Up-regulation of Small Conductance Calcium-Activated Potassium Channels in Rabbit Ventricles with Chronic Myocardial Infarction

Young Soo Lee[1], Peng-Sheng Chen[2]

[1]Catholic University of Daegu, Daegu Korea
[2]Krannert Institute of Cardiology, Indianapolis, IN, USA

Calcium activated K^+ channel

In red blood cells
: Cytosolic Ca^{2+} ↑→ K^+ permeability ↑

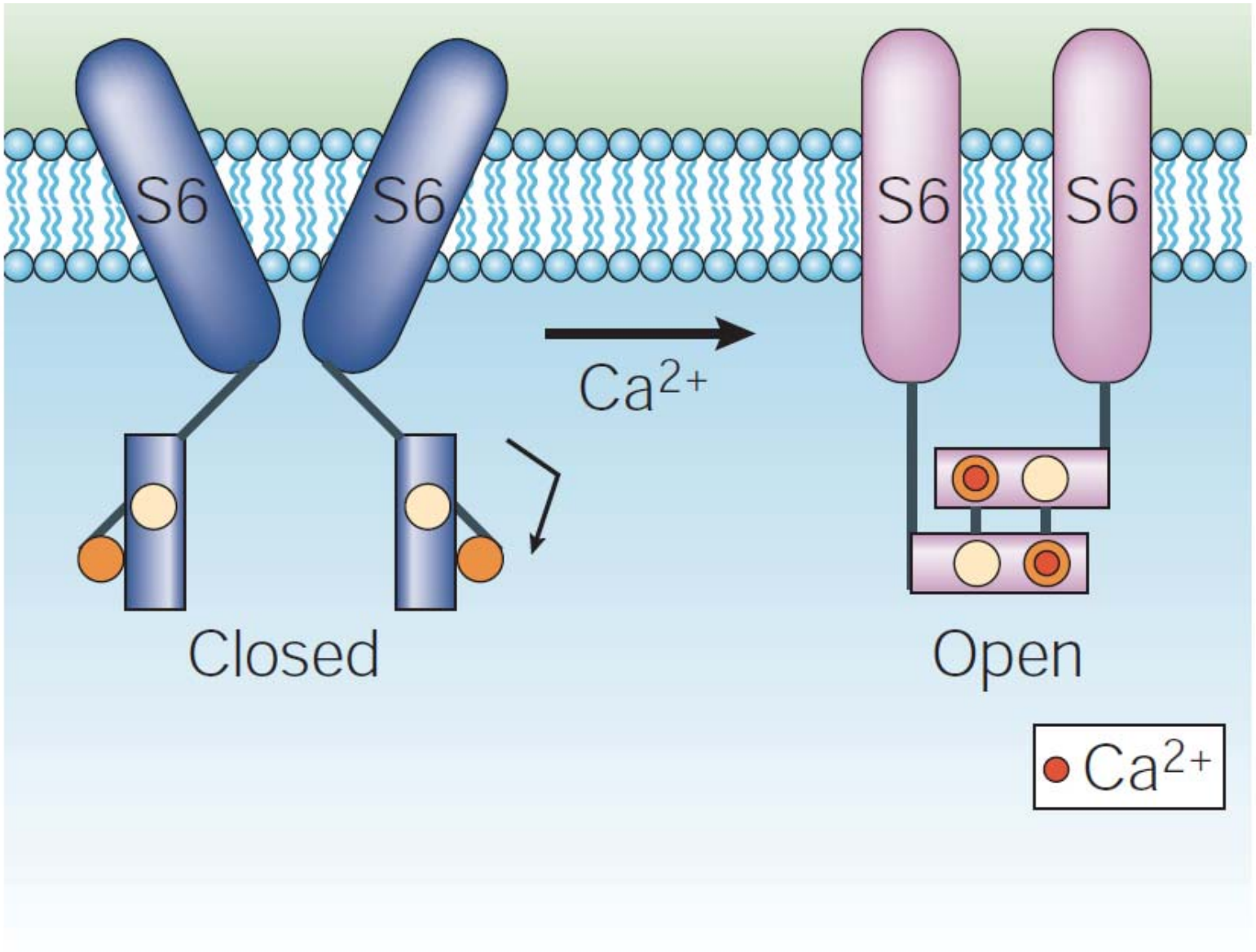
G´ardos G. Biochim. Biophys. Acta 1958;30:653–54

CALCIUM-DEPENDENT
POTASSIUM ACTIVATION
IN NERVOUS TISSUES

R. W. Meech *Ann. Rev. Biophys. Bioeng. 1978. 7:1–18*

: pharmacological manipulation of cytosolic Ca^{2+}
→ the identification of Ca^{2+} -dependent K^+ channels
in molluscan neurons

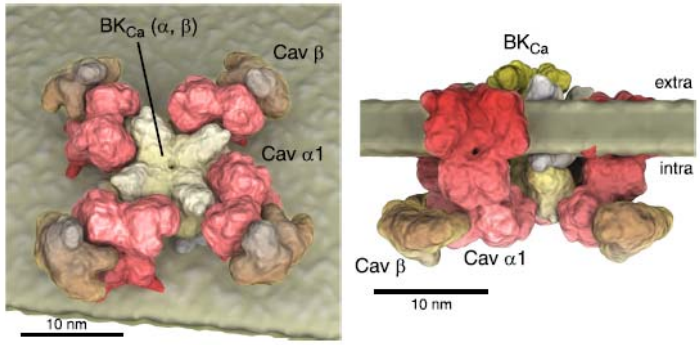
Calcium activated K^+ channel



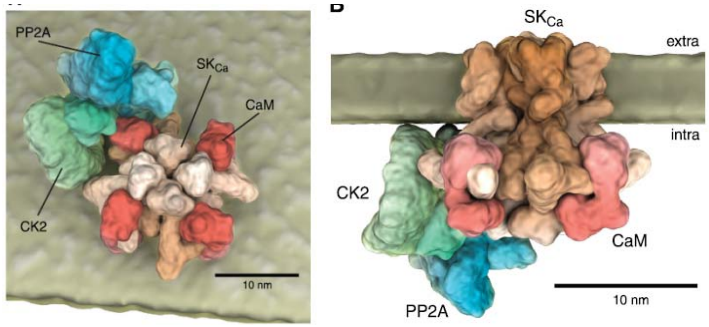
Schumacher, MA. et al. Nature 2001;410:1120–1124.

Calcium activated K^+ channel

1. Big-conductance Ca^{2+} activated K channel
   - BK channel, 100-200 pS
   - smooth muscle, adrenal gland
     brain, auditory sensory hair cell
   - Voltage-dependent



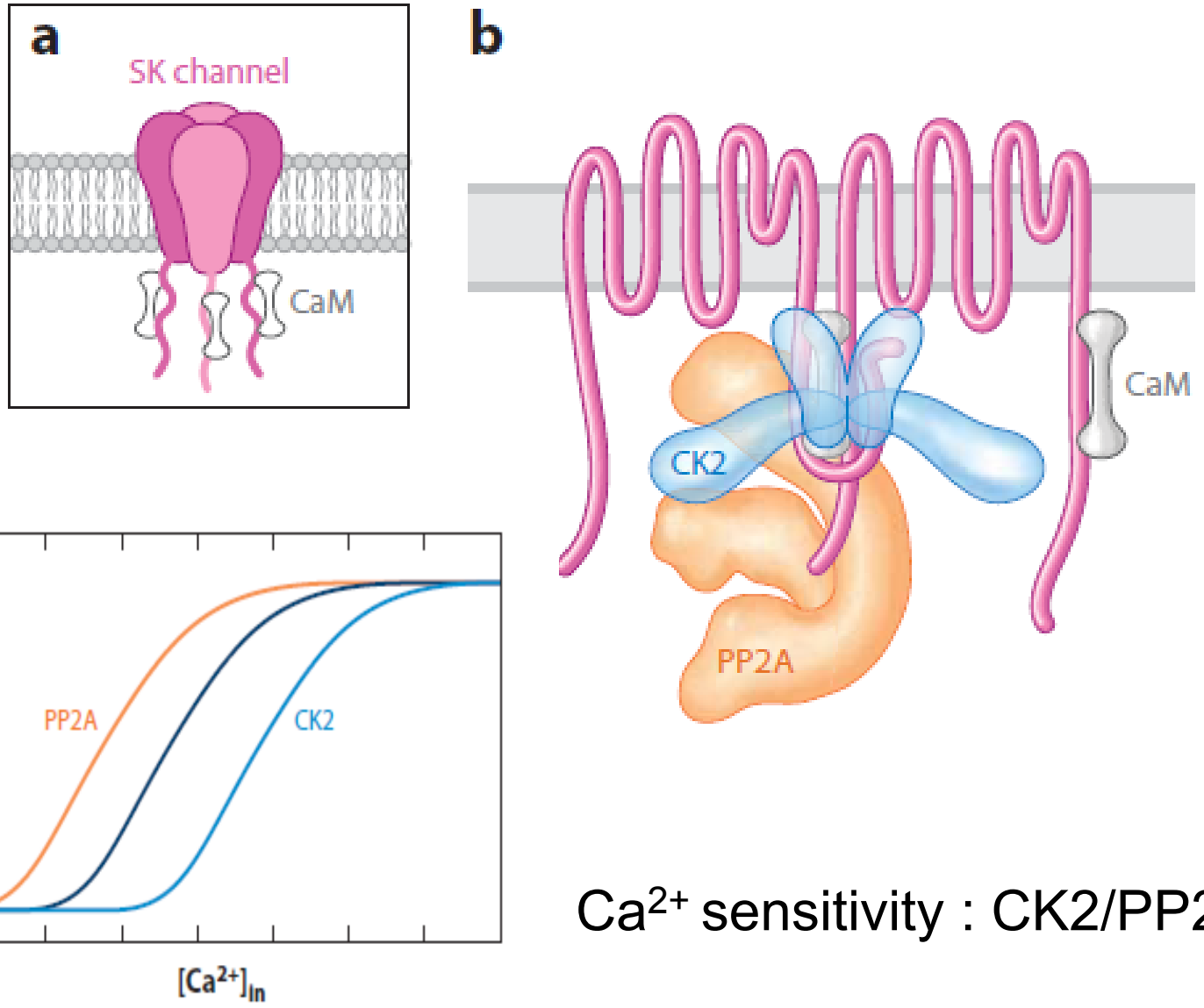
2. Small –conductance Ca^{2+} activated K channel
   - SK channel, 10-20 pS
   - SK1, SK2 : CNS neuron
     SK3 : neuronal and glial cell
   - Voltage-independent



3. Intermediated-conductance Ca^{2+} activated K channel
   - IK channel, SK4,11-40 pS
   - non-neuronal cell : muscle, epithelia, blood cell

Berkefeld H, et al. Physiol rev 2010;90:1437-1459
Vergara C et al. Current Opinion in Neurobiology 1998;8:321-329

