LABORATORY INVESTIGATION

Reduction of Muscle Contractions during Irreversible Electroporation Therapy Using High-Frequency Bursts of Alternating Polarity Pulses: A Laboratory Investigation in an Ex Vivo Swine Model

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ABSTRACT

Purpose: To compare the intensity of muscle contractions in irreversible electroporation (IRE) treatments when traditional IRE and high-frequency IRE (H-FIRE) waveforms are used in combination with a single applicator and distal grounding pad (A+GP) configuration.

Materials and Methods: An ex vivo in situ porcine model was used to compare muscle contractions induced by traditional monopolar IRE waveforms vs high-frequency bipolar IRE waveforms. Pulses with voltages between 200 and 5,000 V were investigated, and muscle contractions were recorded by using accelerometers placed on or near the applicators.

Results: H-FIRE waveforms reduced the intensity of muscle contractions in comparison with traditional monopolar IRE pulses. A high-energy burst of 2-μs alternating-polarity pulses energized for 200 μs at 4,500 V produced less intense muscle contractions than traditional IRE pulses, which were 25–100 μs in duration at 3,000 V.

Conclusions: H-FIRE appears to be an effective technique to mitigate the muscle contractions associated with traditional IRE pulses. This may enable the use of voltages greater than 3,000 V necessary for the creation of large ablations in vivo.

ABBREVIATIONS

A+A = two applicators [configuration], A+GP = single applicator and grounding pad [configuration], H-FIRE = high-frequency irreversible electroporation, IRE = irreversible electroporation

Irreversible electroporation (IRE) is an emerging cancer therapy that uses high-voltage pulsed electric fields to rapidly ablate solid tumors. The therapy involves the insertion of two or more electrodes directly into malignant tissue. A series of 50–100 μs positive-polarity (ie, monopolar) pulses with voltages between 1,000 and 3,000 V are then delivered between electrode pairs. These pulses increase the cells' transmembrane potentials, resulting in the

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M.B.S. and L.X. have two patents pending related to the subject of this article: Methods for Enhancing and Modulating Reversible and Irreversible Electroporation Lesions by Manipulating Pulse Waveforms (US 62/346,903) and Methods for Producing and Evaluating Spherical Ablations Using Irreversible Electroporation (US 62/287,179). M.B.S. also has the following patents related to the subject of this article: Devices, Systems, and Methods for Real-Time Monitoring of Electrophysical Effects During Tissue Treatment (PCT/US15/65792, pending), Mapping of Electric Field and Thermal Contours Using A Simplified Data Cross-Referencing Approach (US Application 61/910,655, with royalties paid to AngioDynamics, Latham, New York), Sub-Lethal Blood Brain Barrier Disruption (US Application 62/022,814, pending), Selective Cell Modulation with Electric Fields (Provisional Patent Application 61790702, pending), Delivery of Electrical Pulses for Electroporation Treatments Utilizing the Vasculature of a Tissue or Organ (US Application 61/416,53, International Application PCT/US2011/062067, with royalties paid by AngioDynamics), and High-Frequency Electroporation for Cancer Therapy (US Application 13/332,133 with royalties paid by AngioDynamics). None of the other authors have identified a conflict of interest.

Table E1, Figures E1–E4, and Videos 1 and 2 are available online at *www.jvir.org*.

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EDITORS' RESEARCH HIGHLIGHTS

- High-intensity, higher-voltage pulse irreversible electroporation (IRE) is explored to minimize muscle contractions; however, the optimal protocols for its delivery are lacking. Such protocols might increase the size of the ablation zone and potentially reduce probe insertions.
- These investigators show that bipolar high-frequency IRE delivered by using a single probe and padding was effective in minimizing the intensity of muscle contractions compared with traditional monopolar IRE at 5,000-V levels in a porcine model. If translatable to humans, this might broaden the utility of IRE, potentially expand applications, and reduce anesthetic requirements, possibly reducing the risk of paralysis.

creation and expansion of nanoscale pores in the cell membrane, which ultimately lead to cell death (1). This nonthermal method of cell destruction makes IRE an attractive ablative therapy for inoperable tumors close to major blood vessels, where risk of vessel occlusion (2) and heat-sink effects (3) may contraindicate thermal ablation techniques (4).

