Modulating the Infarcted Ventricle's Refractoriness with an Epicardial Biomaterial

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Accepted 13 October 2020
Published Online First 28 October 2020

To cite: Chinyere IR, Hutchinson M, Moukabary T, *et al*. *J Investig Med* 2021;**69**:364–370.

ABSTRACT

Patients diagnosed with heart failure with reduced ejection fraction (HFrEF) are at increased risk of monomorphic ventricular tachycardia (VT) and ventricular fibrillation. The presence of myocardial fibrosis provides both anatomical and functional barriers that promote arrhythmias in these patients. Propagation of VT in a reentrant circuit depends on the presence of excitable myocardium and the refractoriness of the circuit. We hypothesize that myocardial refractoriness can be modulated surgically in a model of HFrEF, leading to decreased susceptibility to VT.

Male Sprague-Dawley rats were infarcted via permanent left coronary artery ligation. At 3 weeks post-infarction, engineered grafts composed of human dermal fibroblasts cultured into a polyglactin-910 biomaterial were implanted onto the epicardium to cover the area of infarction. Three weeks post-graft treatment, all rats underwent a terminal electrophysiologic study to compare monophasic action potential electroanatomic maps and susceptibility to inducible monomorphic VT. HFrEF rats (n=29) demonstrated a longer (p=0.0191) ventricular effective refractory period (ERP) and a greater (p=0.0394) VT inducibility compared with sham (n=7). HFrEF rats treated with the graft (n=12) exhibited no change in capture threshold (p=0.3220), but had a longer ventricular ERP (p=0.0029) compared with HFrEF. No statistically significant change in VT incidence was found between HFrEF rats treated with the graft and untreated HFrEF rats (p=0.0834).

Surgical deployment of a fibroblast-containing biomaterial in a rodent ischemic cardiomyopathy model prolonged ventricular ERP as measured by programmed electrical stimulation. This hypothesis-generating study warrants additional studies to further characterize the antiarrhythmic or proarrhythmic effects of this novel surgical therapy.

INTRODUCTION

Patients with heart failure with reduced ejection fraction (HFrEF) have an increased susceptibility to sustained ventricular arrhythmias.[1 2] Sustained monomorphic ventricular tachycardia (VT) can also trigger ventricular fibrillation and ultimately sudden cardiac death.

Significance of this study

What is already known about this subject?

- Biomaterials can be synthesized for nearly any application, with optimization of parameters such as density, degradation time, surface charge and roughness, and electrical resistivity.
- Engineered cardiac grafts that use synthetic biomaterials as the foundational scaffold before cell incorporation are increasing in popularity as basic scientists investigate novel therapies for cardiovascular disease.
- In heart failure with reduced ejection fraction (HFrEF), the creation of arrhythmogenic substrate predisposes to reentrant tachyarrhythmias such as monomorphic ventricular tachycardia.
- Pharmacologic management and catheter ablation are proven therapy options, but have shortcomings that biomedical engineering innovations may address.

What are the new findings?

- In a rat model of HFrEF, epicardial deployment of a fibroblast-containing biomaterial at 3 weeks post-myocardial infarction resulted in a prolonged effective refractory period at 6 weeks post-myocardial infarction.
- While the decrease in the incidence of inducible monomorphic ventricular tachycardia for graft-treated HFrEF rats did not achieve statistical significance, as compared with non-treated HFrEF rats, the decrease may be physiologically relevant and warrants further study.
- Echocardiographic and invasive hemodynamic indices of cardiac function were nearly unchanged in graft-treated HFrEF rats, with respect to non-treated HFrEF rats.

How might these results change the focus of research or clinical practice?

- To date, no surgical biomaterial has been shown to decrease susceptibility to ventricular tachyarrhythmia subsequent to heart failure in the clinical setting.

Significance of this study

- While many biomaterial investigators are attempting to restore conduction through infarcted myocardium, these data suggest that locally prolonging the effective refractory period through infarcted myocardium with a surgical biomaterial may also be a viable mechanism for the next generation of antiarrhythmic therapy.

The mechanism by which VT increases the propensity of conversion to ventricular fibrillation is not currently known; however, it is possible that the energetic demand of high-rate VT in an already damaged heart accentuates the scar-mediated arrhythmogenic substrate.[3] The longer that the VT continues, the greater the degree of energy consumption and substrate accentuation such that after a critical time point, ventricular fibrillation and asystole are the natural progression.[4]

It is for this reason that modulation of the myocardium in ischemic cardiomyopathy to prevent VT is a priority. Monomorphic VT can be viewed as an aberrant electrical circuit, enabled by myocardial cellular and scar substrate.[5] Mechanistically, these arrhythmias are typically due to either fixed reentry or functional reentry within diseased myocardium. The circuits perpetuate due to the presence of an "excitable gap" of myocardium demonstrating spatial and temporal conductive properties.[6] Conceptually, these circuits act as a wave, composed of a leading depolarization component (head) and trailing repolarization component (tail) that can perpetuate indefinitely within the confines of the reentrant circuit if unperturbed.