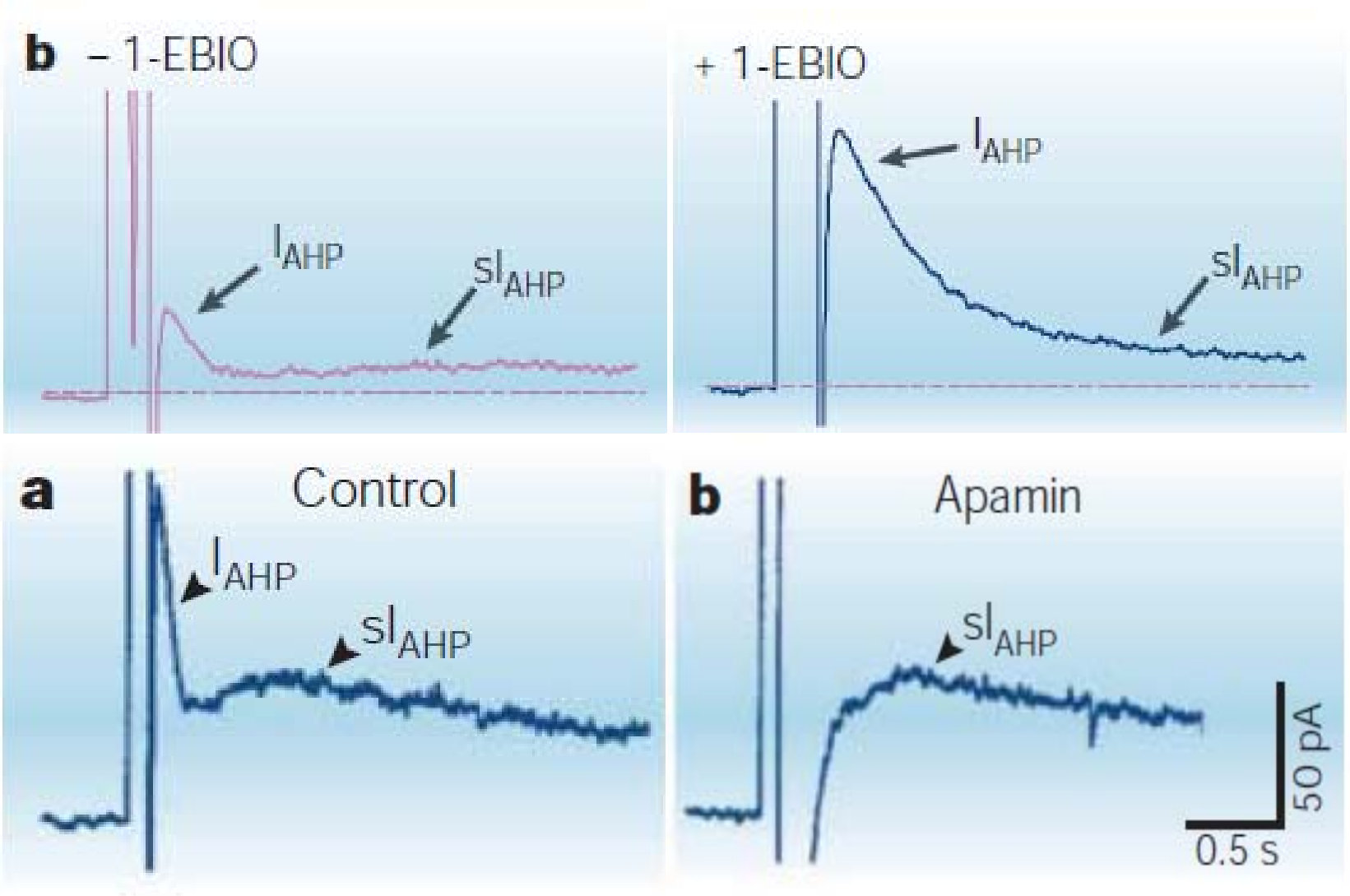
SK channel



Ca^{2+} sensitivity : CK2/PP2A

Adelman JP et al. Annu. Rev. Physiol. 2012. 74:5.1–5.25

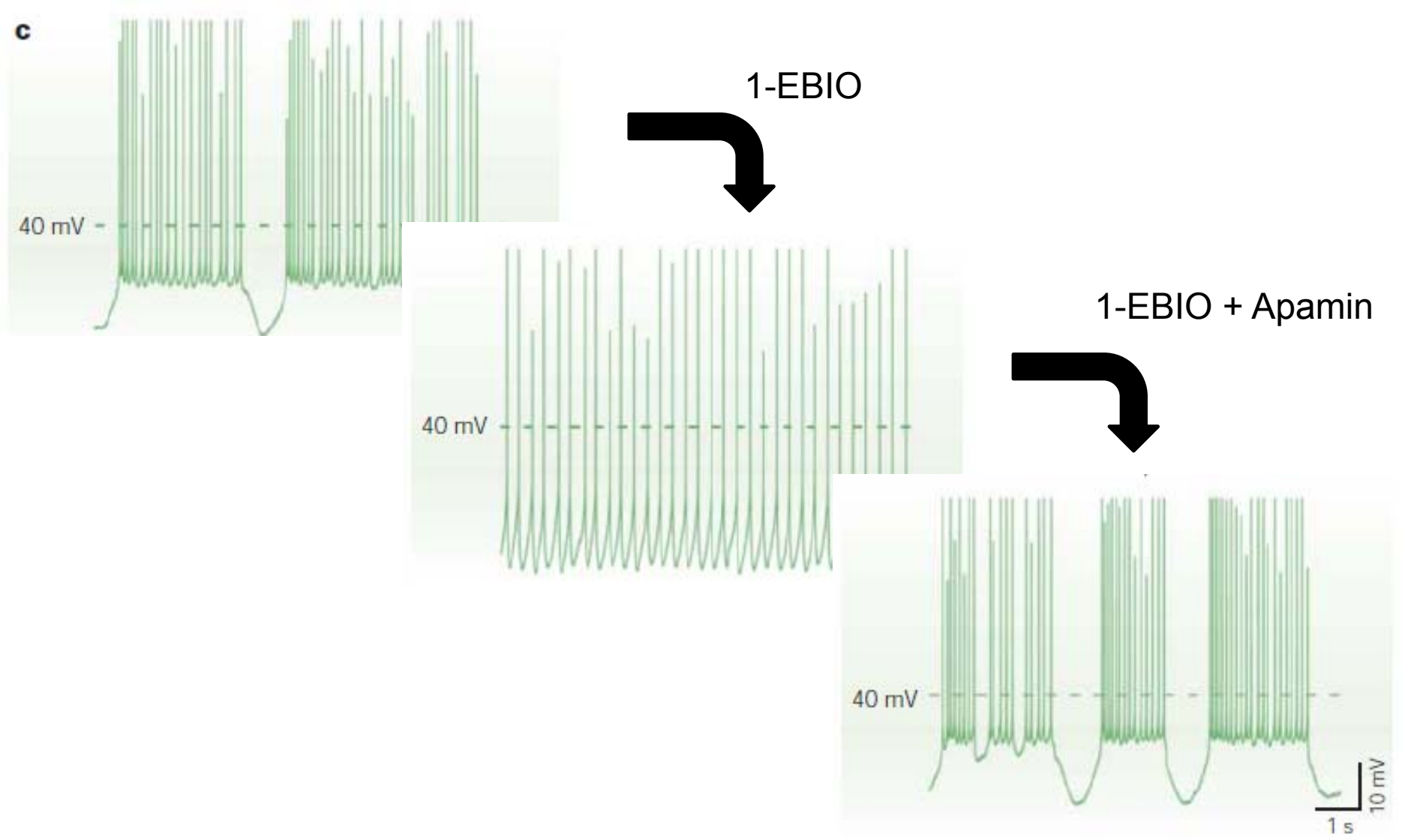
Function of SK channel in Neurons



1-EBIO : 1-ethyl-2-benzimidazolinone
SK channel enhancer

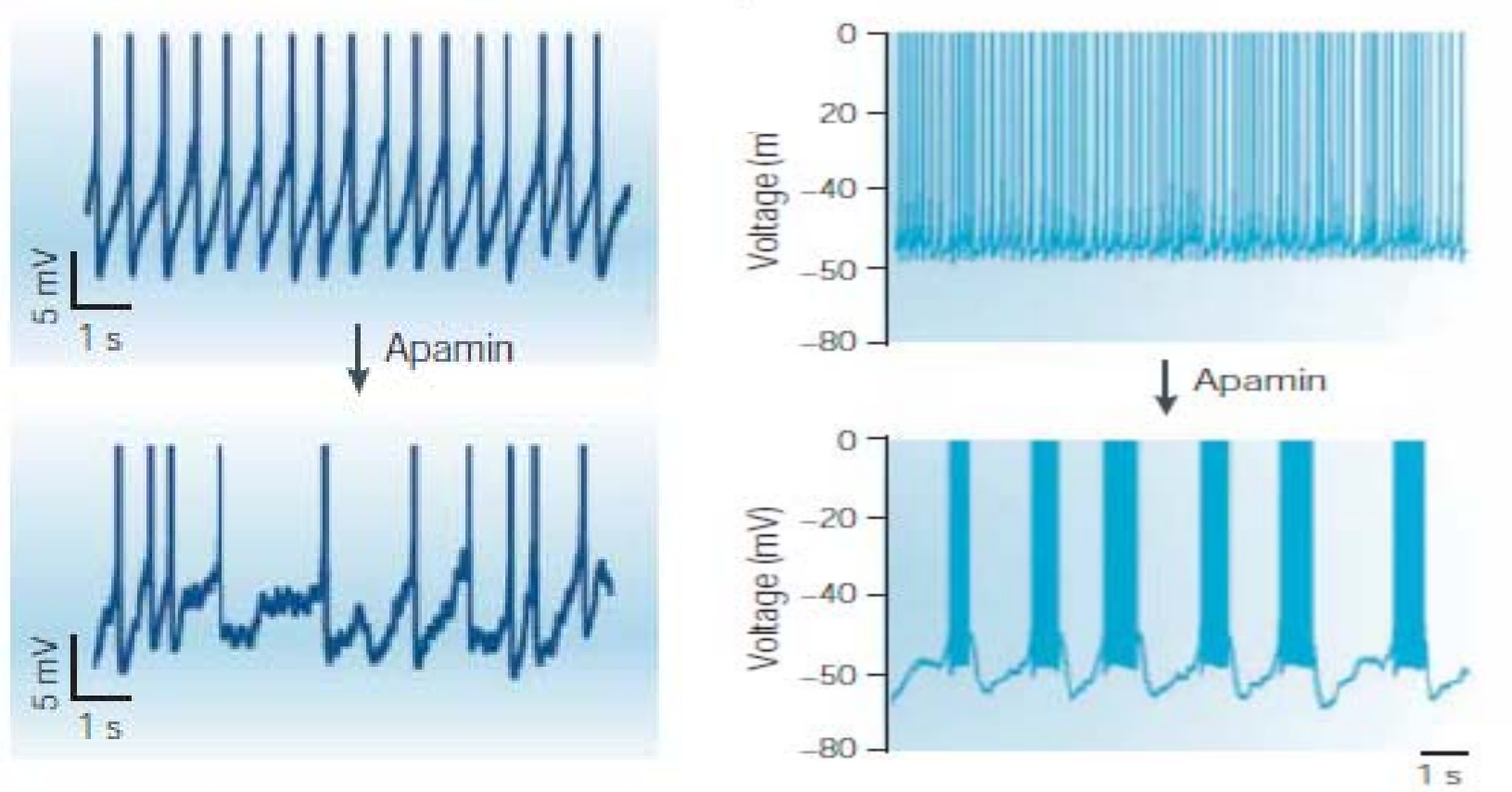
Pedarzani, P. et al. J. Biol. Chem. 2001;276: 9762–9769
Stocker, M. et al. Proc. Natl Acad. Sci. USA 1999;96: 4662–4667

Function of SK channel in Neurons



Cingolani, LA, et al. Neurosci. 2002;22: 4456–4467.

