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A Lanyu Pig Model of Ischemia-Reperfusion Myocardial Infarction for Cardiac Regeneration

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Article

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Abstract

Background

Myocardial infarction (MI) remains a leading cause of heart failure worldwide. Robust large animal models that mimic human physiology are necessary to develop regenerative therapies such as cell transplantation. However, conventional porcine models (e.g., Yorkshire and Göttingen minipigs) are limited by high perioperative mortality, primarily from lethal ventricular arrhythmias, and rapid somatic growth, which hinders long-term study. We hypothesized that the Lanyu pig, a genetically distinct miniature breed, would offer a superior translational platform with better survival and physiological stability than do the conventional models.

Methods

Catheter-based ischemia-reperfusion injury was induced in 17 adult Lanyu pigs through 90-min balloon occlusion of the midsegment of the left anterior descending artery. An insertable cardiac monitor was implanted 3 days before surgery for continuous rhythm tracking. Cardiac function was comprehensively assessed at baseline, immediately after MI, and on Day 14 after MI through transthoracic echocardiography and pressure-volume loop analysis. Intramyocardial injections were administered on Day 14 to simulate the subacute phase and validate the feasibility of therapeutic delivery.

Results

The Lanyu pig model achieved a procedural survival rate of 94.1% (16/17), significantly higher than the historical rates for other breeds. Post-MI assessments confirmed substantial functional decline: left ventricular ejection fraction decreased from 69.4% to 40.0% ($p < 0.001$), and the slope of end-systolic pressure-volume relationship maximum rate of pressure rise decreased from 5.6 to 1.9 mmHg/mL ($p < 0.001$). Although transient arrhythmias were observed, no sustained malignant arrhythmias occurred. Infarcts were consistent, and the animals tolerated the surgical and follow-up procedures well.

Conclusion

Our Lanyu pig MI model demonstrated high survival rates, reproducible cardiac dysfunction, and physiological similarity to humans. This model overcomes key limitations of conventional porcine models, offering a robust and ethically refined tool for preclinical evaluation of novel cardiac regenerative therapies.

Novelty and Significance

What is known?

- Large-animal models are essential to bridge the translational gap in cardiac regeneration research, but conventional porcine models (e.g., Yorkshire pigs and Göttingen minipigs) are limited by high perioperative mortality due to malignant arrhythmias.
- Rapid somatic growth in conventional porcine models complicates the long-term follow-up necessary for regeneration studies.
- Current heart failure treatments delay disease progression but fail to restore lost myocardium, underscoring the need for reliable preclinical platforms to test novel cell-based therapies.

What new information does this article contribute?

- We established a highly reproducible myocardial infarction model by using the Lanyu pig, an indigenous Taiwanese miniature breed; compared with conventional models, our model demonstrated improved survival rates following ischemia-reperfusion injury.
- Lanyu pigs exhibit human-like coronary anatomy and stable genetic characteristics, with a reduced risk of fatal ventricular arrhythmias during the acute postinfarction phase.
- Lanyu pigs in our cohort sustained significant, quantifiable left ventricular systolic dysfunction and hemodynamic impairment (confirmed through pressure-volume loop analysis) up to 14 days after myocardial infarction, providing a stable window for evaluating regenerative interventions.

Overall, our model combines procedural feasibility, low mortality, and continuous rhythm monitoring, thereby supporting the evaluation of the safety and efficacy of regenerative therapies in a clinically relevant large-animal model.

Introduction

Ischemic heart disease remains a key cause of mortality worldwide, often progressing from coronary atherosclerosis to myocardial infarction (MI) and end-stage heart failure.[1, 2] Despite advances in pharmacological and interventional therapies, current treatments are largely palliative, delaying disease progression without regenerating lost cardiomyocytes or reversing fibrotic scarring.[1, 3, 4] Thus, approximately one-third of MI survivors have heart failure within 5 years, underscoring the urgent need for regenerative strategies.[5]

Cell-based therapies, such as treatment with human pluripotent stem cell-derived cardiomyocytes, represent a promising strategy for remuscularizing the infarcted heart.[3, 6, 7] However, successful clinical translation relies heavily on preclinical validation in large-animal models that closely replicate human anatomy and physiology.[6, 8, 9] Pigs are widely regarded as the gold standard for animal models because of their heart size, coronary circulation, and electrophysiological properties are similar to those of humans.[6, 10, 11] Nevertheless, the use of conventional porcine models, such as Yorkshire pigs and Göttingen minipigs, is limited by certain challenges. These models are highly susceptible to ischemia-

induced malignant arrhythmias, which lead to high perioperative mortality rates (17% – 40%).[10–13] Furthermore, their rapid growth complicates the long-term follow-up necessary for regeneration studies.[11, 14]

In this study, we established a novel translational platform by using the Lanyu pig, a miniature breed indigenous to Taiwan. Our findings revealed the unique resistance of Lanyu pigs to fatal arrhythmias following ischemia-reperfusion (IR) injury, which resulted in improved survival. By combining advanced hemodynamic assessment through pressure-volume (PV) loop analysis[12] with continuous rhythm monitoring,[12, 15] we validated the Lanyu pig as a robust, clinically relevant model for evaluating the efficacy and safety of next-generation cardiac regenerative therapies.

Methods

Data Availability

The data that support the findings of this study are available from the corresponding author upon reasonable request. Tse-Wei Chen had full access to all study data and takes responsibility for the integrity of the data and the accuracy of data analysis.

Ethical Approval and Animals

All experimental procedures and protocols were approved by the Institutional Animal Care and Use Committee of the National Center for Biomodels, Taiwan (IACUC approval No.: NLAC-111-M-032 and NLAC-112-M036). The study sample comprised 17 adult Lanyu pigs (11 female and 6 male pigs; mean weight: 34.5 kg; mean age: 7.7 months). The animals were obtained from the Taitung District, Eastern Region Branch, Taiwan Livestock Research Institute, Ministry of Agriculture.

Study Protocol (Fig. 1)

To establish baseline rhythm profiles, insertable cardiac monitors (Reveal LINQ™; Medtronic, Minneapolis, MN, USA) were subcutaneously implanted at 3 days before MI induction (Day – 3). On Day 0, MI was induced through catheter-based IR injury. Functional assessments (transthoracic echocardiography [TTE] and PV loop analysis; Fig. 2) were conducted at baseline (Day 0), immediately after MI, and on Day 14 after MI. On Day 14, thoracotomy was performed to administer an intramyocardial injection, simulating therapeutic delivery (Fig. 3).

Induction of MI Through IR Surgery

On Day 0, catheter-based IR surgery was performed by a qualified interventional cardiologist to induce MI in Lanyu pigs. Percutaneous coronary catheterization was performed with the pig under general anesthesia (Fig. 4). During this procedure, a balloon catheter was advanced to the midsegment of the

left anterior descending (LAD) artery and inflated for 90 min to induce ST-elevation MI. The LAD artery is among the three major coronary branches and supplies a substantial portion of the anterior myocardium.[16] Occlusion of the LAD artery in porcine models leads to transmural infarcts with histopathological and functional changes closely resembling those observed in human MI. Therefore, porcine models represent a robust and translationally relevant large-animal model. After 90 min of ischemia, the balloon was deflated to allow reperfusion of the LAD territory. Cardiac function was evaluated both before and after MI through TTE and PV loop analysis to quantify left ventricular (LV) systolic performance.