Altering either the conduction velocity of the propagating wavefront or the refractoriness of the recruitable myocardial tail can affect both the inducibility and the sustainability of monomorphic VT. This principle underlies the physiological effect of antiarrhythmic agents used in clinical practice.[7] Arrhythmias that are not amenable to antiarrhythmic agents can be disrupted mechanically with catheter ablation, a procedure in which surviving myocardial cells critical to circuit perpetuation are destroyed.

In this preclinical study, we aimed to determine whether treating surgically infarcted HFrEF rats with an engineered cardiac graft composed of a bioabsorbable biomaterial seeded with fibroblasts would reduce the incidence of inducible monomorphic VT via modulating the repolarization component of the reentrant circuit.

METHODS

Rodent heart failure induction

Adult male Sprague-Dawley rats (Envigo, Indianapolis, IN, USA) 6–8 weeks of age were enrolled in this study under the guidance of Institutional Animal Care and Use Committee-approved protocols at the University of Arizona Animal Care Program (number 14-555) and in compliance with the National Institute of Health's "Guide for the Care and Use of Laboratory Animals". As described in previous publications, HFrEF was induced by permanent left coronary artery ligation.[8]

Rats underwent oropharyngeal intubation and ventilation (Harvard Apparatus, Holliston, MA, USA) and were induced using 3% volatile isoflurane in 100% oxygen before receiving an intraperitoneal saline-based cocktail of ketamine (50 mg/kg)+xylazine (5 mg/kg)+acepromazine (1 mg/kg)+atropine (0.5 mg/kg). A left thoracotomy exposed the heart through the anterior mediastinum such that a 5-0 TiCron (Covidien, Minneapolis, MN, USA) ligature could be tied proximally around the left coronary artery, although difficult to visualize with the unassisted eye. The heart was returned to the chest as epicardial lidocaine and epinephrine were applied. Successful ligation was visually confirmed via immediate blanching of the epicardium in the associated circulatory territory. The chest muscle was sutured in layers using 2-0 silk purse string suture (with maximal lung inflation to remove intrathoracic air) and surgical staples were used to close the skin.

The rats were transferred from the heated surgical table to a warmed surgical pad and maintained on the ventilator until they regained consciousness. For postoperative pain, rats were provided intraperitoneal buprenorphine SR (0.8 mg/kg; Wildlife Pharmaceuticals, Windsor, CO, USA) and maintained on carprofen (5 mg/kg) for up to 72 hours during daily monitoring. Sham-operated rats were selected at random and underwent the same exact surgical approach without coronary artery ligation.

Three weeks after coronary ligation, all rats underwent echocardiographic imaging for study arm assignment. Rats with ejection fractions below 40% were assigned to HFrEF. HFrEF rats were subsequently assigned to either graft treatment or untreated HFrEF at random for an additional 3 weeks. Randomization was performed by the animal surgery team during the second left thoracotomy. The surgeon's assistant would transfer a rat to the surgical table without providing any animal identifiers to the surgeon. Immediately before opening each chest, the animal surgeon would request either a graft for surgical deployment or a sterile syringe of 37°C sterile normal saline for epicardial rinse.

Echocardiography

Three weeks post-surgical myocardial infarction, rats were anesthetized with 1.5% isoflurane in 100% oxygen and laid supine on a warming pad with dorsal paw electrodes. Transthoracic echocardiography was performed with a dedicated rodent echocardiography system (Vevo2100 with 13–25 MHz linear transducer; FUJIFILM VisualSonics, Toronto, ON, Canada) by an operator blinded to the animal's status (infarcted or sham). The operator evaluated the heart with views through the parasternal short and long axis as well as two-chamber apical views to evaluate the anterior, lateral, anterolateral, inferior, and posterior walls.[9] Quantification of a left ventricular (LV) ejection fraction below 40% at 3 weeks post-myocardial infarction qualified enrollment in the HFrEF group. HFrEF rats were then randomly assigned to maintenance in the HFrEF group or treatment in the graft group.

