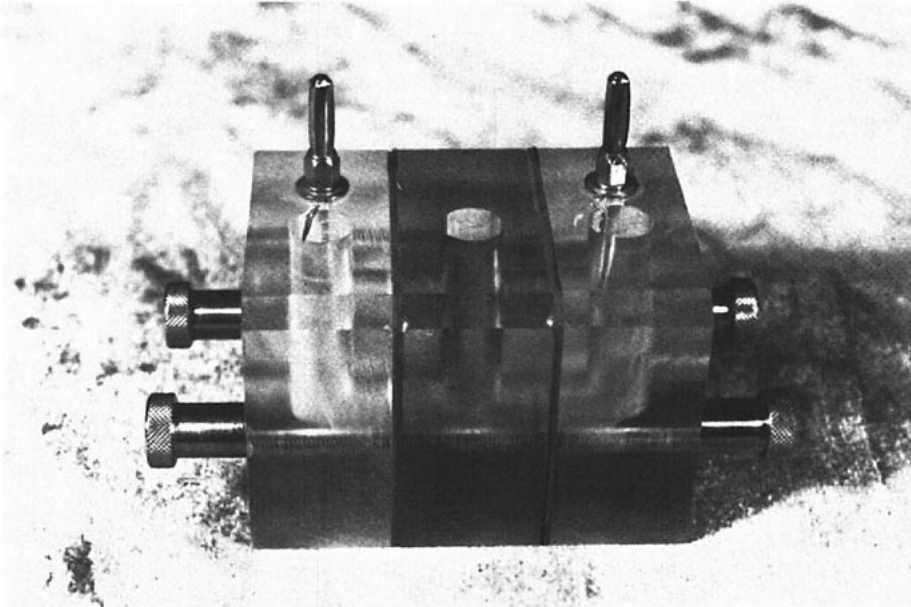


Fig. 1. Electrolysis cell configured to have inter-electrode distance identical to that of commercial ‘stun gun’.

provided continuous power at constant voltage (10 V) for 30 s. The average current was 16 mA. Using the two different power supplies enabled us to compare the difference between electrode reactions and the effects of merely passing an electric current through the venom solution.

The effect of venom electrolysis (‘electroshock’) was determined by comparing the LD_{50} before and after treatment. An increase in the LD_{50} was taken to signify venom inactivation. LD_{50} studies were performed using female, 20–24 g, ICR strain mice (Harlan Sprague-Dawley, Indianapolis, IN). A stock solution of 2.0 mg ml^{-1} freeze-dried rattlesnake (*Crotalus adamanteus*) venom was prepared in 0.15 M NaCl, incubated at 37°C for 30 min and chilled to 0°C until used. The venom solution was added directly to the electrolysis cell or diluted to various concentrations for the LD_{50} determinations. All mice received 0.20 ml of the venom dilutions via tail vein through a 250 μl microsyringe fitted with a 27 gauge needle. Six mice were used for each of five venom doses. These doses were 0.90, 1.35, 1.80, 2.25 and 2.70 mg kg^{-1} .

Results

Figure 2 presents the dose response curves for the four venoms: control, venom from central and electrode compartments of the stun gun experiment, and venom from the central compartment for the BioRad 1000/500 power supply source. Table 1 shows the LD_{50} of each venom, obtained from the curves.

For electrolysis powered by the stun gun, there was no difference in lethality between the control venom and the experimental samples in either the central compartment or

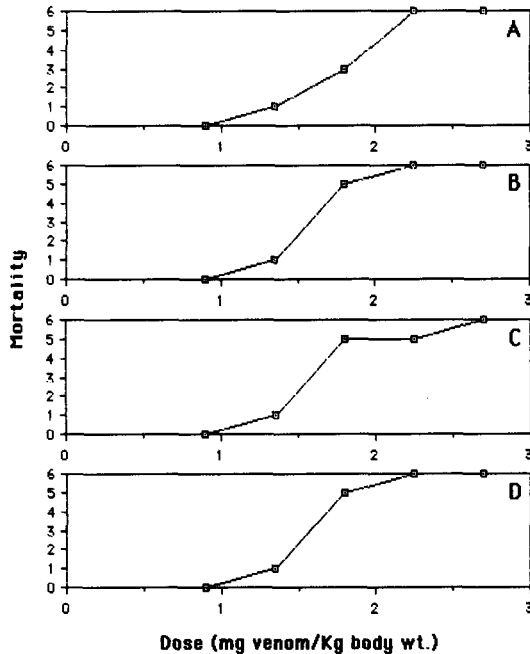


Fig. 2. Dose response curves for determining the LD_{50} s of rattlesnake venom: A, control venom; B and C, venom electroshocked with the stun gun and collected from the central and the electrode compartments, respectively; D, venom electrolyzed at 10 V, recovered from the central compartment.

pooled electrode-containing compartments. That is, the high voltage electric field did not inactivate the venom. For electrolysis using the constant voltage power supply, there was also no measurable inactivation of venom in the central compartment. However, venom pooled from the electrode-containing compartments was completely inactivated. Its corresponding dose response curve, therefore, is not presented.