Although IRE has shown promise in early clinical trials (5), a number of technical challenges exist. The procedure requires the precise placement of multiple applicators with millimeter precision. Ablations created with IRE are typically smaller (1–3 cm) than ablations created with microwave or radiofrequency ablation, requiring the placement of three or more applicators to treat most tumors (6). The ablations created have an irregular, oblong oval, or peanut shape, requiring some degree of preoperative planning for complicated tumor geometries (7–10). Finally, the long-duration monopolar pulses induce strong muscle contractions, requiring an anesthetic regimen that includes chemical paralytic agents (11). There is an existing clinical need for new IRE protocols that produce larger, more predictable ablation volumes while also constraining the induced muscle contractions to levels that can be medically managed.

The present study was performed to investigate the intensity of muscle contractions induced by two modifications developed to address this need. The first is the use of a single applicator and grounding pad (A+GP). The advantage of this modification is the production of larger, more spherical ablations (12). However, this technique exposes a larger volume of muscle tissue to elevated electric fields than the standard two-applicator (A+A) configurations, which increases the potential to induce muscle contractions. Swine studies indicate that 1–3-mg/kg doses of rocuronium are sufficient for the management of muscle contractions for A+A configurations (13), but substantial pulse-induced neuromuscular activation was observed with the A+GP configuration under a similar anesthetic agent dose configuration (14).

The second modification is the use of high-frequency IRE (H-FIRE) pulses (15–17). In H-FIRE, the long-duration monopolar IRE pulses (**Fig 1a**) are replaced with a rapid burst of 0.25–5-μs alternating-polarity (ie, bipolar) pulses (**Fig 1b**). The electric field required to induce muscle contractions with the short-duration bipolar pulses used in H-FIRE is substantially higher than in traditional IRE (18,19). This technique has been used to treat spontaneous large-animal tumors without the need for general anesthesia or neuromuscular blockade (20). However, the ablations created by H-FIRE are typically smaller than those created by traditional IRE at the same voltage (20,21). This requires the use of higher-voltage pulses to achieve ablation volumes equivalent to those created with traditional IRE.

The motivating hypothesis for transitioning to H-FIRE is that the decrease in muscle contractions will enable the use of substantially greater voltages (4–6 kV) with equivalent paralytic agent doses, ultimately yielding larger overall ablations than possible with traditional IRE pulses. The purpose of the present study was to investigate the first component of this hypothesis, if muscle contractions caused by high-voltage H-FIRE waveforms are of similar or lesser intensity compared with traditional IRE protocols implemented clinically. Assessment and optimization of ablation volumes is left for future in vivo investigation.

MATERIALS AND METHODS

Animal Model

This study used female Yorkshire pigs (45–55 kg) that had been recently euthanized as part of other research and training exercises. Institutional review board approval was not required because the animals were euthanized as part of other studies. Treatment protocols with three different applicator configurations were administered on three different animals, and all experiments were completed within approximately 2 hours of euthanasia.

Applicator Configurations

In situ experiments were conducted to assess the feasibility of using a 3-cm umbrella electrode array (LeVeen M001262050; Boston Scientific, Marlborough, Massachusetts) and grounding pad configuration for the ablation of abdominal malignancies. A large incision was made to expose the liver, the LeVeen applicator was inserted approximately 2–3 cm into the liver, with use of the visual indicators on the applicator, and fully deployed. The skin on the hind limb was cleaned with ethanol and allowed to air-dry before placement of the grounding pad. Accelerometers were attached directly to the applicator handle and grounding pad. Additional "worst-case scenario" experiments were conducted with (*i*) an A+A configuration (bipolar probe; AngioDynamics, Latham, New York) with both electrodes directly in muscle tissue and (*ii*) an A+GP configuration with one electrode on the bipolar probe in muscle tissue paired with a distal grounding pad. For all protocols, the applicators were placed once and remained in the exact same location for the duration of the experiment.