Two or three days prior to the terminal invasive hemodynamic study and electrophysiology study at 6 weeks post-myocardial infarction (3 weeks after treatment with the graft), all rats underwent a final transthoracic echo to quantify left ventricular ejection fraction. The operator remained blinded to rat group identity.

Human dermal fibroblasts

Human dermal fibroblasts were used as previously described.[10] In brief, fibroblasts were isolated from neonatal foreskin dermis and expanded into a master cell bank before storage at −196°C. Fibroblasts were thawed for 4 min in a 37°C water bath before resuspension and plating. Plates were maintained in an incubator at 37°C and 5% CO_2 with media changes occurring every other day. Once plates achieved 90% confluence, fibroblasts underwent trypsin-mediated disassociation and centrifugation in preparation for seeding the woven biomaterial.

Engineered grafts

An uncoated polyglactin-910 biomaterial mesh (Ethicon – Johnson&Johnson, Somerville, NJ, USA) was trimmed to generate 1.6 cm diameter matrices. The matrices were rinsed with 1× phosphate-buffered saline before being cellularized with the fibroblasts. Grafts were cultured at 37°C and 5% carbon dioxide in low-adhesion plates to facilitate fibroblast proliferation. Culture media (Thermo Fisher Scientific, Waltham, MA, USA) was changed every 24 hours until surgical deployment of the graft onto a HFrEF rat's LV epicardium.

Prior to surgical implantation, each graft was removed from the incubator, qualitatively assessed using a low-power microscope, and transferred to a 35-mm dish and rinsed with sterile 1× phosphate-buffered saline. The chest was reopened via left thoracotomy, and one graft was implanted over the infarcted area, bridging healthy myocardium on the anterior interventricular septum to healthy myocardium on the lateral left ventricle. The graft was secured to the host's myocardium using three to four circumferential simple interrupted sutures and one central simple interrupted suture.

In vivo cardiac electrophysiology

Using methods previously published, rats underwent an epicardial cardiac electrophysiology study.[11] In brief, in vivo monophasic action potentials (MAPs) were collected with a tungsten concentric bipolar microelectrode (World Precision Instruments, Sarasota, FL, USA). MAP amplitude was quantified as the electric potential difference between phase 4 and the peak of phase 0. Two-dimensional electroanatomic maps were generated using MAP amplitude, and a normal distribution from a previous study[11] was employed to quantify the cut-off values to distinguish normal myocardium (≥7.2 mV), border tissue (7.2>x≥2.8 mV), and scar tissue (<2.8 mV).

Furthermore, a three-lead ECG was obtained (ADInstruments) while programmed electrical stimulation protocols[12] (MATLAB, Natick, MA, USA) were performed using bipolar needle electrodes (BIOPAC Systems, Goleta, CA, USA) on healthy left ventricular myocardium. Ventricular stimulation was performed at twice diastolic threshold (defined as the minimum voltage necessary to capture the heart when pacing and produce a whole-organ depolarization) with a drivetrain of 8 beats at a heart-rate adjusted interval (approximately 240 ms), followed by a single extrastimulus delivered to induce sustained monomorphic VT. After serially decreasing the single extrastimulus by 10 ms, the longest S1–S2 interval in milliseconds that failed to consistently capture the heart was reported as the ventricular effective refractory period (ERP).

Cardiac histopathology

After the terminal cardiac electrophysiology study at 3 weeks post-graft treatment (6 weeks post-myocardial infarction), all cardiac tissue was excised after being arrested in diastole with 3 mL of intracavitary potassium chloride. Hearts were then perfused retrograde through the ascending aorta on a Langendorff apparatus with 10% neutral buffered formalin. Hearts were primed by removing non-LV tissue and then stored in a glass scintillation vial for 1 day before being transferred into 70% ethanol for storage. Finally, LVs were paraffin embedded, sectioned, and stained with Gomori's Trichrome to distinguish healthy myocardium from scar tissue (Tissue Acquisition and Cellular/Molecular Analysis Shared Resource; University of Arizona Cancer Center, Tucson, AZ, USA).