Function of SK channel in Neurons

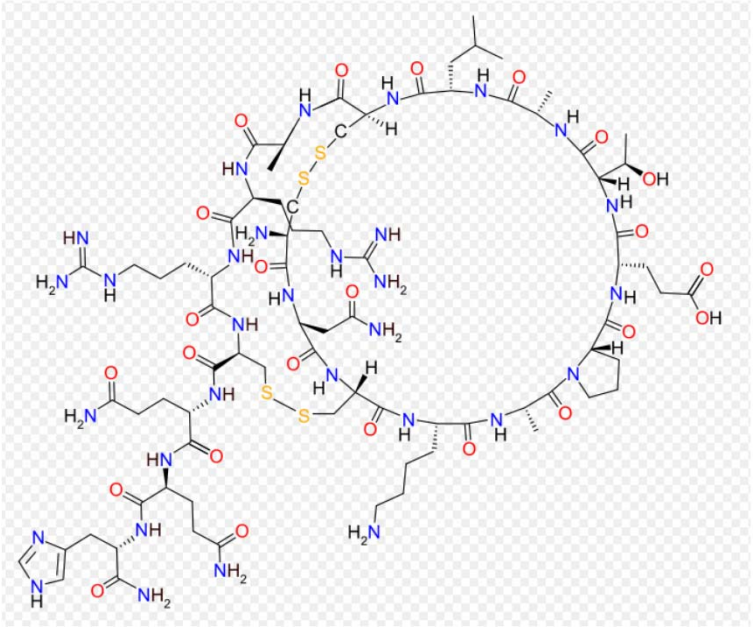


⟶ Spike-frequency adaptation
: Regulator of spike-timing precision

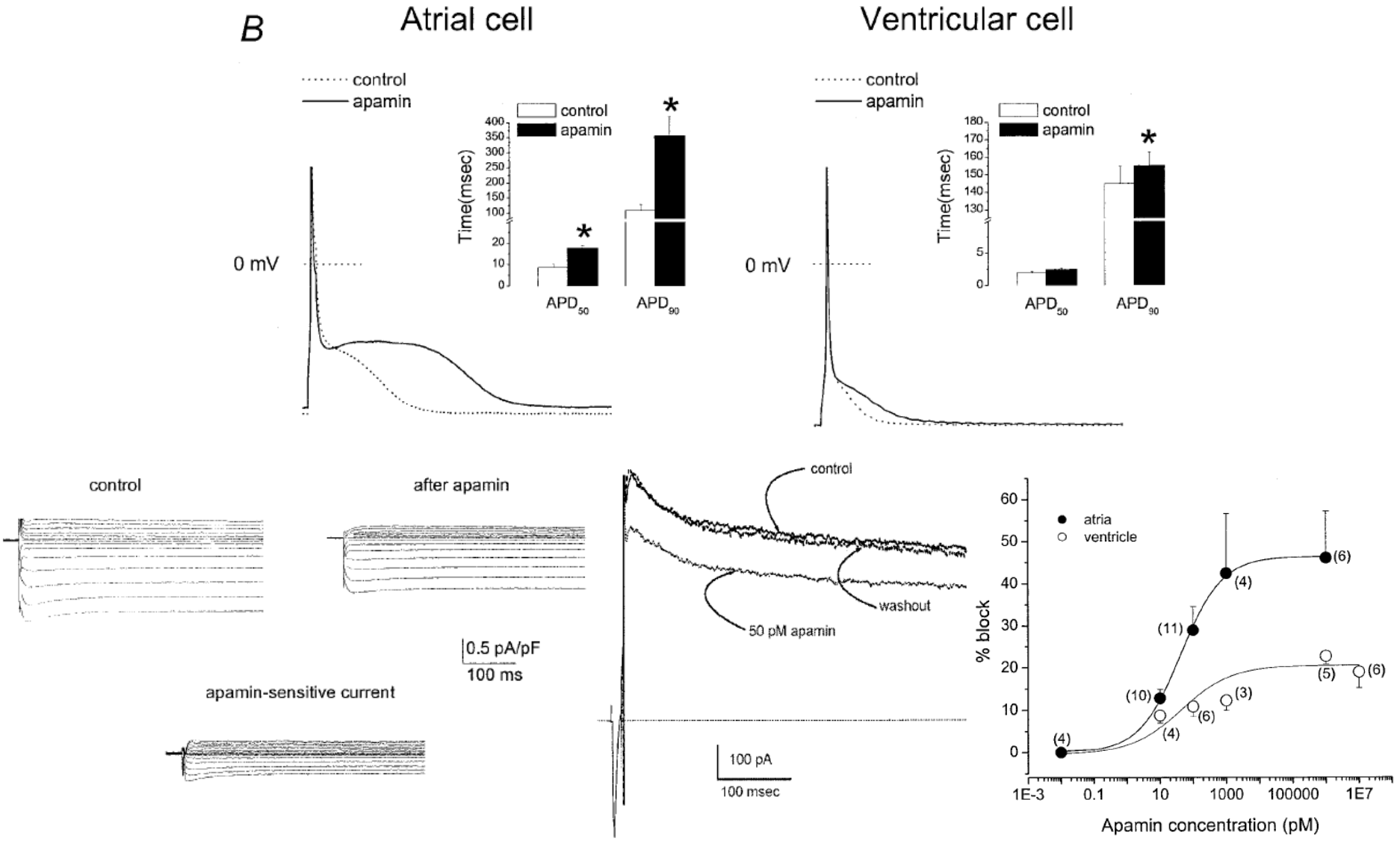
Cingolani, LA, et al. Neurosci. 2002;22: 4456–4467.

Apamin

- ✓ Bee venom toxin
- ✓ Polypeptide of 18 amino acids with two disulfide bridges
- ✓ Isolated from *Apis mellifera*.,



SK channel in heart



Xu Y, et al. JBC, 2003;278:49085-49094

Small-Conductance Calcium-Activated Potassium Channel and Recurrent Ventricular Fibrillation in Failing Rabbit Ventricles

Su-Kiat Chua,* Po-Cheng Chang,* Mitsunori Maruyama, Isik Turker, Tetsuji Shinohara, Mark J. Shen, Zhenhui Chen, Changyu Shen, Michael Rubart-von der Lohe, John C. Lopshire, Masahiro Ogawa, James N. Weiss, Shien-Fong Lin, Tomohiko Ai, Peng-Sheng Chen

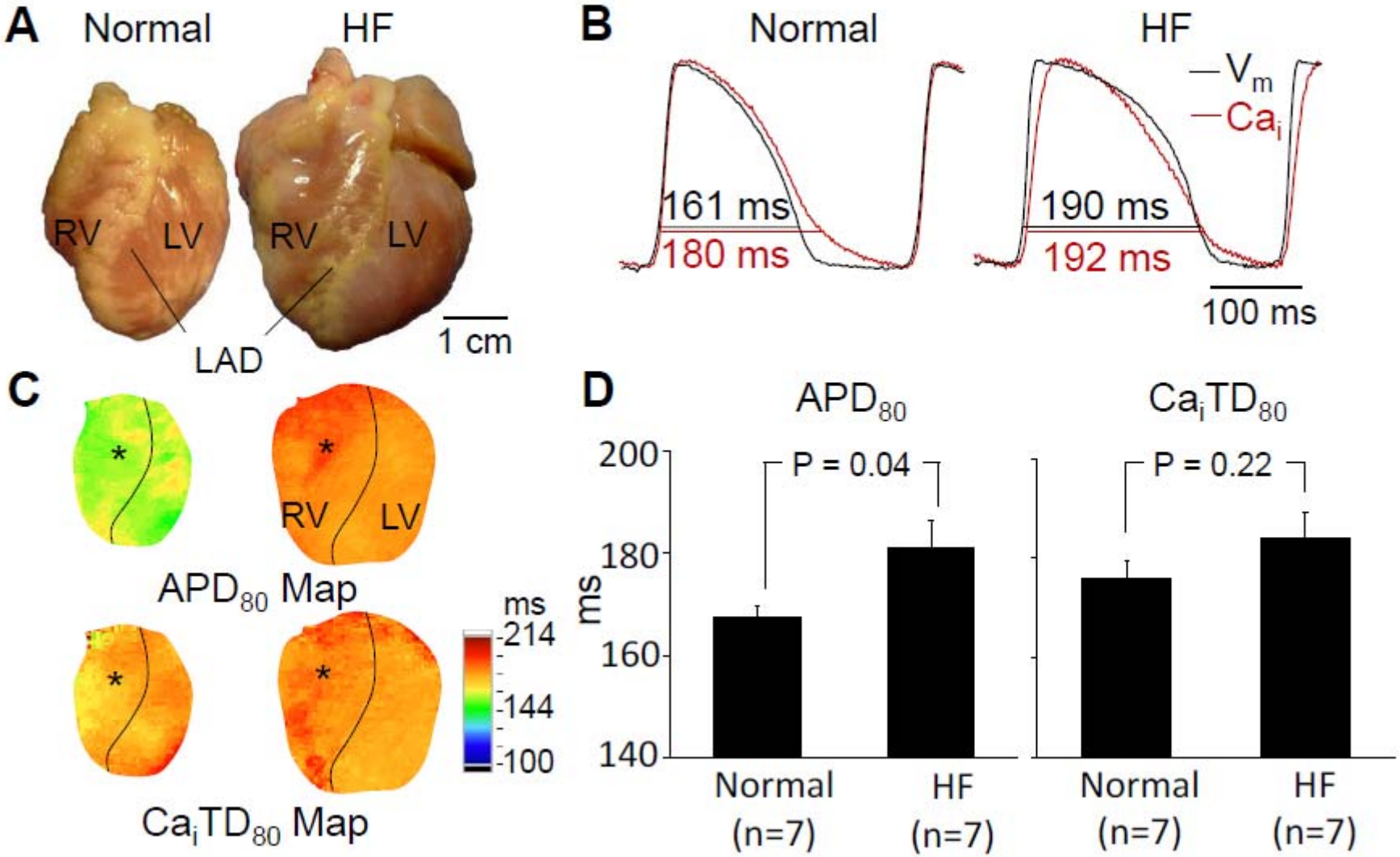
Rationale: **Fibrillation/defibrillation episodes in failing ventricles may be followed by action potential duration (APD) shortening and recurrent spontaneous ventricular fibrillation (SVF).**

Objective: **We hypothesized that activation of apamin-sensitive small-conductance Ca^{2+}-activated K^+ (SK) channels is responsible for the postshock APD shortening in failing ventricles.**