Step-by-Step Coronary Catheterization for LAD Occlusion

To induce IR injury, the following catheter-based procedure was performed with the pig under general anesthesia:

1. *Anesthesia and monitoring*: To induce IR injury, each pig was anesthetized using a combined intramuscular injection of Zoletil/Telazol (5 mg/kg) and xylazine (2.2 mg/kg). Anesthesia was maintained with inhaled isoflurane (0.2% – 6%). The surgical field at the neck or groin was prepared following standard sterile techniques. Vital parameters, such as oxygen saturation, heart rate, arterial blood pressure, body temperature, and end-tidal CO_2, were continuously monitored throughout the procedure.[6]
2. *Perioperative management*: Fentanyl and buprenorphine were used for perioperative and postoperative analgesia. Systemic anticoagulation was achieved with heparin, targeting an activated clotting time of 250–350 s to prevent thrombus formation. Lidocaine was administered prophylactically to prevent or manage ventricular arrhythmias during cardiac procedures and reduce the risks of ventricular tachycardia (VT) and fibrillation. The dosing regimen included an intravenous bolus of 1.0 mg/kg, followed by continuous infusion at 20 μg/kg/min.
3. *Vascular access*: A small incision was made to expose either the carotid or the femoral artery. A vascular sheath was introduced following guidewire placement.
4. *Catheter insertion and hemodynamic assessment*: A 6F Judkins right coronary catheter was advanced toward the LV over a J-tip guidewire. Then, a PV conductance catheter was inserted to facilitate baseline hemodynamic measurement.
5. *Coronary engagement*: The Judkins right coronary catheter was maneuvered near the left coronary ostium. Because complete engagement was not always achievable, a floating wire was used to navigate to the distal LAD artery. If unsuccessful, catheterization was attempted with a 6F Judkins left coronary catheter.
6. *LAD wiring and balloon positioning*: A coronary guidewire (0.014 inches) was advanced into the LAD artery. An angioplasty balloon catheter (2.5 × 15 mm) was delivered over the wire and positioned distal to the first diagonal branch, targeting the midsegment of the LAD artery.

7. *Balloon inflation and LAD occlusion*: The balloon was inflated to 10 atm to achieve complete occlusion of the LAD artery and preserve perfusion to the first diagonal branch. Coronary angiography was performed to confirm successful occlusion.
8. *Ischemia duration and safety precautions*: Balloon occlusion was maintained for 90 min. ST-segment elevation typically occurred within 5 min, confirming myocardial ischemia. Emergency resuscitation equipment and antiarrhythmic agents were made readily available to manage any malignant ventricular arrhythmias.
9. *Reperfusion*: After the occlusion period, the balloon was gradually deflated. Coronary angiography confirmed restoration of LAD blood flow and subsequent reperfusion - .

Feasibility of Intramyocardial Cell Injection

On Day 14, left thoracotomy was performed with the pig under general anesthesia for targeted intramyocardial delivery of stem cells or vehicle control. After heart exposure, five injection sites were identified: two within the central infarct zone and three along the peri-infarct border (Fig. 3). Injections were administered using 1-mL insulin syringes (BD Biosciences) fitted with 29G needles to ensure precision and minimize tissue trauma. All cell- or vehicle-containing syringes were kept on wet ice and used within 30 min of preparation to maintain cell viability. All surgical procedures were performed by experienced cardiovascular surgeons. Postoperative care included analgesia and continuous monitoring to ensure recovery. This standardized injection protocol allowed for consistent delivery of cell therapy across all experimental groups.

Hemodynamic and Functional Assessments

LV function was evaluated using a GE Vivid-i ultrasound system. For each view, images were acquired over at least three consecutive cardiac cycles to minimize beat-to-beat variability. The LV end-diastolic dimension (LVEDD) and end-systolic dimension (LVESD) were measured at the mid-ventricular (papillary muscle) level in the short-axis view. The LV ejection fraction (LVEF) was calculated following the M-mode method. Load-independent analysis was performed using a PV conductance catheter (Millar Instruments). The inferior vena cava was transiently occluded to assess contractility (end-systolic PV relationship [ESPVR] and maximum rate of pressure rise in the LV [dP/dt_{max}]) and relaxation.

Statistical Analysis

Statistical analyses were performed using IBM SPSS Statistics version 22.0 (IBM Corp., Armonk, NY, USA). All data are presented as mean ± standard deviation values. A nonparametric Friedman test was performed to compare results across time points. For subsequent post hoc pairwise comparisons, a Wilcoxon signed-rank test with Bonferroni correction was performed to account for Type I error. A p value of < 0.05 indicated statistical significance.

Results

Lanyu Pigs Exhibit Improved Survival Following MI

Of the 17 animals included in this study, 16 survived the IR procedure and subsequent 14-day observation period. Only one animal died of acute heart failure shortly after reperfusion, corresponding to a mortality rate of 5.9%. This rate is markedly lower than that typically reported in the Yorkshire pig and Göttingen minipig models.

MI Induces Substantial Functional Decline

TTE confirmed successful induction of MI. A marked, sustained decline was noted in systolic function by Day 14 compared with the baseline value (Table 1). LVEF decreased from 69.4% to 40.0% ($p < 0.001$; Fig. 5A), stroke volume decreased from 43.7 to 22.0 mL ($p < 0.001$; Fig. 5B), fractional shortening decreased from 38.3% to 19.1% ($p < 0.001$; Fig. 5C), interventricular septal thickness increased from 0.78 to 0.89 cm ($p = 0.045$; Fig. 5D), posterior wall thickness at end-diastole increased from 0.65 to 0.74 cm ($p = 0.005$; Fig. 6A), LV internal dimension during systole increased from 2.34 to 2.89 cm ($p < 0.001$; Fig. 6B), and end-systolic volume increased from 19.3 to 32.9 mL ($p < 0.001$; Fig. 6C). These findings confirmed substantial systolic dysfunction and ventricular remodeling following MI, validating the effectiveness of our model for the investigation of postinfarct cardiac injury.

Table 1
Transthoracic echocardiography revealed a marked reduction in left ventricular function following ischemia-reperfusion injury, confirming successful model establishment

Interventricular septum thickness (cm)	**Day 0 (Pre MI)**	**Day 0 (Post MI)**	**Day 14**
	0.78 ± 0.13	**0.89 ± 0.14 (*p* = 0.045)**	**0.95 ± 0.20 (*p* = 0.007)**
Posterior wall thickness at end-diastole (cm)	0.65 ± 0.11	0.74 ± 0.11 (*p* = 0.005)	0.67 ± 0.09 (*p* = 0.92)
Left ventricular internal diameter during diastole (cm)	3.81 ± 0.34	3.58 ± 0.43 (*p* = 0.063)	3.49 ± 0.46 (*p* = 0.027)
Left ventricular internal diameter during systole (cm)	2.34 ± 0.25	2.89 ± 0.36 (*p* < 0.001)	2.83 ± 0.39 (*p* < 0.001)
End-diastolic volume (mL)	63.03 ± 13.06	54.89 ± 15.62 (*p* = 0.071)	53.48 ± 17.04 (*p* = 0.072)
End-systolic volume (mL)	19.33 ± 4.77	32.91 ± 9.92 (*p* < 0.001)	31.51 ± 12.37 (*p* < 0.001)
Stroke volume (mL)	43.67 ± 10.08	22.02 ± 6.11 (*p* < 0.001)	22.20 ± 5.45 (*p* < 0.001)
Fraction shortening (%)	38.28 ± 4.26	19.08 ± 2.01 (*p* < 0.001)	20.18 ± 2.03 (*p* < 0.001)
Ejection fraction (%)	69.43 ± 5.37	40.02 ± 3.90 (*p* < 0.001)	42.11 ± 3.78 (*p* < 0.001)
Data are presented as mean ± standard deviation values.			
N = 17; *p* values indicate comparisons with baseline measurements (before MI). **Abbreviation:** MI, myocardial infarction.			