Discussion

The Nova XR-5000 stun gun-powered electrolysis provided twenty 11 A, 7000 V spikes s^{-1} for a duration of 90 s. However, as each spike only exists for 5 μs , only 1.0 μM of electrons was generated during this experiment. Assuming that the size of an average venom protein is 30 kDa, there was 0.13 μM of venom protein in each compartment. Furthermore, if it required a minimum of one electron to denature a venom protein molecule at the electrode surface, then there would be sufficient electrons to inactivate 7–8 times the venom present in the electrode compartments. The fact that there was no observed venom inactivation in any of the cell compartments clearly indicates that DC electric current from the stun gun did not inactivate lethal components of the venom. It further implies that at the electrodes, either water and/or salt were oxidized (anode) and reduced (cathode), rather than the venom.

In both experiments, only 4 mg of *C. adamanteus* venom were present in each compartment; however, the entire sample was presented to the electric field. With a stun

Table 1. Lethal dose 50 for sampled venom pools

<i>Treatment</i>	<i>Compartment</i>	<i>pH</i>	<i>LD₅₀</i>
Control		5.47	1.7
Nova power supply	central electrode pool	5.47	1.6
		anode, 5.47 cathode, 5.50	1.5
BioRad power supply	central electrode pool	5.48	1.5
inactivated		anode, 2.22 cathode, 11.46	

gun treatment of an actual human envenomation, it is unlikely that the entire venom sample would be presented to the electric field. This would further reduce the likelihood of an electric current from a stun gun affecting snake venom proteins. If it is assumed that an envenomation capable of causing significant morbidity could contain as much as 100 mg of venom, then the design of our experiment represented an electrolysis of 4% of the dose of such an envenomation. Electrolysis of 90 s is 18 times the duration of the 5 s recommended for snakebite treatment by the makers of stun guns. Therefore, the total energy per mg of venom during the electrolysis in our cell was 450 times greater than that used for snakebite treatment. Still, there was no evidence of venom inactivation.

Electrolysis of the venom by the constant voltage power supply was 10 V at 16 mA for 30 s. Thus, we applied 4–5 times the charge delivered by the stun gun operated at 7000 V, which is pulsed for 90 s. Even under the conditions of constant voltage power supply, venom in the central compartment of the electrolysis cell was unaffected by the electric field. Inactivation of venom in the electrode-containing cells was a result of the extreme pH observed in these compartments.

Snake venom delivered into a human victim from fang punctures is subject to absorption by lymphatics and venous capillaries. However, it is our opinion that *in vivo* distribution of venom would only serve to remove the venom from any potential effects of an electric field. We purposely designed our experiments to immobilize the venom within the electric field and remove these variables. Quite frankly, the notion that electricity applied to a snake bite would eliminate the toxicity by any mechanism other than venom degradation, such as alteration of vascular reactivity or augmentation of immune response, seems exceedingly far-fetched.

We only studied the lethal potential of venom and did not attempt to quantify any effects that electrical shock might have upon local tissue reactions, such as necrosis, or nonlethal systemic derangements, such as coagulopathy.

Perhaps, some things have not changed much since the following was written over 30 years ago:

'It is apparent that folklore cures are more than historical curiosities. Whatever their source, they are hazardous: first, because they often involve dangerous methods; second, because they delay the use of really effective therapeutic procedures. There are probably two principal reasons why folklore remedies remain so prevalent in the snakebite literature: 1) The emergency usually occurs in remote

areas that may be distant from skilled medical help. 2) Snakebite is an accident highly variable in the gravity of its results. It is one in which the most fantastic remedy may gain its reputation among credulous people by having cured a malady that required no treatment whatever' [9].

Within the recent past, the Food and Drug Administration has taken action against marketing the stun gun for use in snakebite.

On the basis of our experiments and the absence of compelling alternate biophysical or biochemical evidence, any recommendation for stun gun treatment for snake venom poisoning is as yet without scientific merit.

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