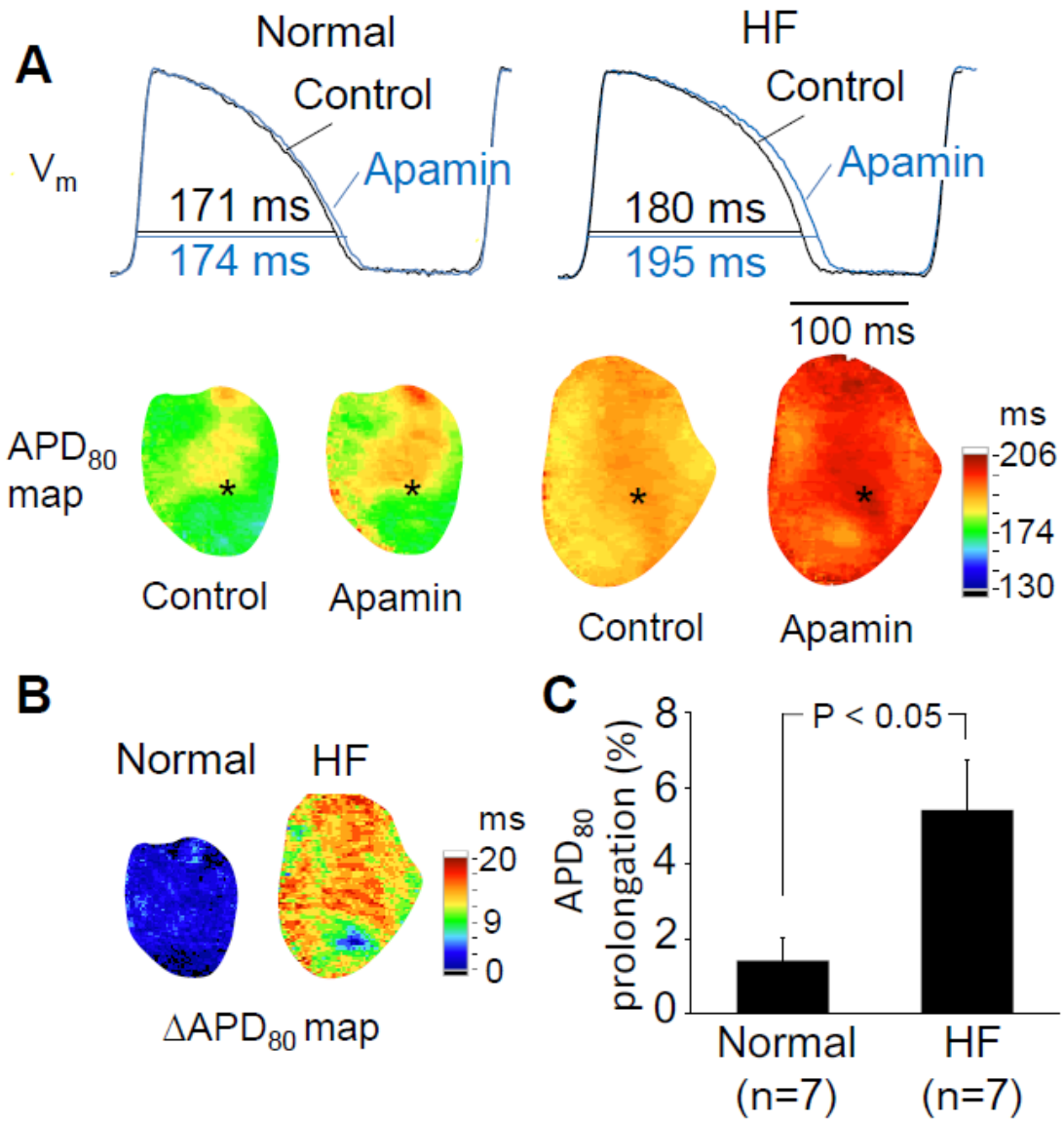
Methods and Results: **A rabbit model of tachycardia-induced heart failure was used. Simultaneous optical mapping of intracellular Ca^{2+} and membrane potential (V_m) was performed in failing and nonfailing ventricles. Three failing ventricles developed SVF (SVF group); 9 did not (no-SVF group). None of the 10 nonfailing ventricles developed SVF. Increased pacing rate and duration augmented the magnitude of APD shortening. Apamin (1 μmol/L) eliminated recurrent SVF and increased postshock APD_{80} in the SVF group from 126±5 to 153±4 ms ($P<0.05$) and from 147±2 to 162±3 ms ($P<0.05$) in the no-SVF group but did not change APD_{80} in nonfailing group. Whole cell patch-clamp studies at 36°C showed that the apamin-sensitive K^+ current (I_{KAS}) density was significantly larger in the failing than in the normal ventricular epicardial myocytes, and epicardial I_{KAS} density was significantly higher than midmyocardial and endocardial myocytes. Steady-state Ca^{2+} response of I_{KAS} was leftward-shifted in the failing cells compared with the normal control cells, indicating increased Ca^{2+} sensitivity of I_{KAS} in failing ventricles. The K_d was 232±5 nmol/L for failing myocytes and 553±78 nmol/L for normal myocytes ($P=0.002$).**

Conclusions: **Heart failure heterogeneously increases the sensitivity of I_{KAS} to intracellular Ca^{2+}, leading to upregulation of I_{KAS}, postshock APD shortening, and recurrent SVF. (*Circ Res*. 2011;108:971-979.)**

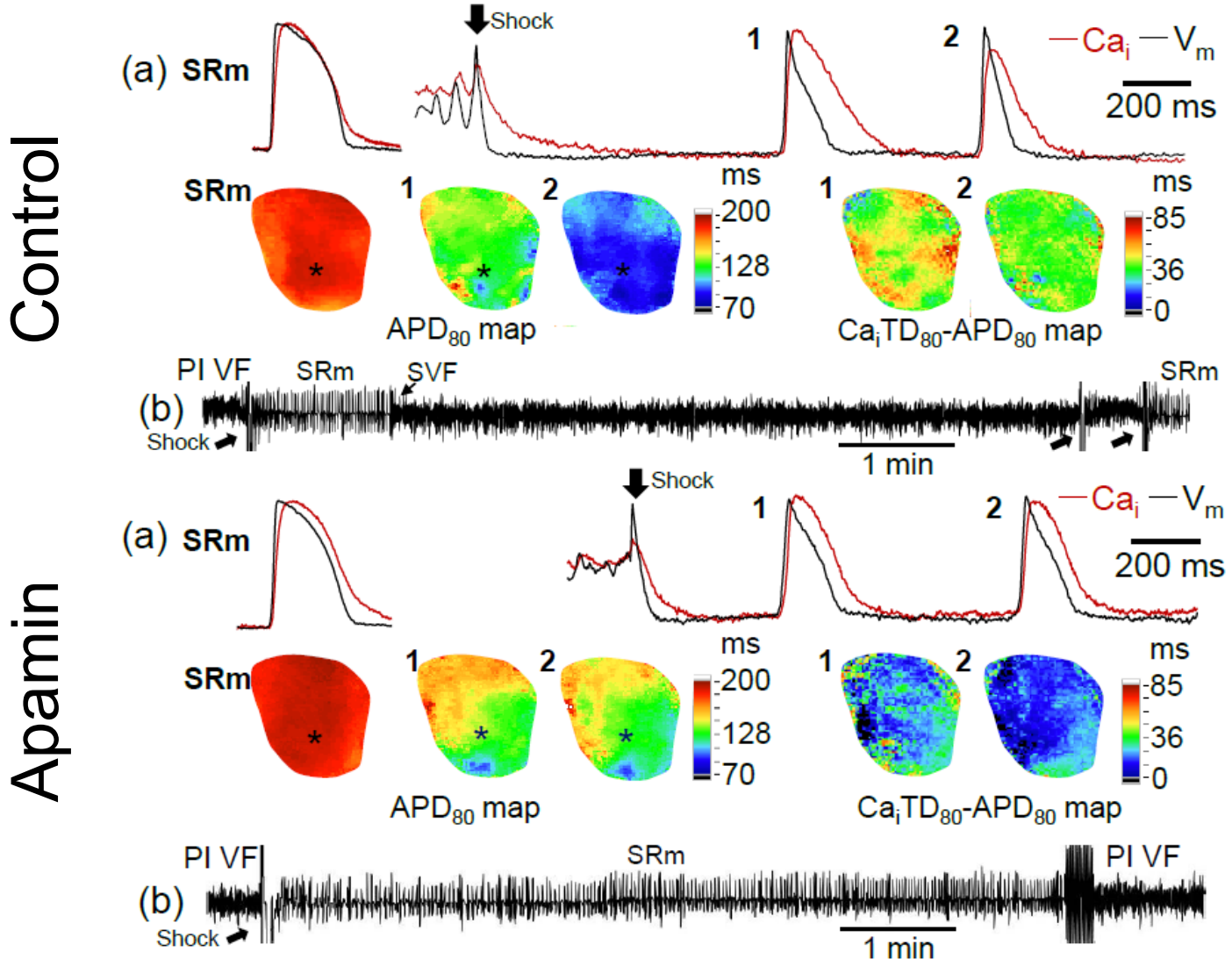
APD in normal and failing ventricles



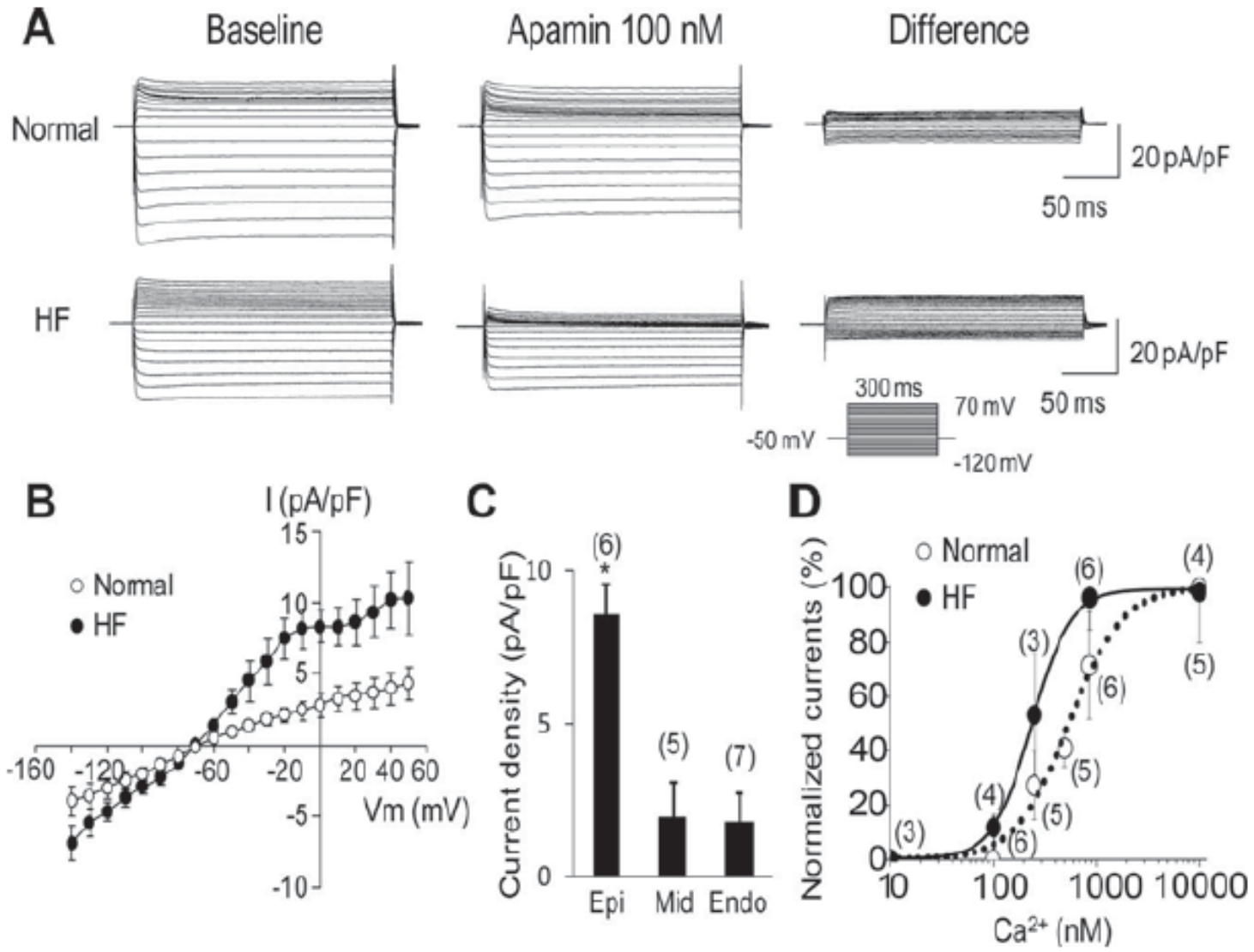
Effect of apamin in normal and failing ventricles