Hemodynamic Validation Through PV Loop Analysis

Invasive PV loop analysis corroborated the TTE findings, revealing load-independent evidence of contractile failure (Table 2). LVEF decreased from 66.1% to 46.2% ($p < 0.001$; Fig. 6D), slope of ESPVR decreased from 5.6 to 1.9 mmHg/mL ($p < 0.001$; Fig. 7A), and dP/dt_{max} decreased from 2977.4 to 2164.1 mmHg/s ($p = 0.013$; Fig. 7B). These PV loop parameters independently validated the TTE evidence of

impaired systolic function and contractility, reinforcing the reliability of our model for the investigation of postinfarct cardiac dysfunction.

Table 2
Pressure-volume loop analysis revealed a marked decline in left ventricular contractile performance following ischemia-reperfusion injury, supporting the functional effect of MI

Cardiac output (mL/min)	**Day 0 (Pre MI)**	**Day 0 (Post MI)**	**Day 14**
	3927.3 ± 1544.8	**4197.7 ± 1979.4 (*p* = 0.48)**	**3202.9 ± 1544.1 (*p* = 0.064)**
Stroke volume (mL)	40.6 ± 19.5	37.4 ± 18.5 (*p* = 0.35)	30.6 ± 15.0 (*p* = 0.009)
Heart rate (bpm)	101.9 ± 20.6	114.5 ± 15.4 (*p* = 0.004)	110.8 ± 24.1 (*p* = 0.24)
Stroke work (mmHg*mL)	3606.5 ± 1781.5	3006.6 ± 1283.6 (*p* = 0.16)	2365.8 ± 1399.4 (*p* = 0.004)
Maximum rate of LV pressure rise (mmHg/s)	2977.4 ± 1235.6	2164.1 ± 518.6 (*p* = 0.013)	2738.5 ± 952.8 (*p* = 0.61)
Ejection fraction (%)	66.1 ± 7.2	46.2 ± 5.6 (*p* < 0.001)	46.1 ± 9.7 (*p* < 0.001)
End-systolic PV relationship	5.6 ± 1.8	1.9 ± 0.9 (*p* < 0.001)	2.1 ± 0.8 (*p* < 0.001)
Data are presented as mean ± standard deviation values.			
N = 17; *p* values indicate comparisons with baseline measurements (before MI). **Abbreviation:** MI, myocardial infarction.			

Electrocardiographic Characterization and Management of Ventricular Arrhythmias

Continuous LINQ monitoring, together with intraprocedural observation during IR surgery, revealed transient ventricular arrhythmias, particularly during the acute ischemic and reperfusion phases. In total, 152 episodes of VT were recorded. Using the prespecified electrocardiographic criteria, VT was defined as a sequence of ≥ 3 consecutive ventricular premature complexes with a ventricular rate of ≥ 180

beats/min and a QRS duration of ≥ 60 ms.[3] Notably, when VT occurred during IR surgery, the first procedural step was to confirm that the guiding catheter was not deeply engaged in the left coronary ostium and thereby compromising left coronary perfusion. When catheter-induced coronary hypoperfusion was excluded, VT episodes were corrected with lidocaine bolus administration and amiodarone infusion as needed. All VT episodes were self-limiting and nonfatal, and no sustained malignant ventricular arrhythmias were observed beyond the acute peri-procedural phase.

Discussion

To the best of our knowledge, this study is the first to establish the Lanyu pig as a feasible large-animal model for MI, demonstrating the baseline physiological stability required for rigorous cardiac regeneration research. Compared with the conventional Yorkshire pig and Göttingen minipig models, the Lanyu pig model exhibited superior physiological tolerance and survival following MI induction. High perioperative mortality in porcine models is frequently attributed to malignant ventricular arrhythmias, with reported rates reaching 37%.[8, 17, 18] By contrast, the procedural mortality rate in our sample was only 5.9% (1 of 17 pigs). This resilience highlights the Lanyu pig model as a robust, translationally relevant platform for preclinical intervention studies (Fig. 8). The precise biological mechanisms conferring this unique arrhythmia resistance remain to be elucidated, underscoring a key avenue for future mechanistic studies.

The duration of coronary occlusion is a critical determinant of infarct size and procedural survival, with longer ischemia periods typically resulting in larger myocardial damage and higher mortality risks. For instance, a 90-min occlusion leads to more extensive infarction than does a 45-min occlusion.[11] We selected the LAD artery as the occlusion site to create a substantial infarct area. To reduce procedural mortality, care was taken to preserve patency of the first diagonal branch, ensuring partial perfusion of the anterior LV wall.[14, 19] In addition, prophylactic antiarrhythmic medications and prompt defibrillation protocols were implemented to manage and minimize arrhythmia-related complications during the IR period. These strategies collectively contributed to the high survival rate observed in our Lanyu pig MI model.

Echocardiography enables quantitative evaluation of multiple functional indices, including LVEF, which reflects the percentage of blood ejected from the LV per cardiac cycle. However, the application of echocardiography in porcine models is limited by anatomical challenges, such as the keeled thoracic conformation, which affects image quality and measurement accuracy. LVEF is influenced by several hemodynamic variables, such as preload, afterload, lusitropy, and inotropy; this influence limits the utility of LVEF as a standalone measure of myocardial performance.[20, 21]

Key parameters derived from PV loop analyses offer detailed insights into myocardial contractility and ventricular performance. ESPVR captures the maximal pressure the ventricle can generate at varying volumes during peak systole. The slope of the ESPVR line is considered a relatively load-independent marker of contractile function. A steeper slope indicates greater myocardial contractility, whereas a

more gradual slope suggests impaired contractile performance. The steepest rate of LV pressure increase during isovolumetric contraction can be quantified as $dP/dt_{max,}$ a sensitive index of intrinsic myocardial contractility. Higher dP/dt_{max} values indicate better ventricular performance, whereas lower values indicate myocardial dysfunction. Stroke work measures the amount of mechanical work performed by the ventricle per cardiac cycle, representing the efficiency with which chemical energy is converted into hemodynamic output. A decline in stroke work may indicate myocardial ischemia or diminished contractile capacity. However, because stroke work depends on preload and afterload, it should be interpreted in conjunction with ESPVR and other load-independent indices for accurate assessment of myocardial function.

A further important contribution of the present study is the electrocardiographic characterization of ventricular arrhythmias in the Lanyu pig. Although definitions of VT in large-animal models are often extrapolated from human criteria or other swine breeds, continuous rhythm monitoring in our model enabled a more specific electrophysiological characterization.[3] We found that the upper limit of sinus-rhythm QRS duration in Lanyu pigs was 60 ms; accordingly, ventricular ectopic events, including ventricular premature complexes and VT, were identified by a widened QRS complex of ≥ 60 ms in the appropriate rhythm context (Fig. 9). Establishing this Lanyu pig-specific threshold may improve the accuracy of arrhythmia recognition and facilitate timely management during the acute phase of ischemia-reperfusion injury. In addition, this operational definition provides a practical and standardized reference for future preclinical studies, particularly those evaluating the potential arrhythmogenicity of novel cell-based regenerative therapies.

We performed both TTE and PV loop analysis to characterize cardiac function. Although TTE facilitates noninvasive monitoring, it is limited by its load-dependency. PV loop analysis confirmed that the observed reduction in LVEF was driven by intrinsic contractile failure rather than loading conditions alone. This rigorous validation supports the sensitivity of the Lanyu pig model to detect functional improvements in future regenerative trials.

Limitations

This study has some limitations. Despite its advantages, PV loop analysis requires specialized expertise to ensure accurate data acquisition and interpretation. Precise catheter placement within the LV and meticulous conductance calibration are necessary to obtain reliable hemodynamic measurements. Misplacement of the catheter or calibration errors can lead to major inaccuracies in volume and pressure data, compromising study validity. Therefore, successful implementation of PV loop analysis requires experienced personnel, rigorous physiological monitoring, and adherence to standardized protocols to ensure data fidelity and reproducibility in large-animal cardiovascular studies.