Data analysis

Data are expressed as mean±SEM. An alpha level of 0.05 was set to assess statistical significance; echocardiographic and electroanatomic mapping differences between groups were assessed by one-way analysis of variance (ANOVA) with the post hoc Holm-Sidak method for all pairwise comparisons. The a priori planned comparison (HFrEF vs HFrEF treated with the graft) was also assessed using two-tailed unpaired Student's t-tests and yielded the exact same relationships as found with the ANOVA+Holm-Sidak. The difference in incidence of inducible monomorphic VT was assessed via Fisher's exact test. No SEM range is provided for the incidence of VT as it is a categorical yes/no result based on at least 15 consecutive premature ventricular complexes on the surface electrocardiogram. The definition of rodent sustained VT is based on a conversion from the clinical definition of 100+ beats/minute for at least 30 s, accounting for the elevated heart rates observed in rats.[13]

RESULTS

Rodent heart failure induction

A graphic of the study timeline has been provided (figure 1). All 29 rats that underwent left coronary artery ligation developed objective heart failure, with documented changes in echocardiographic and invasive hemodynamic variables (online supplemental figures 1 and 2). Sham rats (n=7) were also successfully survived and served as accurate study controls.

Engineered grafts

Culture of the fibroblasts into the biomaterial mesh yielded consistently confluent engineered grafts; the polyglactin-910 mesh fibers and gaps were encapsulated in fibroblast-derived organic extracellular matrix as observed in a previous study from our laboratory.[10] The grafts were easily handled by the animal surgeon for successful implantation without tearing. Multiple simple interrupted epicardial sutures secured the graft and each chest was successfully closed without incident despite it being the second thoracic surgery.

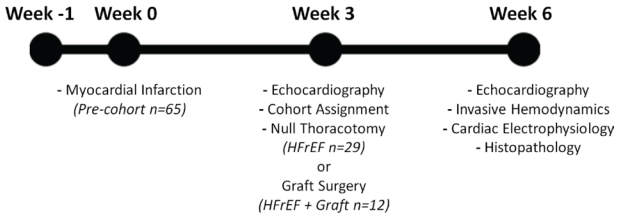

Figure 1 Study timeline. A graphic describing the study timeline with cohort sizes. All rats were acclimated on arrival for 1 week (week –1) before myocardial infarction (week 0). Sham rats underwent a left thoracotomy without left coronary artery ligation to serve as appropriate surgical controls. A minority of infarcted rats did not survive the acute ischemic event. Left ventricular ejection fraction below 40% was confirmed via echocardiography (week 3) for all infarcted pre-cohort rats before randomized cohort assignment to either HFrEF (n=29) with a null thoracotomy or HFrEF+graft (n=12). All rats were survived (week 6) for a final echocardiographic study, invasive hemodynamic assessment, and cardiac electrophysiologic evaluation. Finally, cardiac tissue was harvested for histopathology.

Echocardiography

Six-week echo data (figure 2) revealed a significantly lower left ventricular ejection fraction (EF) for HFrEF compared with sham rats (33±2 vs 76%±3%, p<0.0001). HFrEF rats treated with the graft (n=12) had no change in EF compared with untreated HFrEF rats (39±5 vs 33%±2%, p=0.2549).

In vivo cardiac electrophysiology

Two-dimensional electroanatomic maps were generated using MAP amplitude. Quantification of the amount of scar tissue, relative to the entire mapped area (figure 2), revealed a physiologically negligible amount of scar tissue for sham rats (0.3%±1.5%) and a substantial amount of scar tissue for HFrEF rats (33.0±2.4 vs 0.3%±1.5%, p<0.0001). Treatment of HFrEF rats with the graft showed no difference in MAP amplitude-derived scar area (25.2±6.6 vs 33.0%±2.4%, p=0.4310) compared with the HFrEF group.

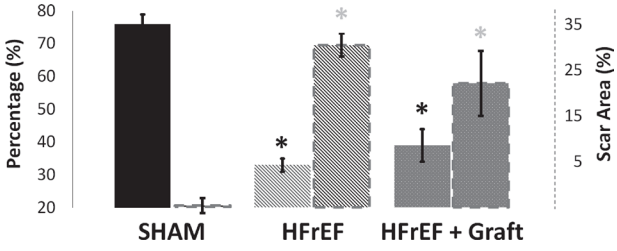

Figure 2 Ejection fraction and scar surface area. Transthoracic echocardiography (*left axis, solid bars*) at the study endpoint (6 weeks post-myocardial infarction, 3 weeks post-graft therapy) revealed a normal left ventricular ejection fraction (EF) for sham (n=7), a significantly decreased EF (33%±2% vs 76±3%, p<0.0001) for the HFrEF group (n=27), and a comparable EF (39%±5% vs 33±2%, p=0.2549) for the graft-treated HFrEF group (n=13). Furthermore, epicardial monophasic action potential amplitude was used to create a two-dimensional color map of the epicardium (maps shown in figure 3), which revealed a large region of scar tissue in the vascular territory of the left coronary artery in HFrEF rats (n=29; *right axis, dashed bars*). This scar area was not observed in sham rats (n=7; 33.0%±2.4% vs 0.3±1.5%, p<0.0001), and graft-treated rats (n=12) exhibited a slightly smaller percentage of scar area (25.2%±6.6% vs 33.0±2.4%, p=0.4310). * denotes statistical significance vs sham.