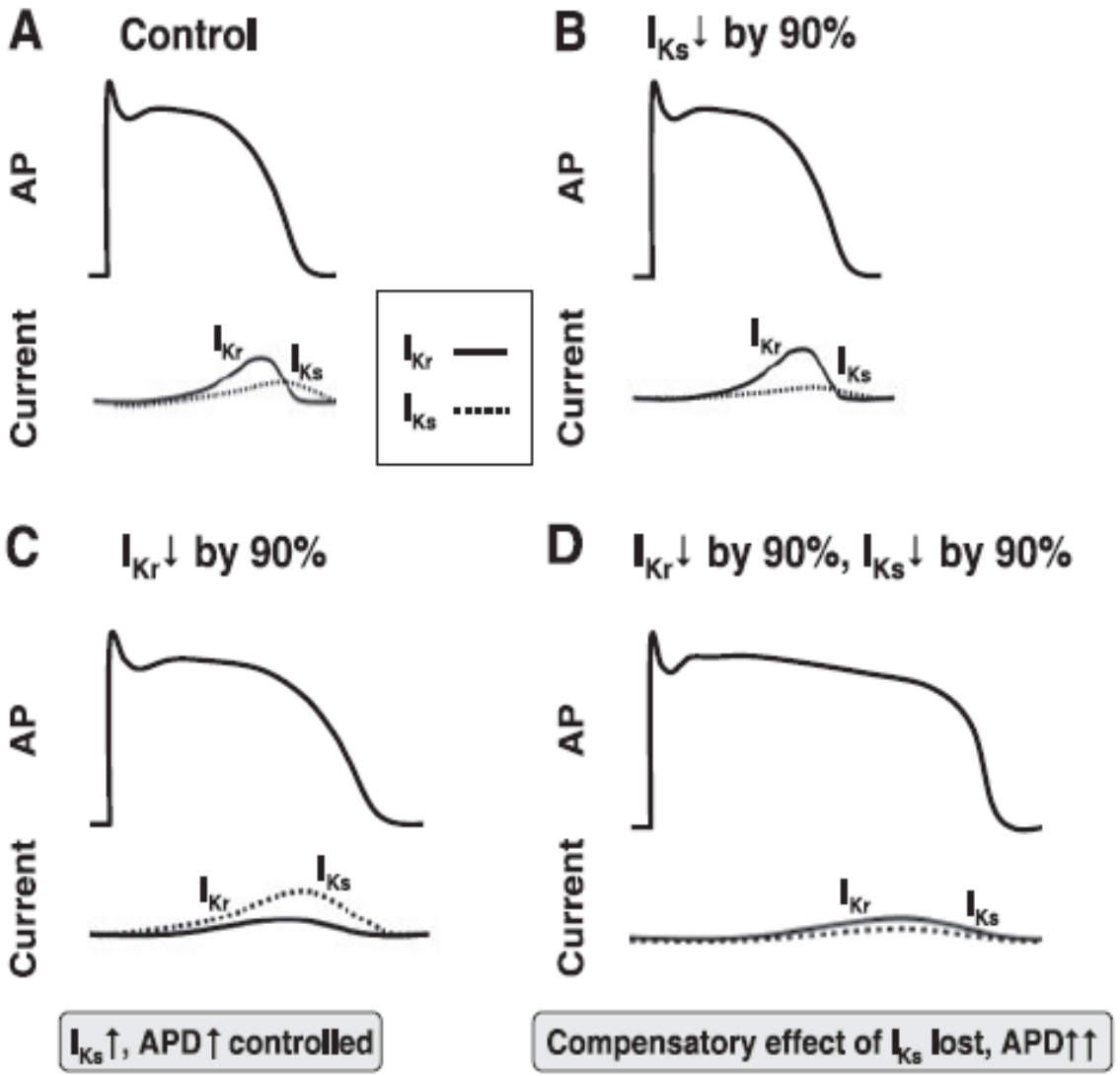
Effect of apamin on postshock APD in failing ventricles



Apamin-sensitive currents in normal and failing ventricles



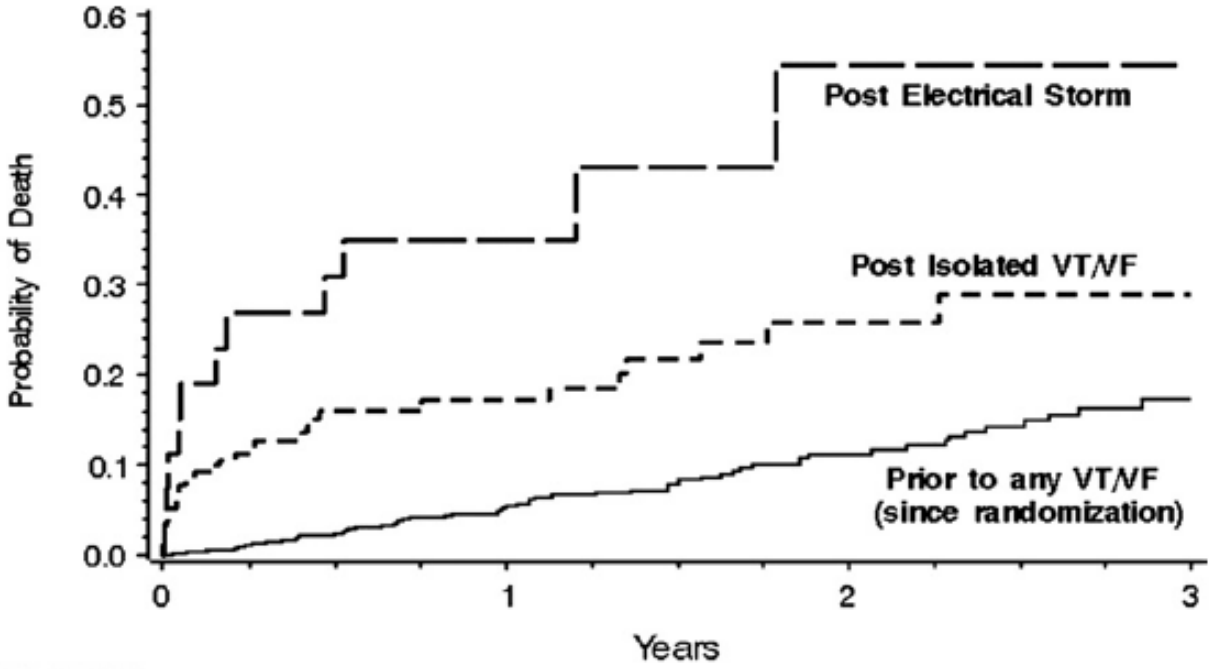
Repolarization reserve



Nattel D, et al. Physiol Rev 2007;87:425–456.

Ventricular arrhythmia storms in postinfarction patients with implantable defibrillators for primary prevention indications: A MADIT-II substudy

(Heart Rhythm 2007;4:1395–1402)

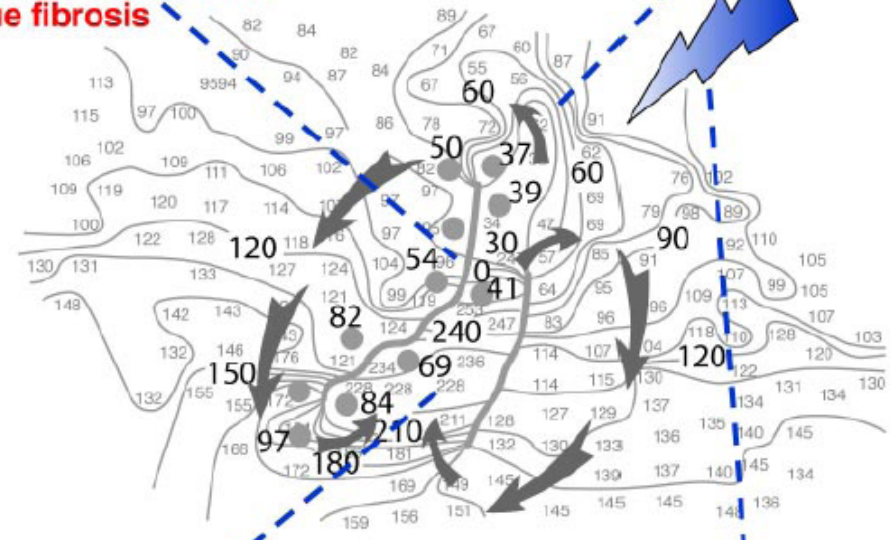


PATIENTS AT RISK				
Electrical Storm	27	9 (0.35)	3 (0.55)	2 (0.55)
Isolated VT/VF	156	67 (0.17)	28 (0.26)	6 (0.29)
Prior to any VT/VF	719	419 (0.06)	206 (0.11)	75 (0.17)

Mechanism of post-MI ventricular arrhythmia

Increased tissue anisotropy
- Connexin downregulation
- Smaller and fewer gap junctions
- Fewer side-to-side connections
- Tissue fibrosis

Slowed conduction
- Connexin downregulation and redistribution
- I_{Na} down-regulation
- I_{CaL} down-regulation



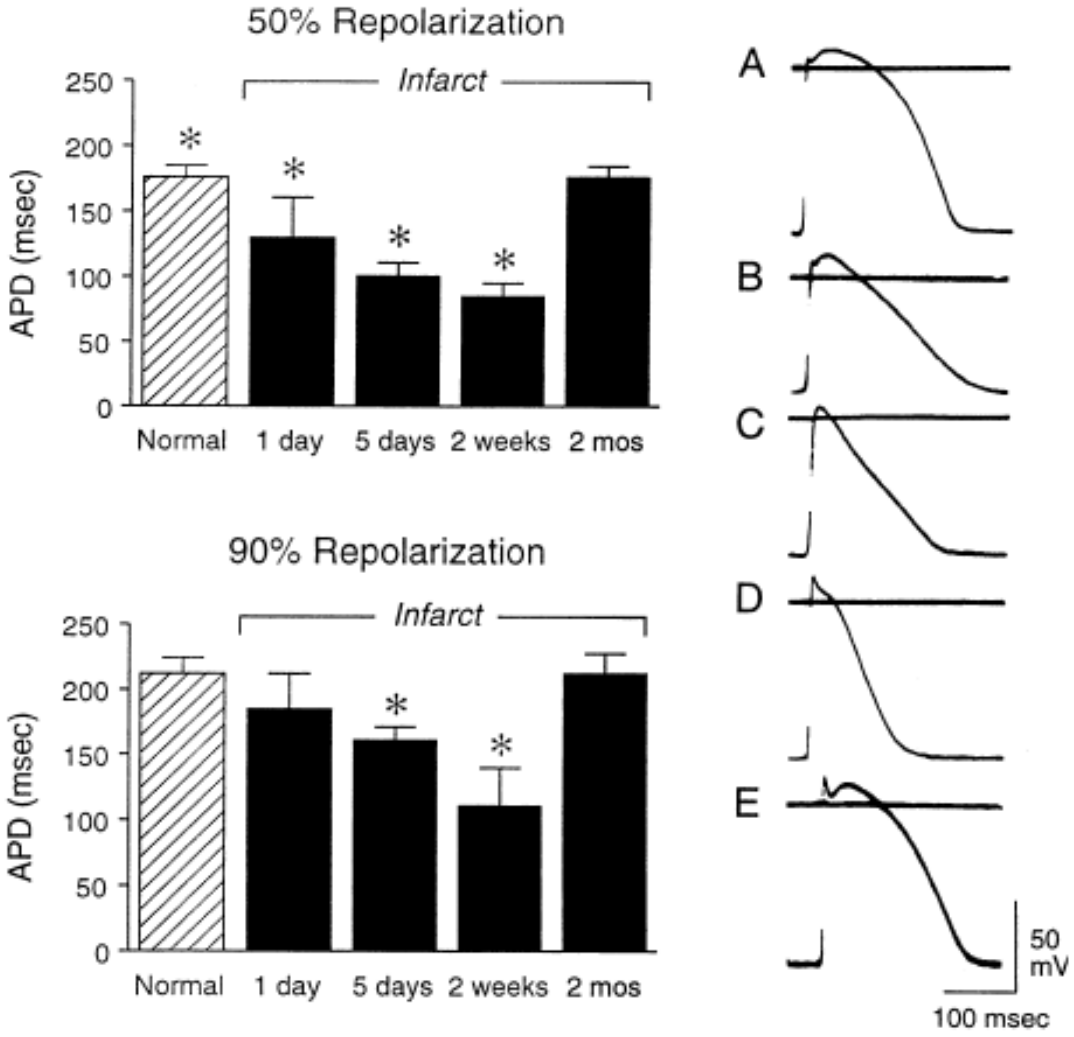
Refractoriness heterogeneity
- Heterogeneous K^+ channel down-regulation
- Post-repolarization refractoriness (I_{Na} down-regulation)

Ectopic complexes
- EADs (decreased K^+ currents)
- DADs (spontaneous Ca^{2+} microreleases)

Ion current and transporter remodeling underlying anisotropic reentry post-MI

Peters NS, e t al. Circulation 1997;95:988–996.