Conclusion

The Lanyu pig represents a robust, clinically relevant, and survival-favorable model for cardiovascular research. Its use can accelerate the translation of novel cardiac regeneration therapies from the bench to the bedside.

Article Information

Abbreviations

- ESPVR: End-systolic pressure-volume relationship
- IR: Ischemia-reperfusion
- LAD: Left anterior descending
- LVEF: Left ventricular ejection fraction
- MI: Myocardial infarction
- PV: Pressure-volume
- TTE: Transthoracic echocardiography
- VT: Ventricular tachycardia

Declarations

Article Information

Funding

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Disclosures

The authors used ChatGPT (OpenAI) for language editing and grammar correction during manuscript preparation. This manuscript was edited by Wallace Academic Editing. All scientific content, analyses, and conclusions were developed and verified by the authors, who take full responsibility for the manuscript.

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Figures

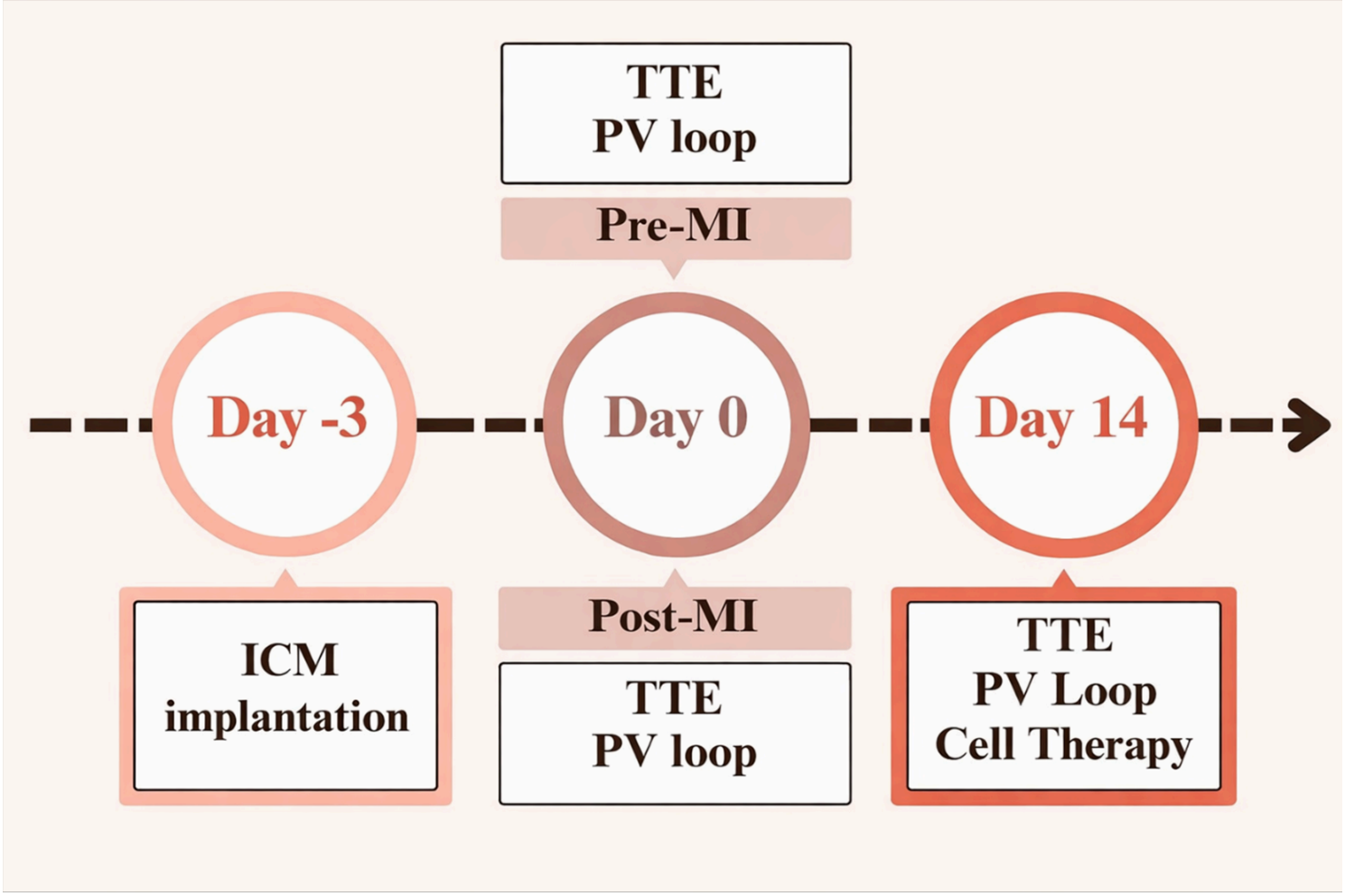


Figure 1

Experimental timeline.

Lanyu pigs were subjected to ischemia-reperfusion surgery on Day 0 to induce MI. ICMs were implanted on at 3 days before surgery for continuous rhythm monitoring. Vehicle or cells were intramyocardially injected at 14 days after MI. **Abbreviations:** MI, myocardial infarction; PV, pressure-volume; ICM, insertable cardiac monitor; TTE, transthoracic echocardiography.

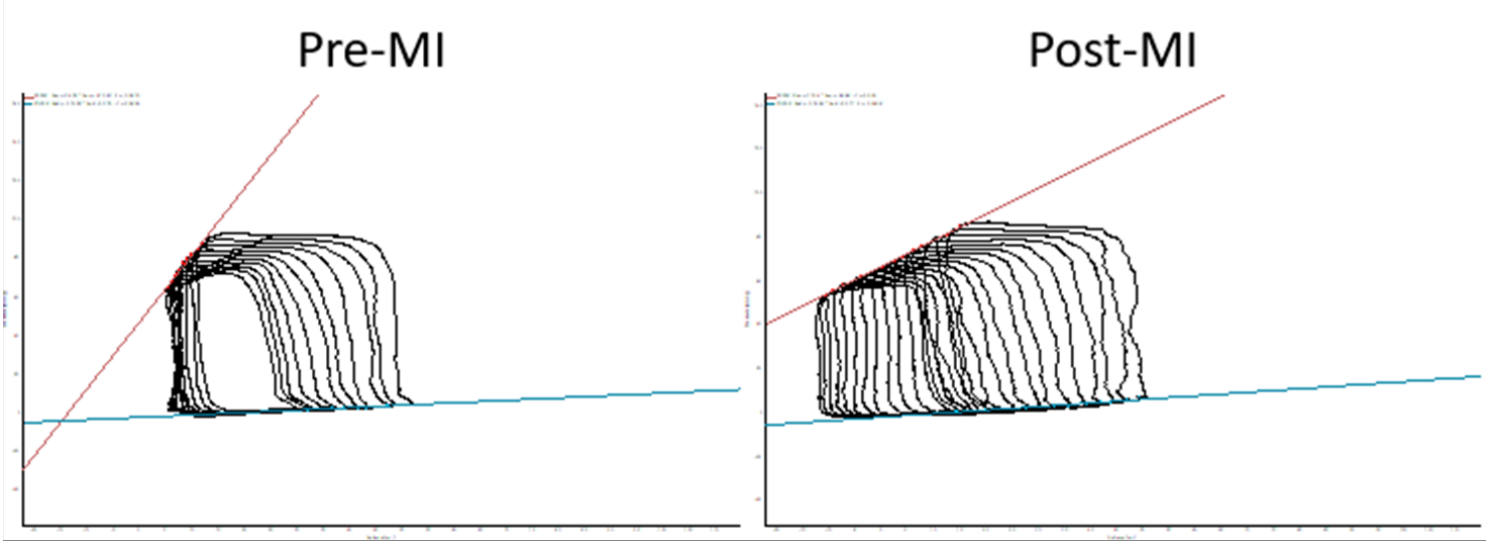


Figure 2

Pressure-volume loop analysis.