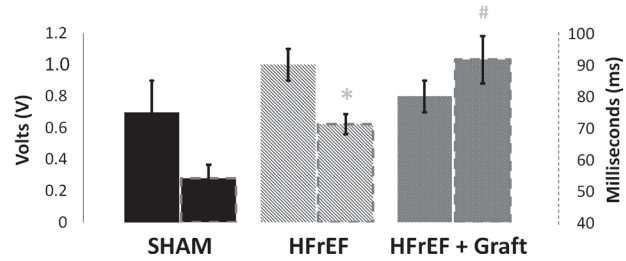

Figure 3 Capture threshold and effective refractory period. Pacing of the heart with an epicardial electrode revealed an acceptable capture threshold (*left axis, solid bars*) for sham rats (n=7) that had no cardiac abnormalities. This value increased to a non-significant degree with onset of HFrEF (n=29; 1.0±0.1 vs 0.7±0.2 V, p=0.1430). Graft-treated HFrEF rats (n=12) elicited a comparable capture threshold (0.8±0.1 vs 1.0±0.1 V, p=0.3220), with no statistically significant relationships found for any of the three groups. In addition, programmed electrical stimulation drivetrains with eight equidistant S1 stimuli and a single premature S2 stimulus revealed a short effective refractory period (ERP; *right axis, dashed bars*), defined as the longest S1(8)–S2 interval that fails to capture the heart and produce a global depolarization, for the sham group (n=7). This value increased with onset of HFrEF (n=28; 68±3 vs 53±4 ms, p=0.0191) and further increased with graft therapy (88±7 vs 68±3 ms, p=0.0029). Graft-treated HFrEF rats (n=12) exhibited the longest ERP. */# denotes statistical significance vs SHAM/HFrEF, respectively.

Capture threshold was assessed quantitatively (figure 3). Sham rats exhibited a capture threshold of 0.7±0.2 V; however, comparison with the HFrEF group yielded no difference (1.0±0.1 vs 0.7±0.2 V, p=0.1430), nor did the comparison between HFrEF and the graft-treated HFrEF group (0.8±0.1 vs 1.0±0.1 V, p=0.3220).

Utilization of a programmed electrical stimulation protocol with a single premature stimulus (S1–S2) enabled quantification of the LV ERP (figure 3). The sham group had the shortest ERP (53±4 ms), which prolonged in HFrEF (69±3 vs 53±4 ms, p=0.0191), and further prolonged in the graft-treated HFrEF group (88±7 vs 69±3 ms, p=0.0029).

Finally, the S1–S2 drivetrain was also used to assess the incidence of inducible monomorphic VT in rats that had undergone MAP electroanatomic mapping (figures 4 and 5). As expected, sham rats exhibited no VT whereas HFrEF rats experienced successful induction to VT at a rate of 72% (72 vs 0%, p=0.0394). HFrEF rats treated with the graft had no change in the incidence of VT compared with untreated HFrEF rats (42 vs 72%, p=0.0834).

Qualitative histopathological data supported no difference in scar burden between HFrEF and graft-treated HFrEF hearts, and revealed transmural infarcts with foci of residual biomaterial in graft-treated HFrEF hearts (figure 6).