Electrical remodeling in ischemia and infarction



Down regulation

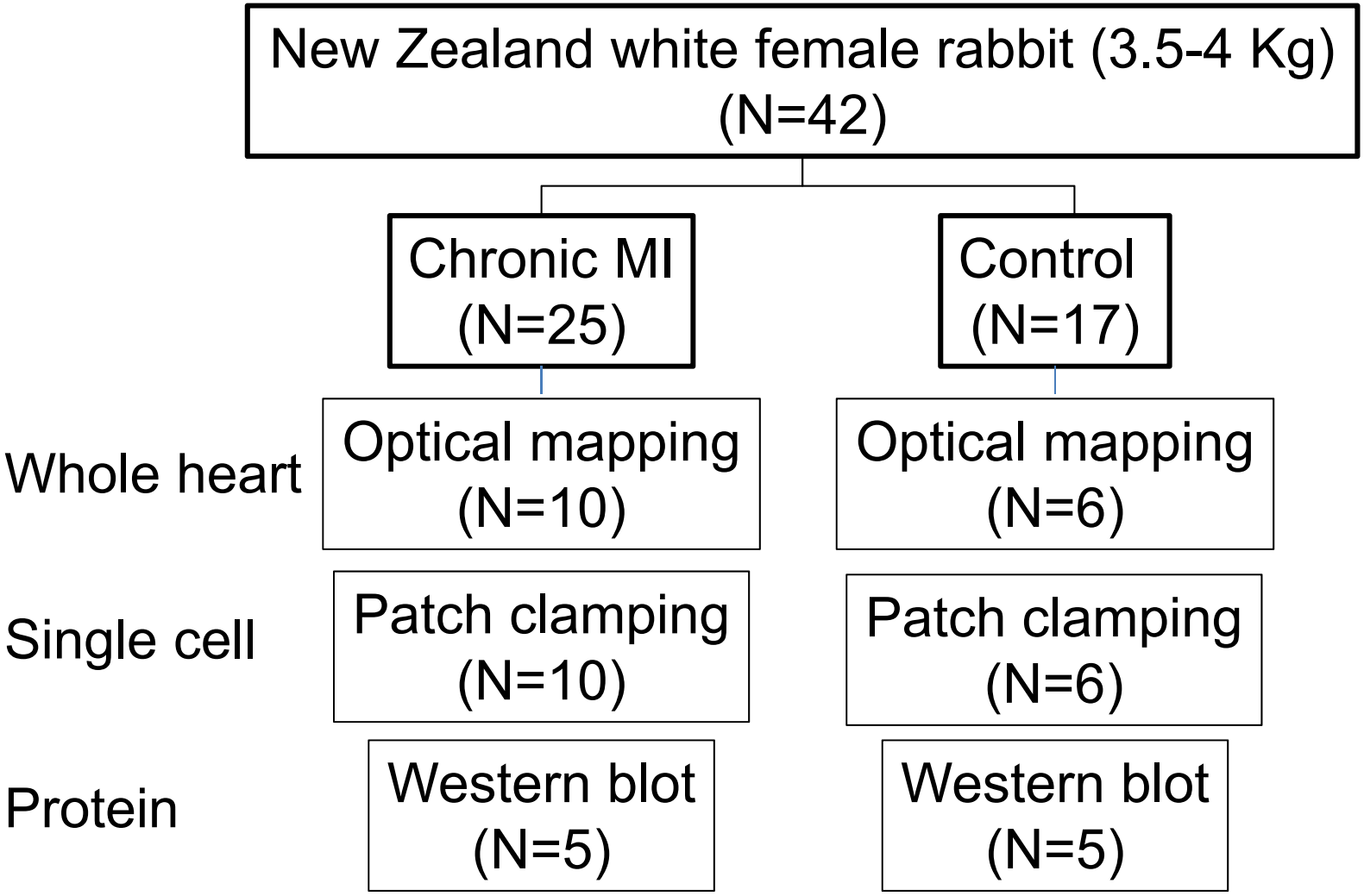
- I_{Na}, I_{to}, I_{Kr}, I_{Ks}, I_{CaL}

Ursell PC, et al. Circ Res, 1985;56:436-451

Purpose

To perform optical mapping studies and patch clamp studies to test the hypothesis that there is up-regulation of I_{KAS} in rabbit ventricles with chronic MI, and that I_{KAS} contributes significantly to ventricular repolarization in chronic MI ventricles.

Method


New Zealand white female rabbit (3.5-4 Kg)
(N=42)
Chronic MI
(N=25)
Control
(N=17)
Whole heart
Optical mapping
(N=10)
Optical mapping
(N=6)
Single cell
Patch clamping
(N=10)
Patch clamping
(N=6)
Protein
Western blot
(N=5)
Western blot
(N=5)

Creation of MI

(A)

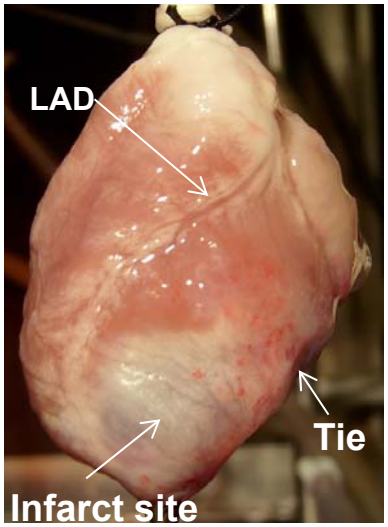
Baseline

pECG

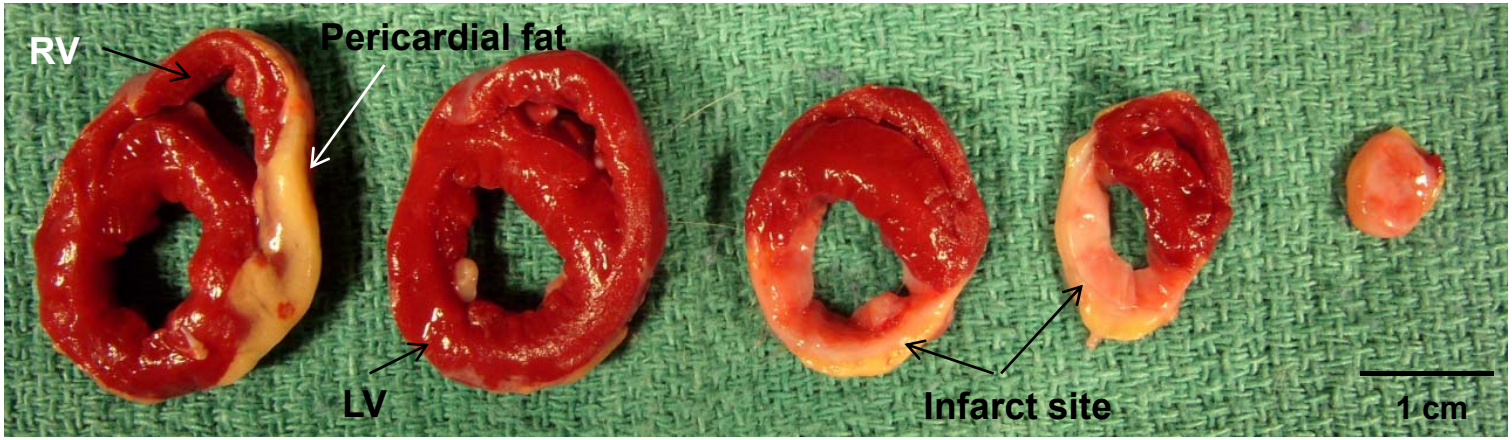
After ligation of coronary artery

200ms

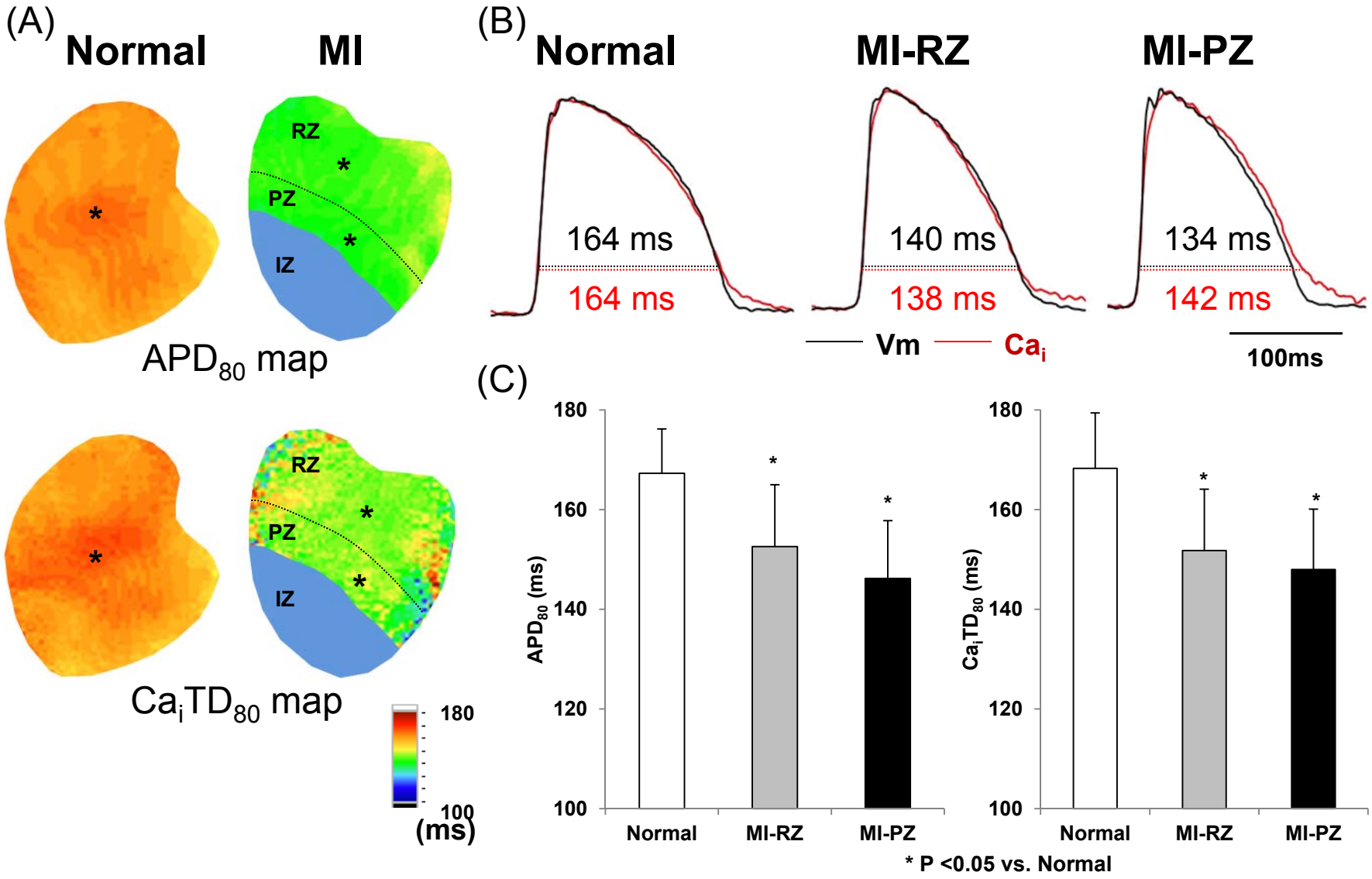
(B)