Pressure-volume loop analysis offers information on key indices of ventricular performance, including the ESPVR (red line) and EDPVR (blue line), which reflect myocardial contractility and ventricular stiffness, respectively. After MI, the ESPVR slope decreases, indicating impaired contractile function. Conversely, the EDPVR slope becomes steeper, reflecting increased ventricular stiffness; the ventricle becomes less compliant and requires higher filling pressures during diastole, similar to a stiffened balloon that is difficult to inflate. Collectively, these changes indicate the progression of both systolic dysfunction and diastolic rigidity in postinfarct remodeling. **Abbreviations:** EDPVR, end-diastolic pressure-volume relationship; ESPVR, end-diastolic pressure-volume relationship; MI, myocardial infarction.

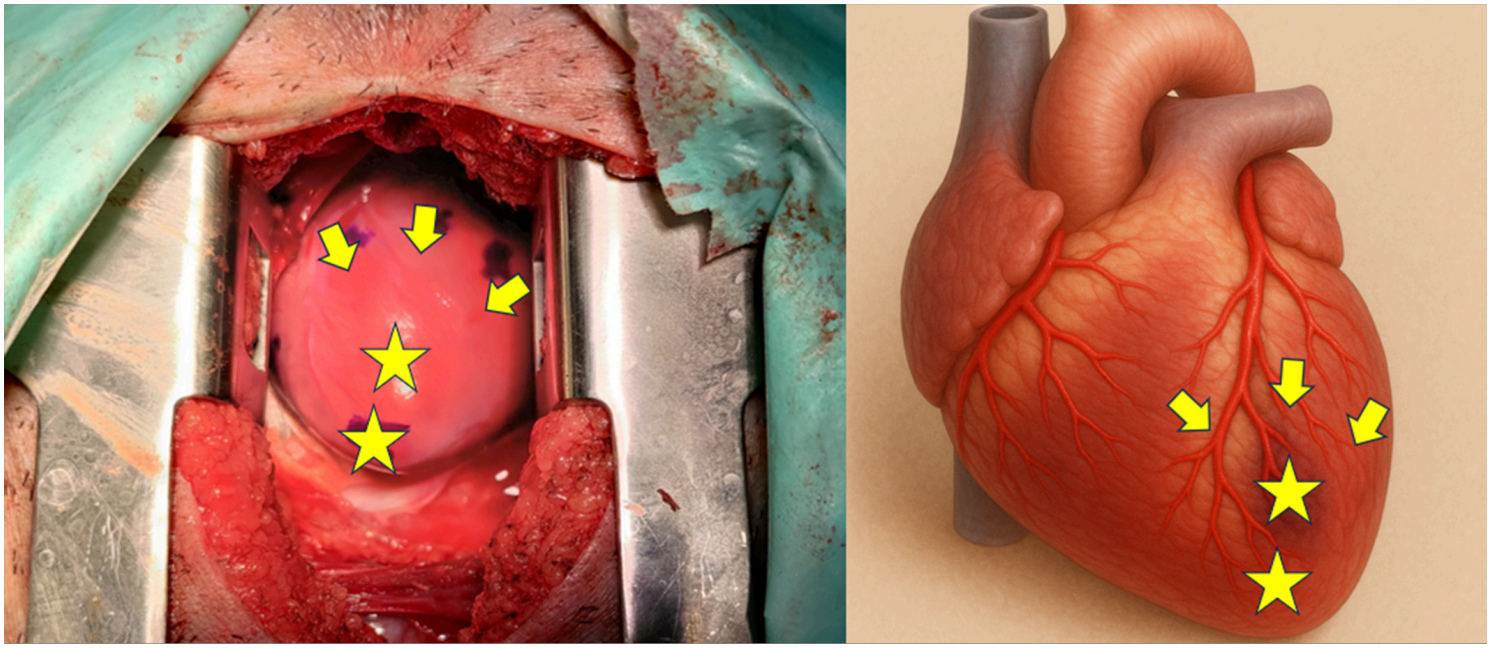


Figure 3

Cell delivery through thoracotomy.

Vehicle or cells were intramyocardially injected through a limited left thoracotomy at 14 days after myocardial infarction. This approach enabled precise delivery under direct visualization into five myocardial sites: two within the central infarct zone (stars) and three along the peri-infarct border (arrows).

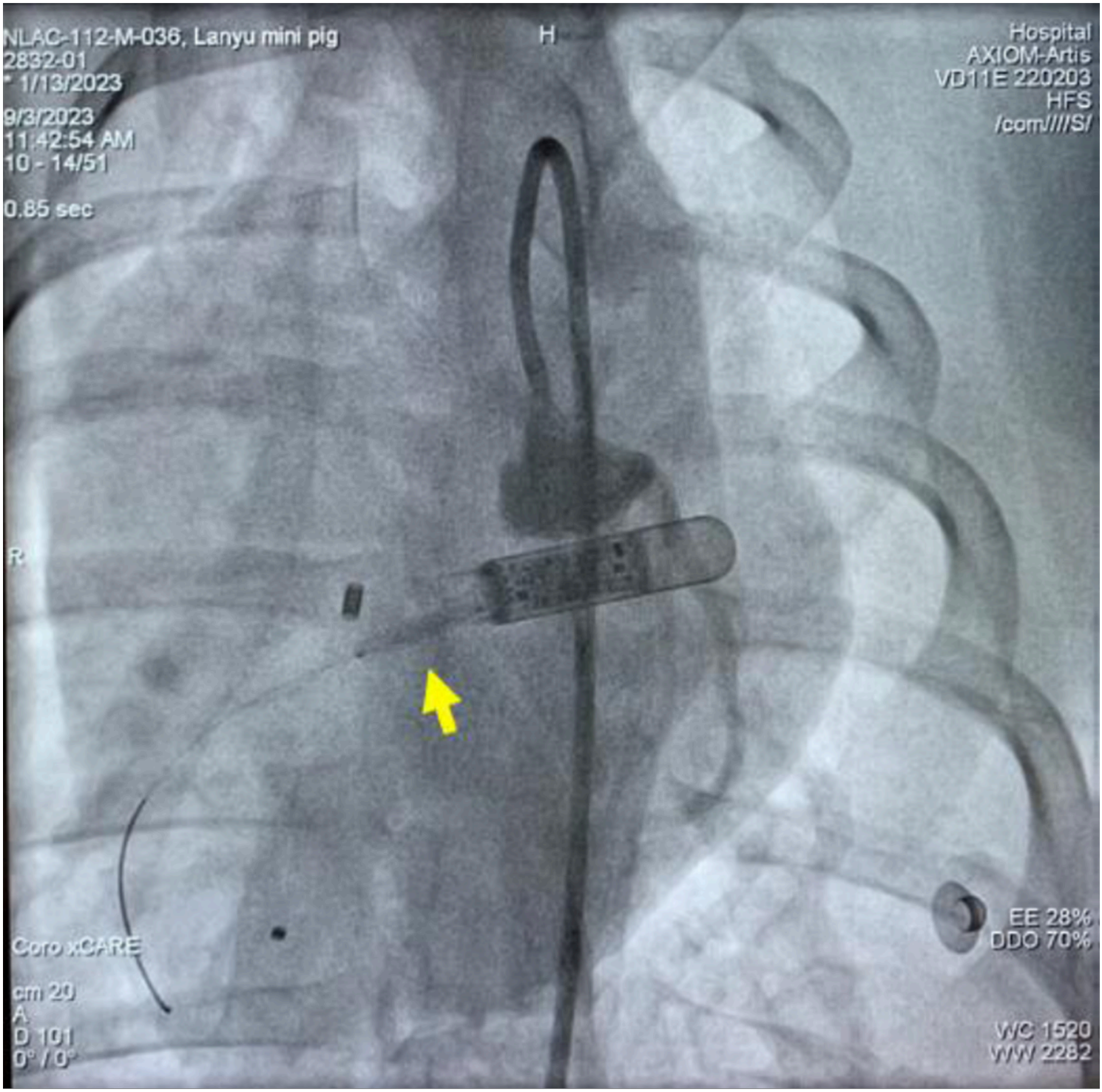


Figure 4

Ischemia-reperfusion surgery.