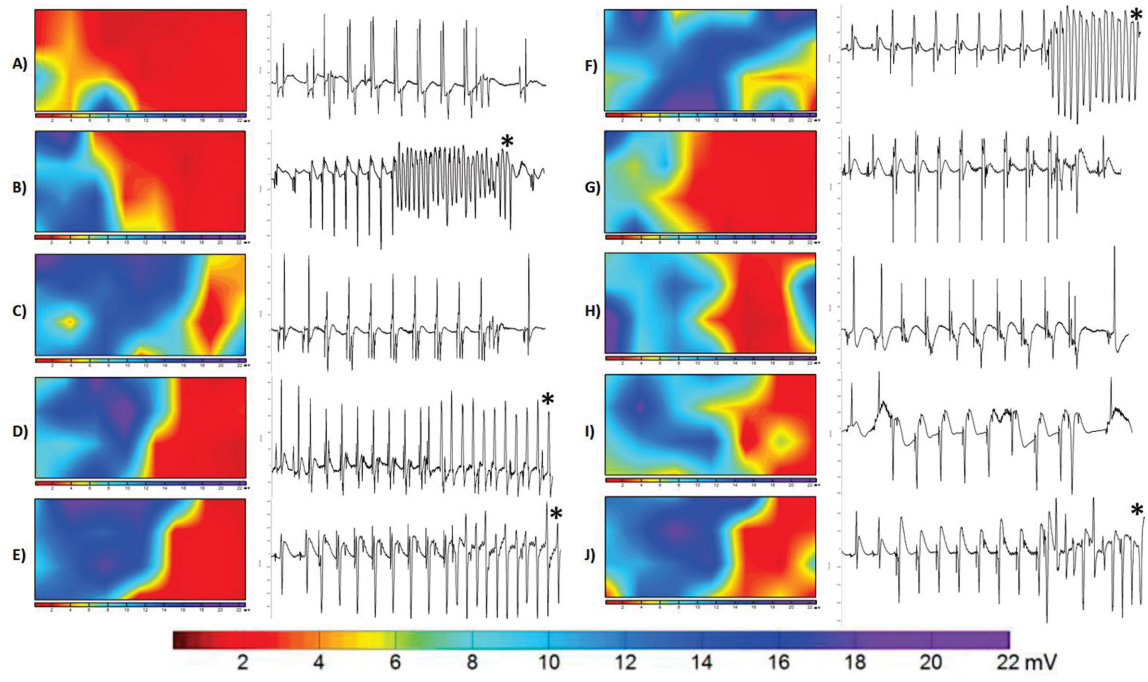

Figure 4 Monophasic action potential amplitude electroanatomic maps with surface electrocardiograms. Interpolation of monophasic action potential amplitudes into two-dimensional electroanatomic color maps from the anterolateral epicardial surface of 10 HFrEF rats treated with the graft (panels A–J). The color bar is depicted below each color map in millivolts, and a larger color bar is located at the bottom of the figure. For each rat, electrical infarct represented by red is seen in the vascular territory of the left coronary artery while healthy tissue can be seen to its left. In addition, beside each color map is a surface ECG tracing during programmed electrical stimulation to induce monomorphic ventricular tachycardia (VT). Induction protocols that successfully resulted in sustained monomorphic VT have been denoted with an asterisk (*).

DISCUSSION

This preclinical study aimed to evaluate the effects of a fibroblast-containing engineered graft on ventricular electrophysiologic properties in a rodent model of ischemic cardiomyopathy. It is important to note that this model represents ischemic cardiomyopathy following an acute anterior wall myocardial infarction and not cardiomyopathy in general. After a treatment timeline of 3 weeks, for a total study timeline of 6 weeks (figure 1), graft-treated HFrEF rats were found to have an unchanged LV ejection fraction (figure 2), a comparable percentage scar (figure 2), an unchanged capture threshold (figure 3), a prolonged ERP (figure 3), and no statistically significant change in the incidence of inducible monomorphic VT (figures 4 and 5). Histopathology revealed transmural infarcts for all rats that underwent permanent left coronary artery ligation and foci of residual biomaterial in graft-treated HFrEF rat hearts (figure 6).

Although LV ejection fraction decreases as expected between the sham study arm and the HFrEF study arm, the maintenance of a low LV ejection fraction with the graft treatment was anticipated as the grafts did not contain any contractile cells such as ventricular cardiomyocytes. This finding is also consistent with a 2010 study from our laboratory using the same engineered graft.[10] Interestingly, the only echocardiographic parameter found to change with graft treatment was LV posterior wall thickness in diastole (online supplemental figure 1). We postulate that the angiogenic effect of this graft therapy[10] may have increased retention of hibernating myocardium and thus decreased global compensatory hypertrophy.

Capture threshold, also known as diastolic threshold, can be viewed as a measurement of the myocardium's content of repolarized excitable cardiomyocytes, assuming stable electrode contact with the myocardium. A low capture threshold correlates with the presence of cardiomyocytes that are repolarized and excitable. In this study, we report no significant changes in capture threshold between the three study arms (figure 4), suggesting either a lack of sufficient assay sensitivity to detect any changes in resistance between the groups or possibly no difference in the aforementioned fibroblast parameters that are likely to modulate electrical resistance.

Finally, graft-treated HFrEF rats exhibit no statistically significant change in the incidence of inducible monomorphic VT (figure 5). Prolongation of the LV ERP (figure 5) at local epicardial sites could theoretically lead to elimination of the excitable gap necessary for the reentry circuit of monomorphic VT, similar to what is accomplished with ablation.[14] Although the graft treatment resulted in no significant change in the incidence of VT, previous investigators have cautioned the sole reliance on p values to define physiologic significance.[15] It is on this basis that we highlight the physiological relevance of a 30-point decrease in the incidence of inducible monomorphic VT in graft-treated HFrEF rats. This finding is hypothesis-generating with regard to reproducibility and clinical feasibility.