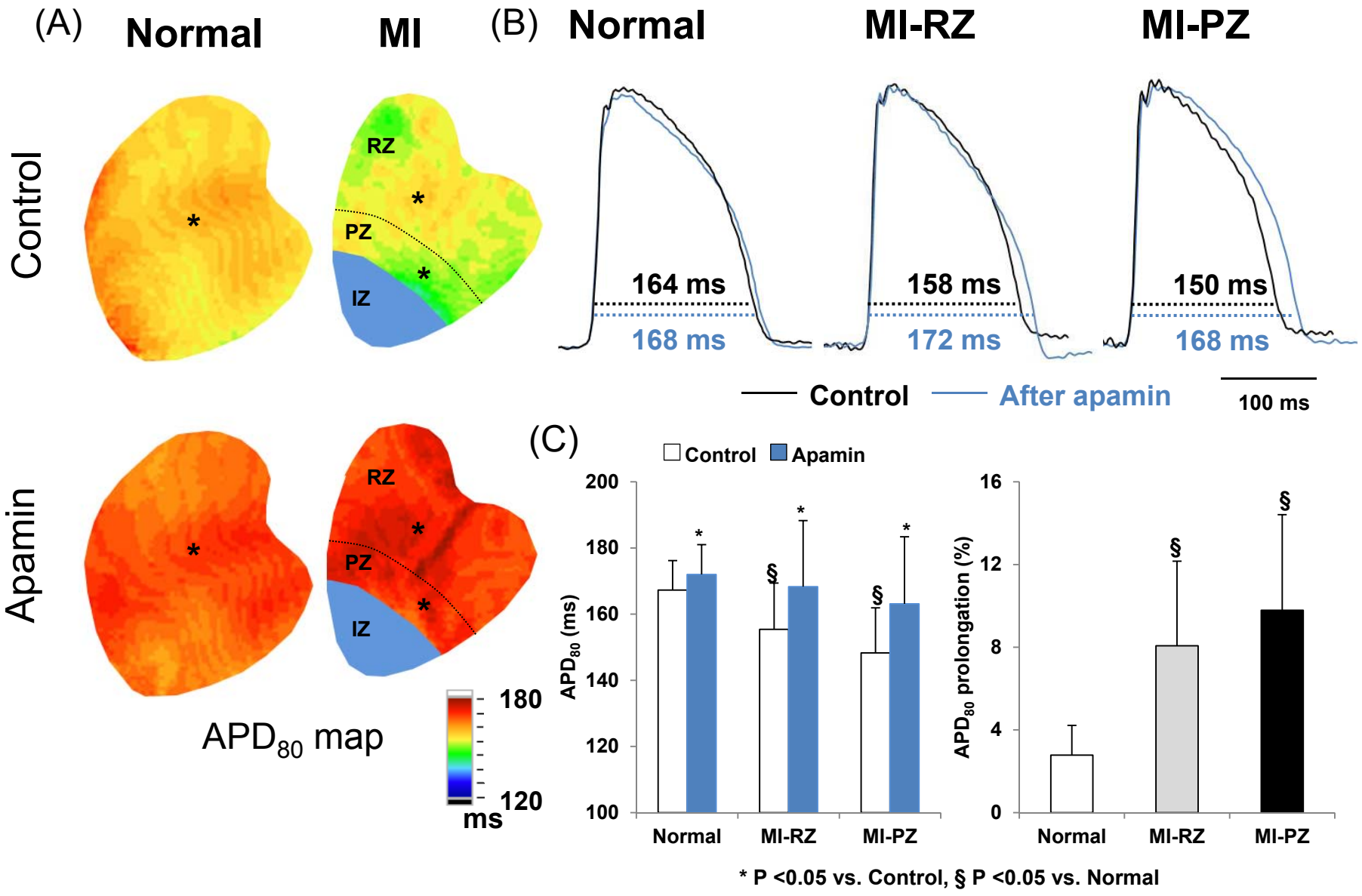
(C) After 1% triphenyl tetrazolium chloride staining



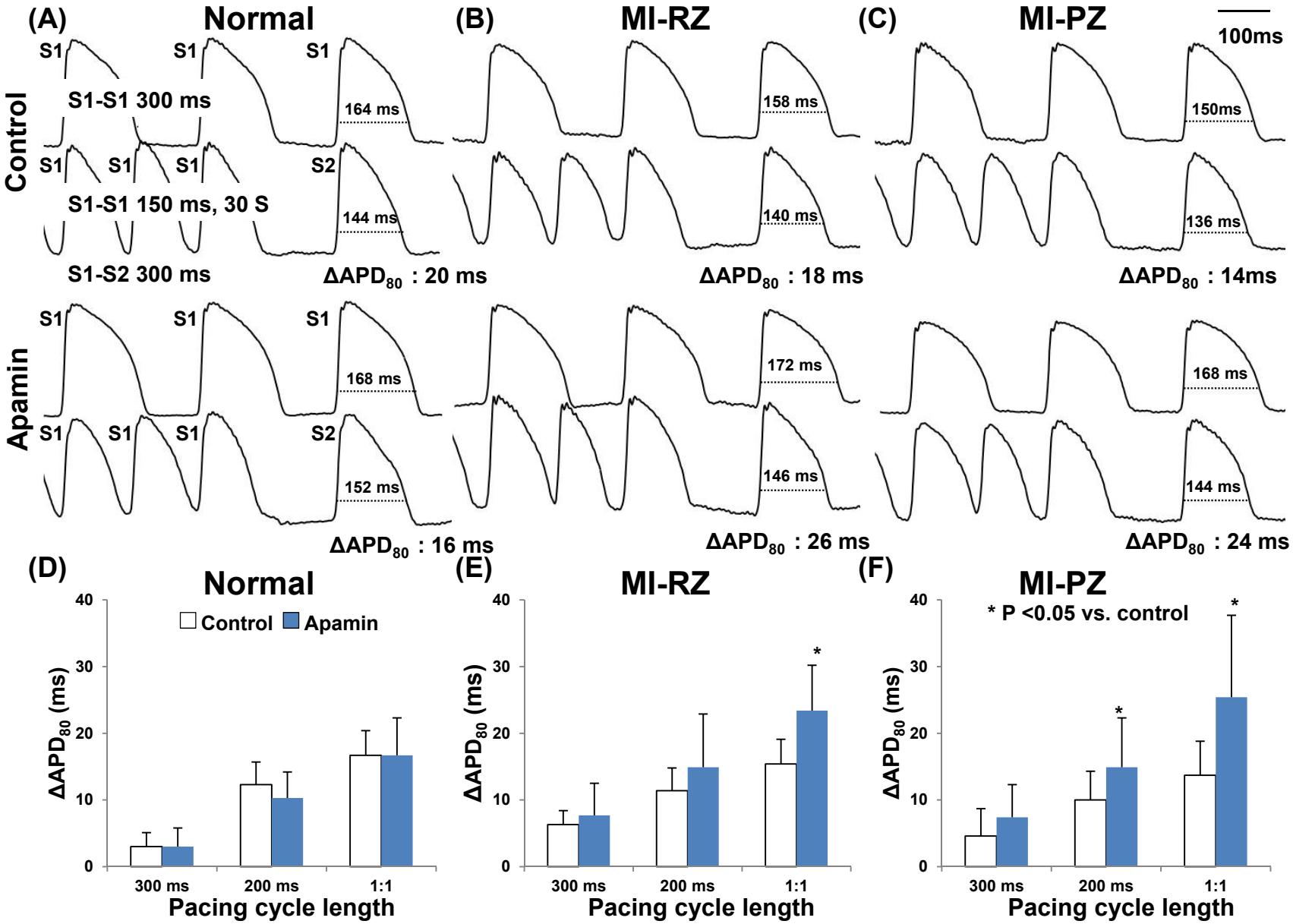
APD in the normal and MI rabbit ventricles