Myocardial infarction was induced through catheter-based occlusion of the left anterior descending artery for 90 min, followed by reperfusion. Diagonal branch flow was preserved to ensure consistent infarct size and model reproducibility. Yellow arrow: angioplasty balloon catheter.

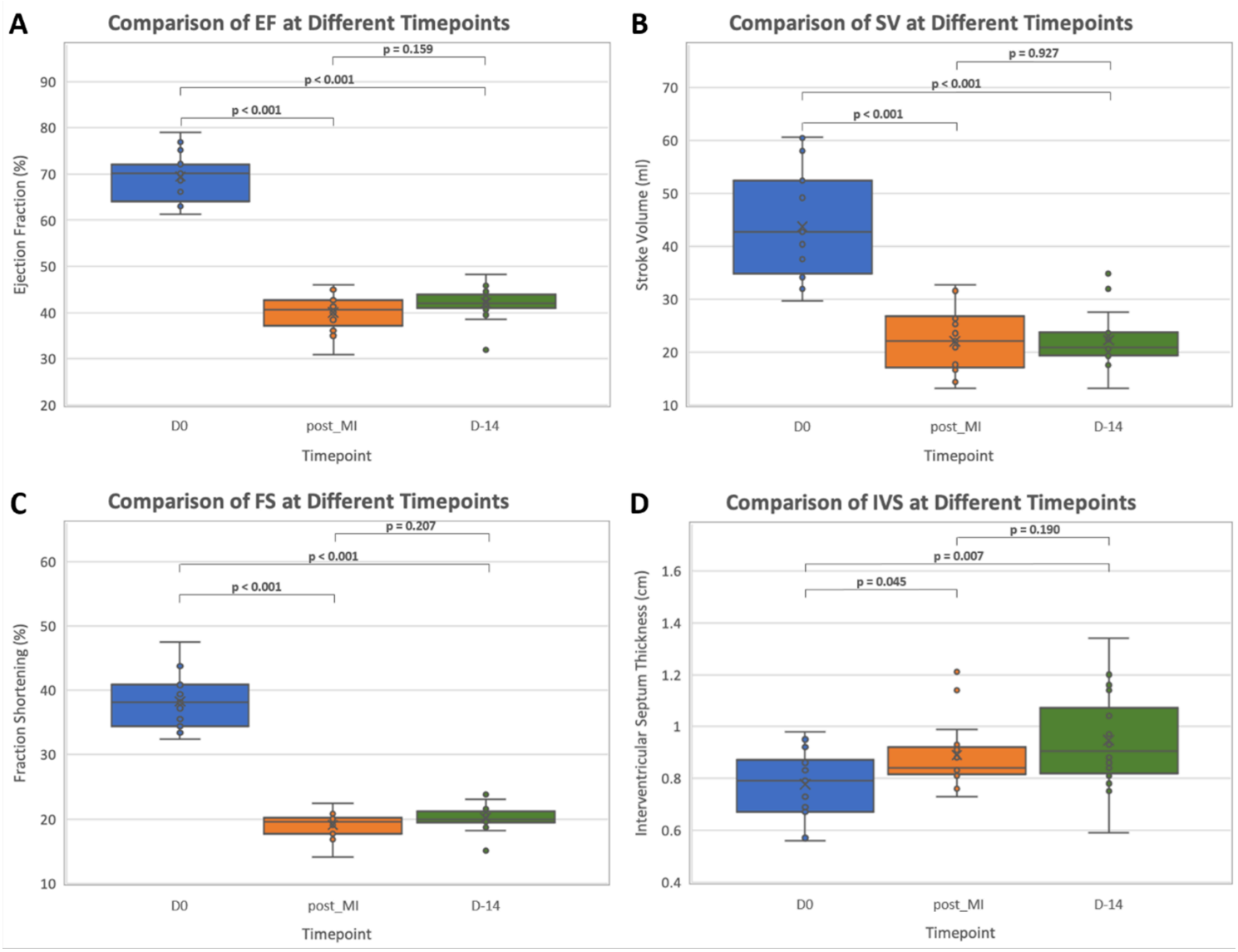


Figure 5

Left ventricular systolic dysfunction, SV reduction, FS reduction, and IVS thickening following MI.

(A) Transthoracic echocardiography revealed a significant decline in LVEF from 69.4% at baseline (Day 0) to 40.0% after MI. LVEF remained at 41.3% on Day 14 ($p < 0.001$ vs. baseline), confirming successful induction of systolic dysfunction.

(B) SV decreased significantly from 43.7 mL at baseline to 22.0 mL after MI and remained reduced at 21.7 mL on Day 14 ($p < 0.001$ vs. baseline), indicating impaired left ventricular pump function.

(C) FS decreased from 38.3% at baseline to 19.1% after MI and remained at 20.3% on Day 14 ($p < 0.001$ vs. baseline), indicating sustained systolic impairment.

(D) IVS thickness increased from 0.78 cm at baseline to 0.89 cm after MI and further to 0.96 cm on Day 14 ($p = 0.007$ vs. baseline), indicating early structural remodeling.

Abbreviations: EF, ejection fraction; IVS, interventricular septum; LVEF, left ventricular ejection fraction; MI, myocardial infarction; SV, stroke volume.