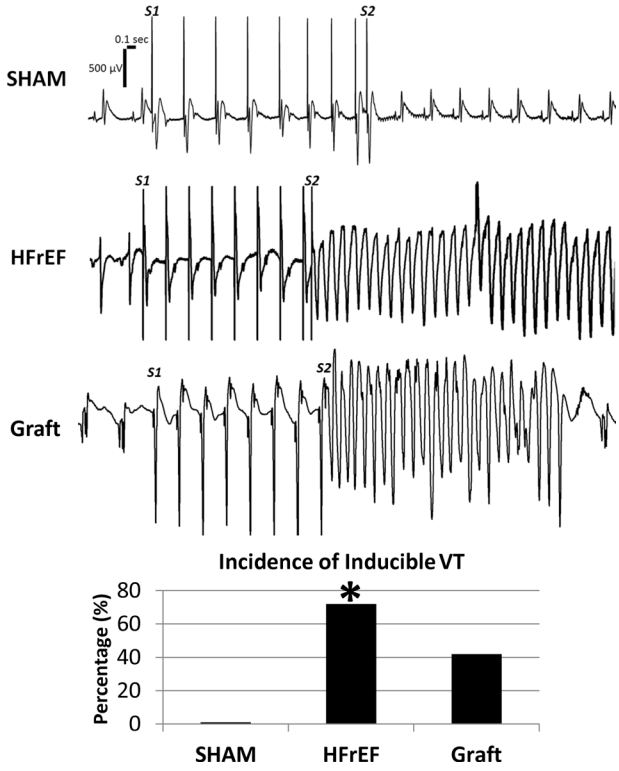

Figure 5 Incidence of inducible monomorphic ventricular tachycardia. S1–S2 drivetrains were unsuccessful in inducing monomorphic ventricular tachycardia (VT) in any sham rats; however, the majority of HFrEF rats were able to be induced, with representative tracings shown (72% vs 0%, p=0.0394). HFrEF rats treated with the graft were able to be successfully induced into VT to a lesser degree, although this did not reach statistical significance (42% vs 72%, p=0.0834). * denotes statistical significance vs sham.

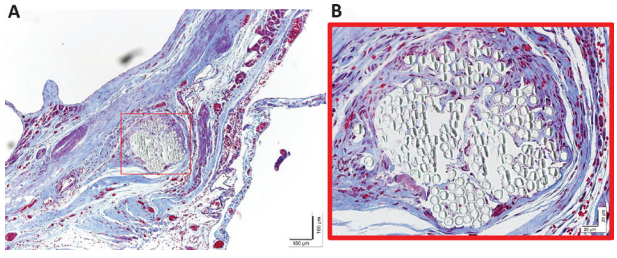

Figure 6 Cardiac histopathology. After the terminal cardiac electrophysiology study at 3 weeks post-graft treatment (6 weeks post-myocardial infarction), cardiac tissue was harvested and stained with Gomori's Trichrome. Myocardial tissue and red blood cells stain red in color while collagen bundles and connective tissue stain blue in color. As expected, this representative graft-treated HFrEF rat heart exhibits a large transmural infarct with dense connective tissue presence (A, ×10 magnification). Thin streaks of surviving cardiomyocytes can be observed near the epicardium and endocardium. Interestingly, foci of residual biomaterial with septa of myocardial tissue can be observed throughout the infarcted myocardium (B, ×40 magnification).

However, it is equally important to note that ERP prolongation can also be a negative prognostic factor, predisposing to malignant ventricular tachyarrhythmias by way of action potential duration heterogeneity between the endocardium and epicardium, or action potential duration prolongation leading to triggered activity or Torsades de Pointes. Nonetheless, we did not observe any afterdepolarization-related ectopy, Torsades de Pointes, nor ventricular fibrillation. The graft may be antiarrhythmic by locally prolonging ERP and thus providing a physiologic block to reentrant VT. This could be accomplished locally without global myocardial effects.