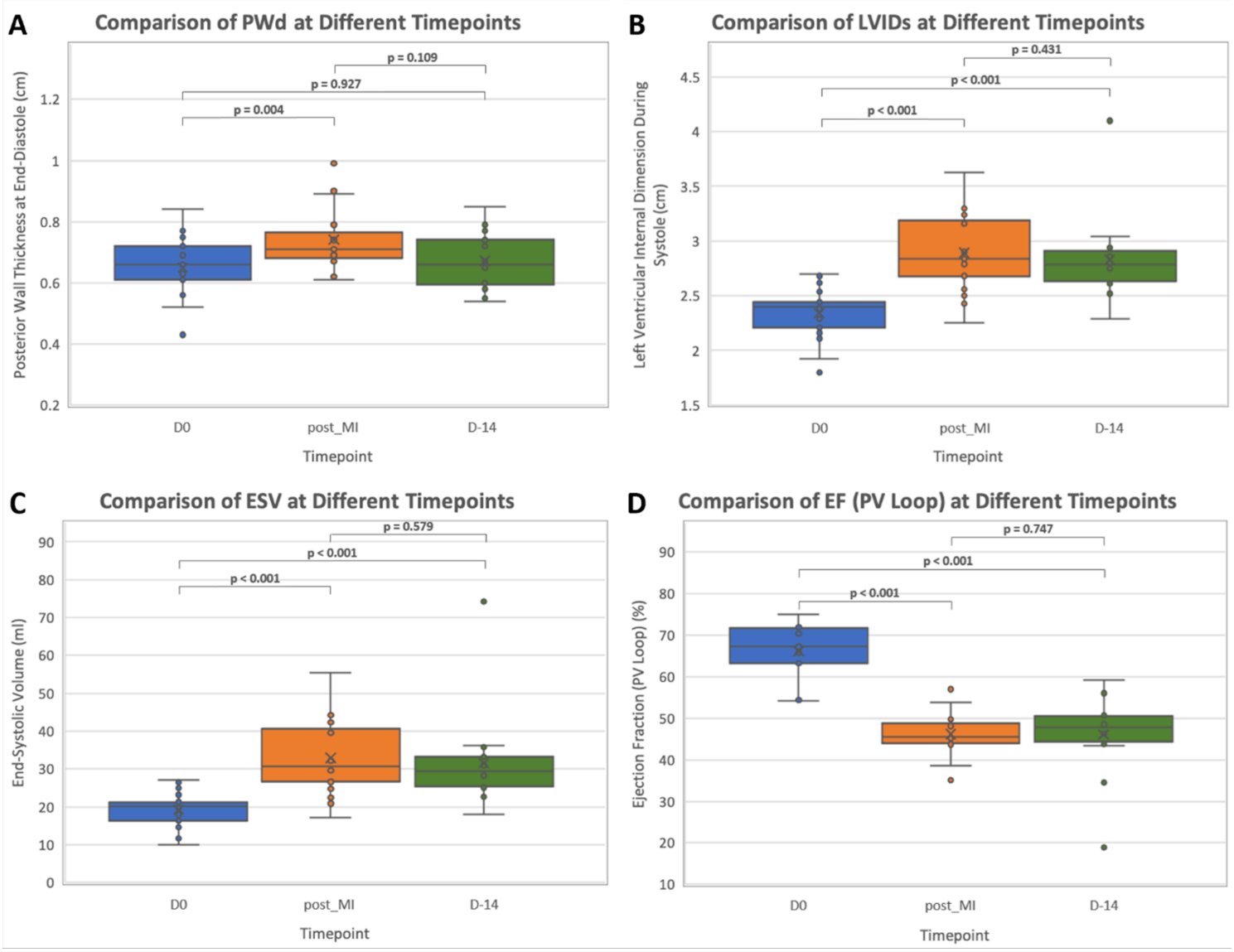


Figure 6

PV loop analysis of PWd increase, systolic chamber dilation, ESV increase, and LVEF reduction following MI.

(A) PWd increased from 0.65 cm at baseline to 0.74 cm after MI (p = 0.004) and partially improved to 0.72 cm on Day 14 (p = 0.927 vs. baseline), indicating incompletely compensatory hypertrophic remodeling.

(B) LVID during systole increased from 2.34 cm at baseline to 2.89 cm after MI (p< 0.001) and remained at 2.78 cm on Day 14 (p < 0.001 vs. baseline), indicating progressive systolic chamber dilation.

(C) ESV increased from 19.3 mL at baseline to 32.9 mL after MI (p < 0.001) and remained at 31.2 mL on Day 14 (p < 0.001 vs. baseline), indicating impaired systolic emptying and ventricular remodeling.

(D) LVEF decreased from 66.1% at baseline to 46.2% after MI ($p < 0.001$) and remained at 45.5% on Day 14 ($p < 0.001$ vs. baseline), confirming significant and sustained systolic dysfunction.

Abbreviations: ESV, end-systolic volume; LVEF, left ventricular ejection fraction; LVID, left ventricular internal diameter; MI, myocardial infarction; PWd, posterior wall thickness at end-diastole.

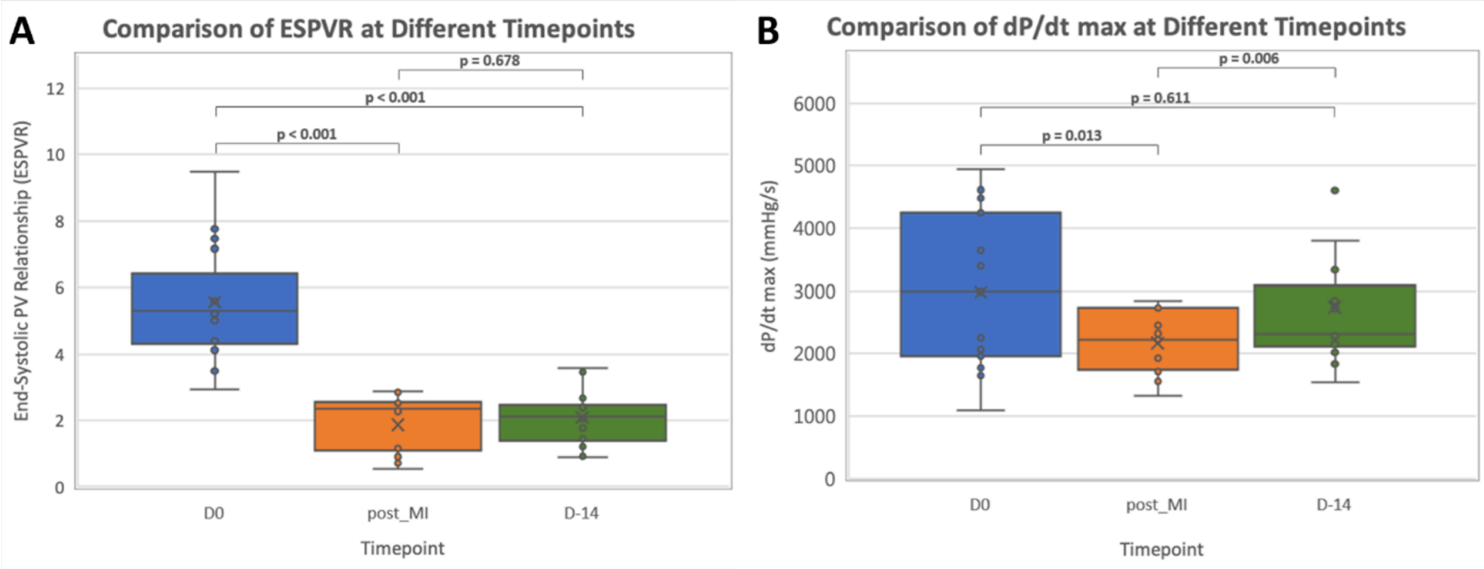


Figure 7

Reductions in ESPVR and dP/dt_{max} following MI.

(A) ESPVR decreased from 5.6 mmHg/mL at baseline to 1.8 mmHg/mL after MI ($p < 0.001$) and remained at 1.9 mmHg/mL on Day 14 ($p < 0.001$ vs. baseline), indicating a pronounced, sustained reduction in contractile function.

(B) dP/dt_{max} in the LV decreased from 2977.4 mmHg/s at baseline to 2164.1 mmHg/s after MI ($p = 0.013$) and partially improved to 2625.7 mmHg/s on Day 14 ($p = 0.611$ vs. baseline), indicating persistent but incompletely reversible impairment of myocardial contractility.

Abbreviations: dP/dt_{max}, maximum rate of pressure rise; ESPVR, end-diastolic pressure-volume relationship; MI, myocardial infarction.