A previous study from our laboratory investigated whether a cardiac graft consisting of human induced pluripotent stem cell–derived cardiomyocytes, in addition to the fibroblasts and biomaterial described in this present study, had any electrophysiologic effect on the infarcted ventricle.[16] The study found a decrease in the incidence of inducible VT; however, no mechanism was revealed. This present study was designed as a follow-up "sufficiency" study, where we sought to evaluate if the fibroblast-biomaterial graft alone was sufficient to reproduce the same electrophysiologic effect. Furthermore, in addition to reporting a physiologically relevant difference in the incidence of inducible monomorphic VT with this fibroblast-biomaterial graft, we also provide evidence of the potential mechanism: local prolongation of ventricular effective refractory period, disrupting the reentrant circuit necessary for monomorphic VT propagation.

Interestingly, the synthetic polyglactin-910 biomaterial used in this graft preparation has never been evaluated alone for its electrical effects on the cardiac system. It has been characterized as a negatively charged insulating biomaterial with a resistance sufficiently strong to stop current flow at up to ~90 V of direct current. This degree of electrical insulation would likely overcome any natural cardiac electrophysiologic process. Thus, theoretically it is possible that the effects described in this study could be due, at least in part, to the capacitive effects of this biomaterial. Whether polyglatcin-910 alone is necessary and/or sufficient to recapitulate these cardiac electrophysiology findings is of potential interest. To date, no synthetic biomaterial has been shown to decrease arrhythmia generation in the clinical setting, although biomaterial investigators have shown enhanced conduction through infarcted myocardium using carbon nanotubes[17] and created electroconductive injectable hydrogels using gold nanorods.[18]

Limitations

The permanent coronary artery occlusion methodology used to create HFrEF often creates transmural infarcts in these rats. It is probable that the electrical circuit necessary for the reentrant monomorphic ventricular tachycardia involves the epicardial surface, given the transmural nature of the scar tissue. Thus, the electrophysiologic effects observed 3 weeks after surgical deployment of the engineered graft on the epicardium may require that the electrical circuit reaches the epicardium. Should the infarct pattern be subendocardial or intramural and not transmural, as is often the case clinically, it is unclear whether this engineered graft would exert an equally efficacious effect.

One limitation of this study is the selected animal model. Compared with human cardiomyocytes, rat cardiomyocytes are known to express different isoforms and relative abundances of ion channels, which gives rise to their shorter action potential durations and increased heart rates, ability to withstand large transmural myocardial infarctions, and decreased susceptibility to ventricular fibrillation.[12] These differences, and potentially other currently unknown differences, may limit future translation of this graft therapy to human patients with HFrEF, and warrant comprehensive investigation with respect to discrepancies between rat and human cardiac electrophysiology and ischemic remodeling.

Furthermore, the age of rats at surgical infarct is younger than what is observed clinically in patients with atherosclerotic acute coronary events. Nonetheless, on using a rat-to-human age converter,[19] it becomes apparent that the rats employed in this study were closer to the human equivalent of middle age, which is reasonable for modeling structural heart disease.

CONCLUSION

This evaluation of an engineered graft composed of human dermal fibroblasts in a bioabsorbable biomaterial to treat arrhythmogenesis in ischemic cardiomyopathy may be the first of its kind. This epicardial biomaterial therapy in HFrEF rats yielded an unchanged ejection fraction, a comparable percentage scar, an unchanged capture threshold, but a prolonged effective refractory period. Should this increase in refractoriness locally target the substrate-mediated reentrant circuit, this graft could decrease myocardial susceptibility to monomorphic ventricular tachycardia in HFrEF.

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Acknowledgements A special thank you to Sherry Daugherty, Maribeth Stansifer, Mary Kaye Pierce, Grace Gorman, and Dr. Joseph Bahl for their advice and technical assistance.

Contributors IRC—electrophysiology investigator, performed data acquisition and analyses. MH—clinical cardiac electrophysiologist, assisted in data interpretation. TM—clinical cardiac electrophysiologist, assisted in data interpretation. JWK—biomedical engineer, assisted in creation of biomaterial. EJ—heart failure cardiologist, assisted with rodent model and echocardiography. SG—senior physician scientist, provided scientific guidance. JJL—research scientist, provided biomaterial and scientific guidance.

Funding This work was supported by the NHLBI T32 HL007249-43, WARMER Research Foundation, Sarver Heart Center, and the University of Arizona.

Competing interests JWK, SG, and JJL have disclosed a financial interest in Avery Therapeutics, Inc. to the University of Arizona. In addition, the University of Arizona has a financial interest in Avery Therapeutics, Inc. These interests have been reviewed and are being managed by the University of Arizona in accordance with its policies on outside interests. All other authors have no relevant conflicts to disclose.

Patient consent for publication Not required.

Provenance and peer review Commissioned; externally peer reviewed.

Data availability statement All data relevant to the study are included in the article or uploaded as online supplemental information.

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