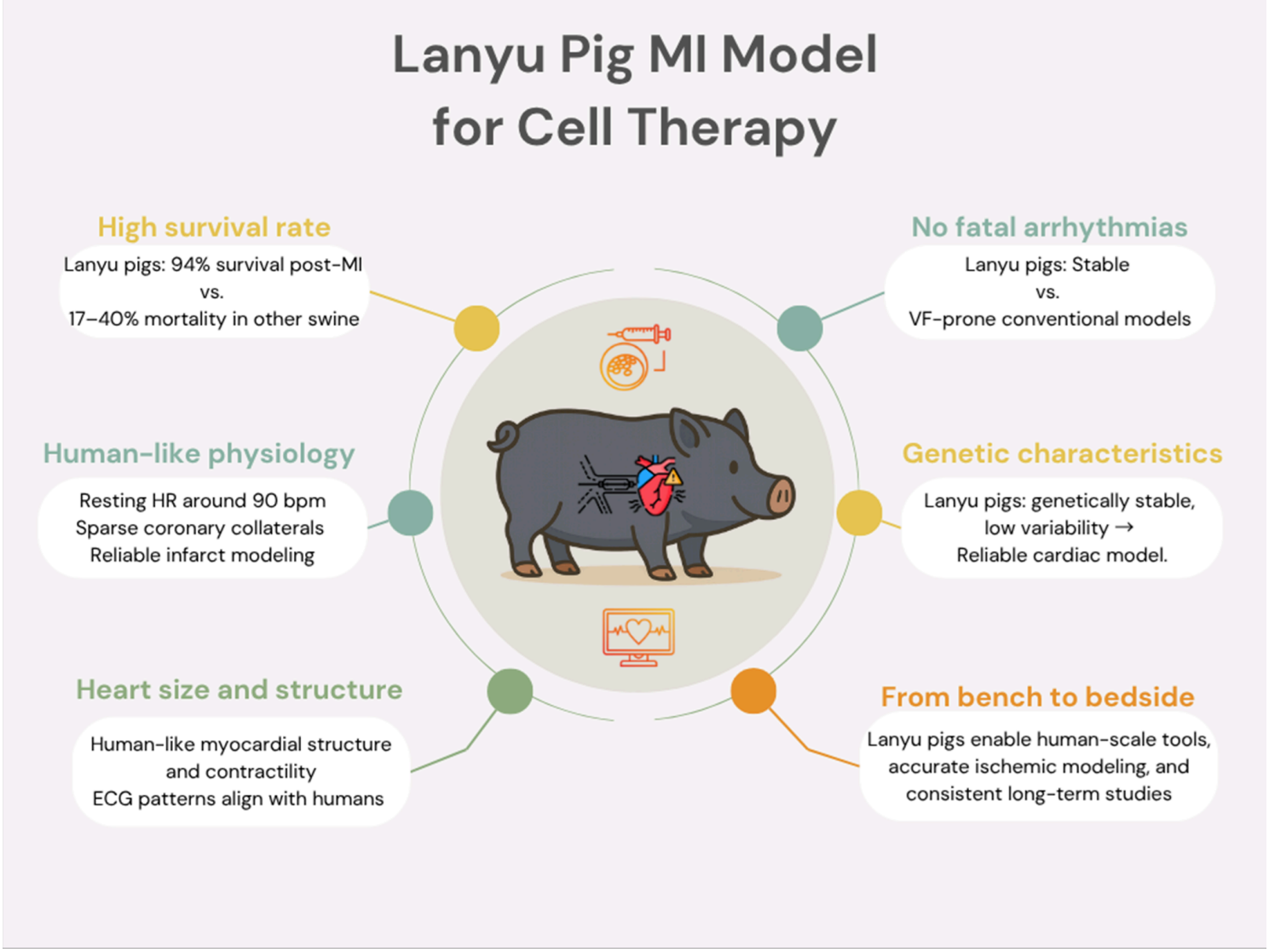


Figure 8

Lanyu pigs as a robust large-animal MI model.

According to our review of the literature, this is the first study to establish the Lanyu pig as a large-animal myocardial infarction model, demonstrating improved tolerance and survival compared with those of the conventional Yorkshire pig and Göttingen minipig models.

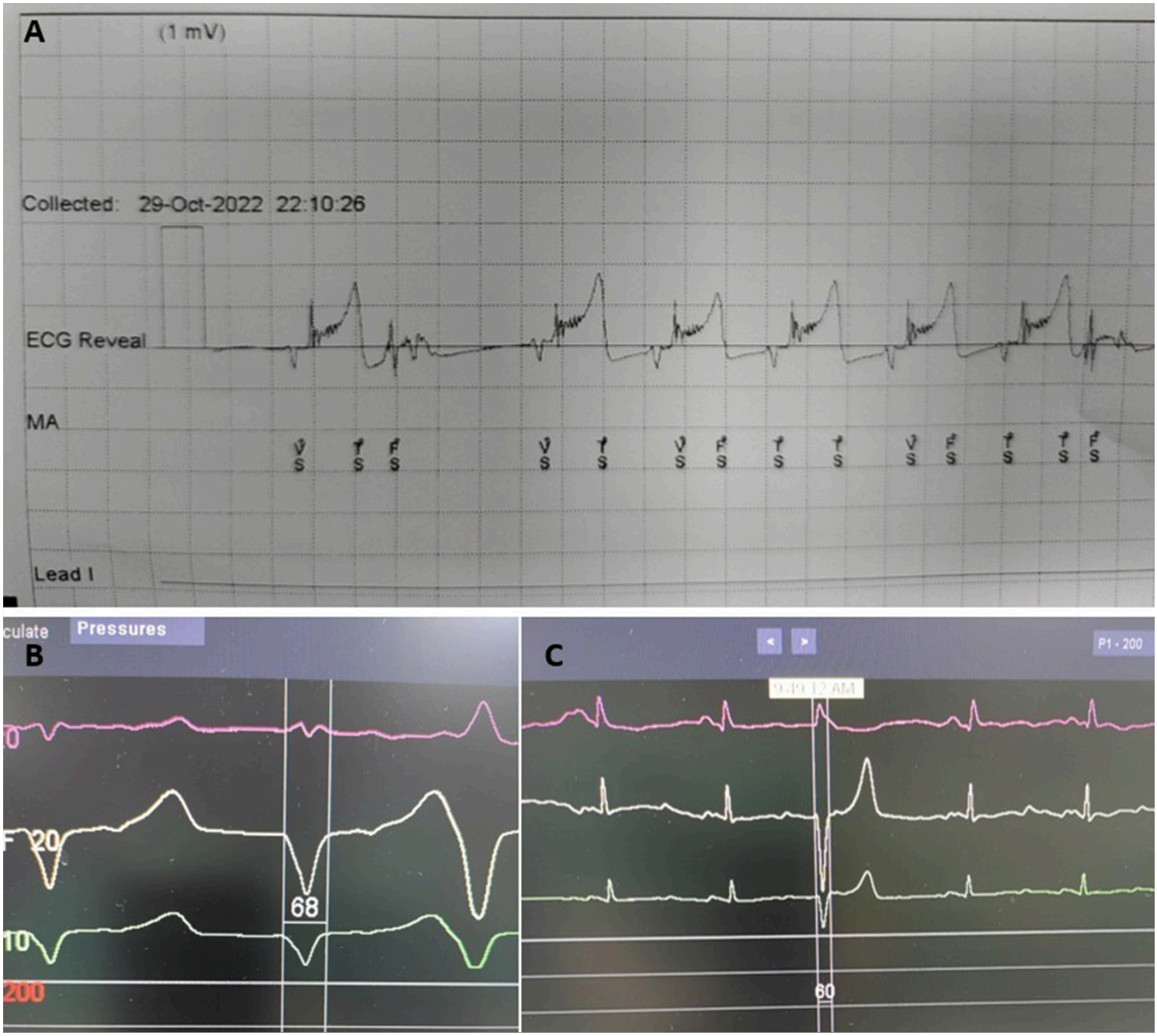


Figure 9

Electrocardiographic characterization of baseline rhythm and ventricular arrhythmias in the Lanyu pig model.

(A) A baseline normal sinus rhythm demonstrates a narrow QRS complex strictly within 60 ms.

(B) An isolated ventricular premature complex exhibits a widened QRS duration measured at 60 ms.

(C) An episode of ventricular tachycardia is characterized by a further widened QRS complex measured at 68 ms.