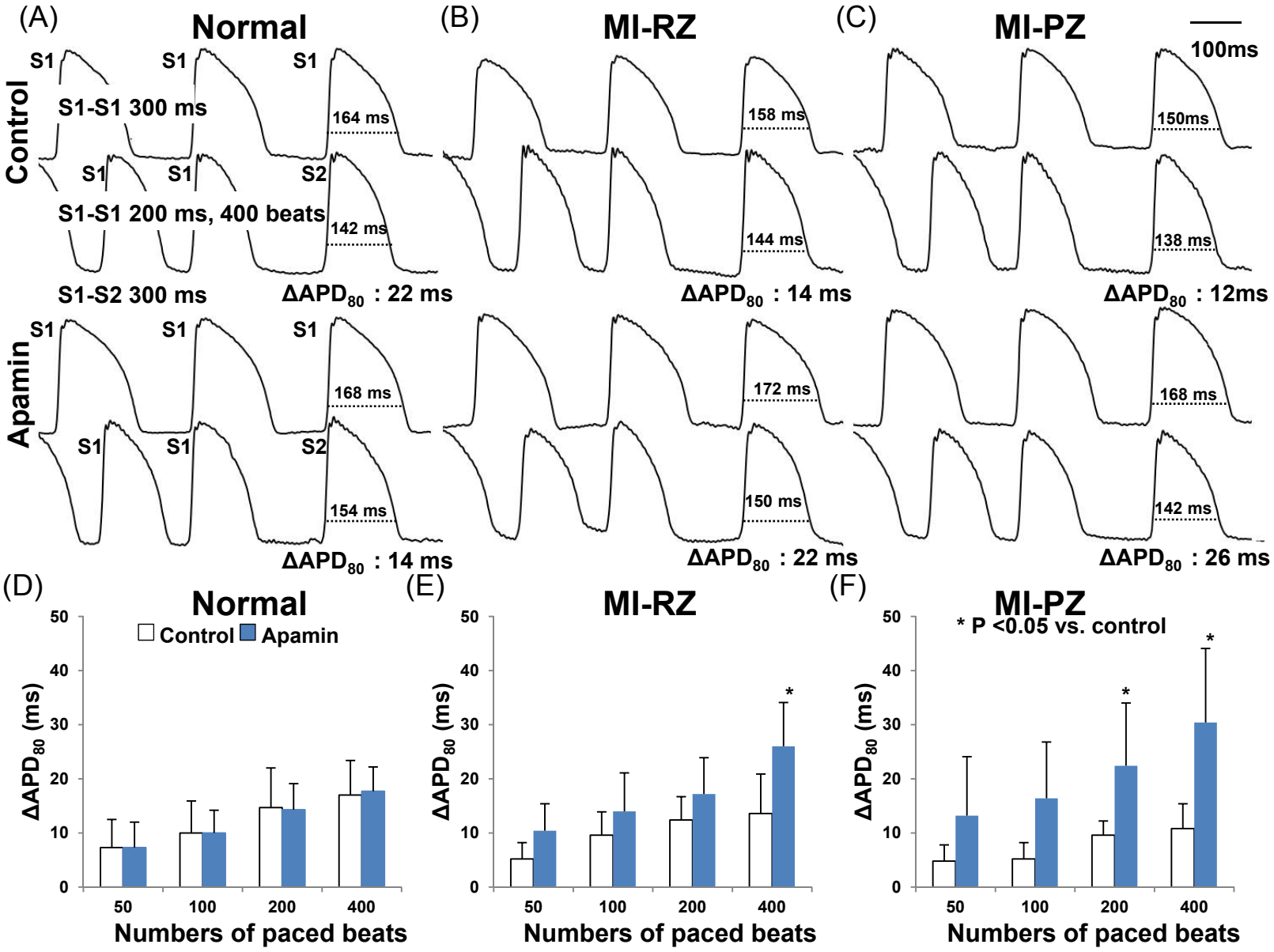
Effect of apamin on APD



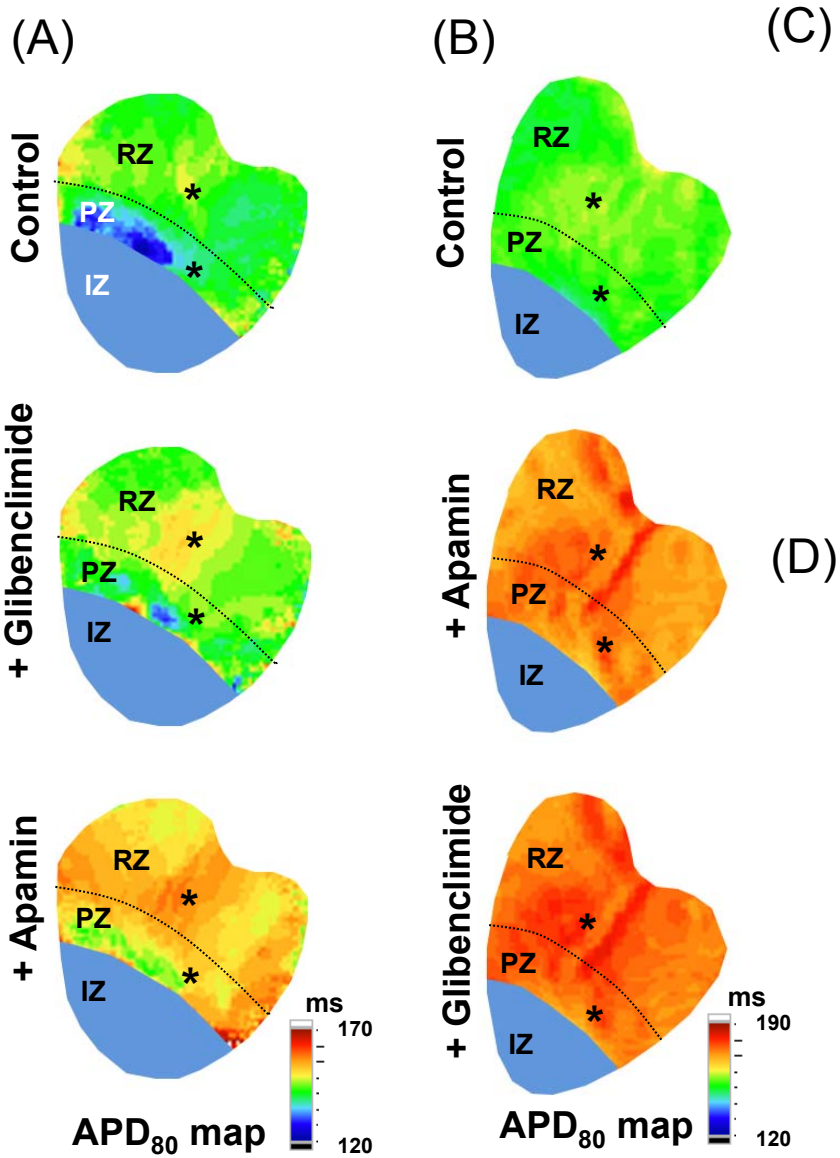
Effect of apamin on ΔAPD according to PCL

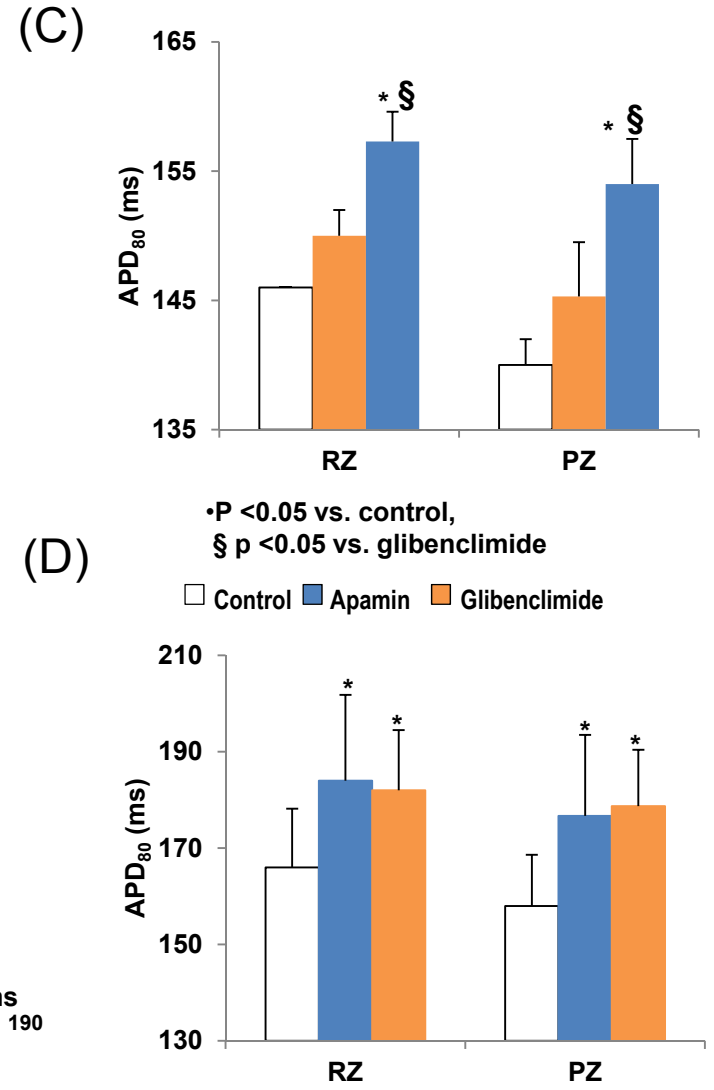


Effect of apamin on ΔAPD according to pacing No.

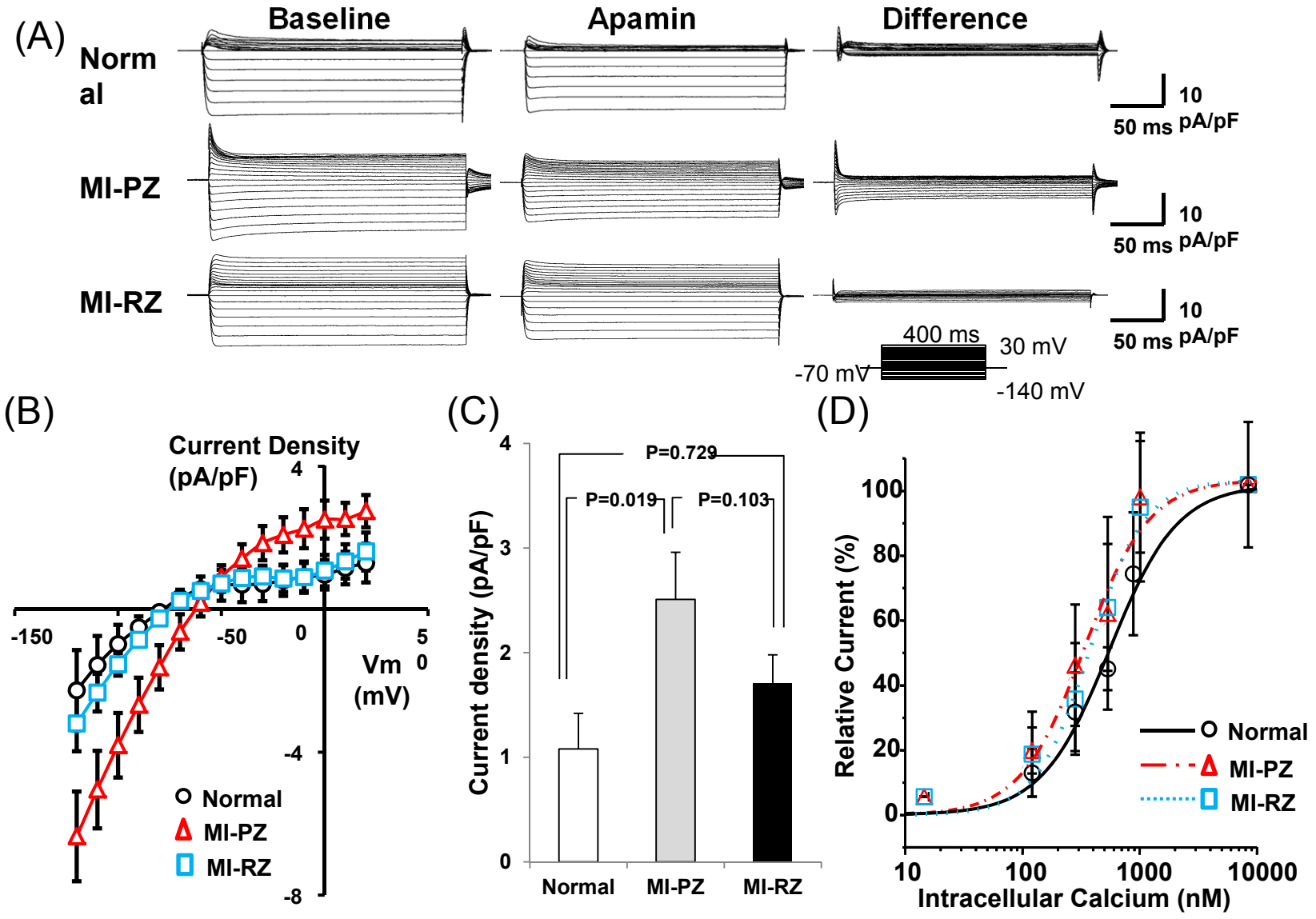


Effect of K_{ATP} blocker on APD

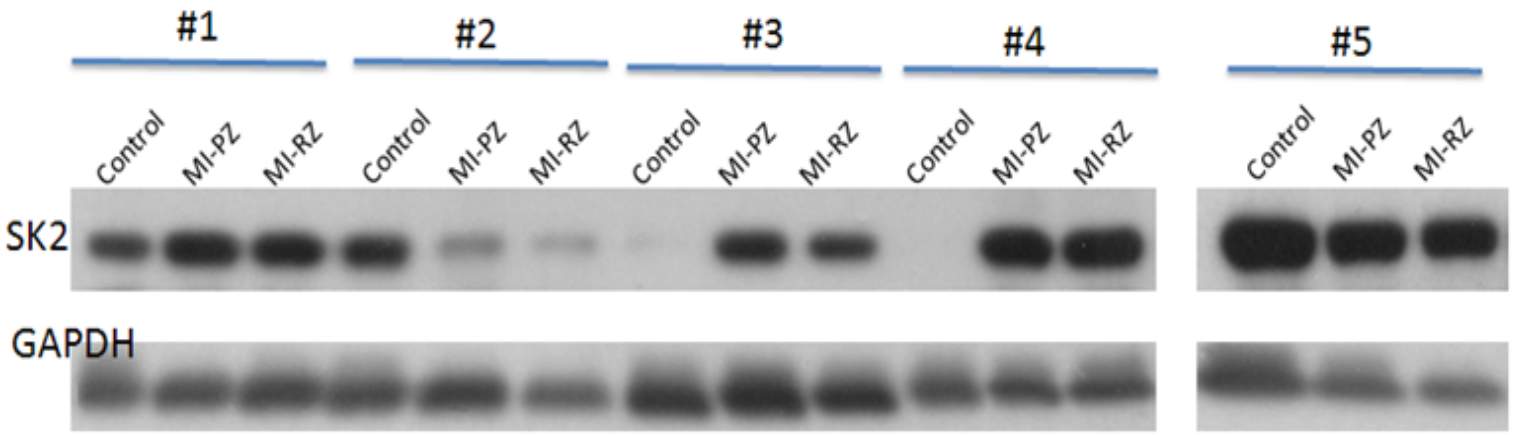


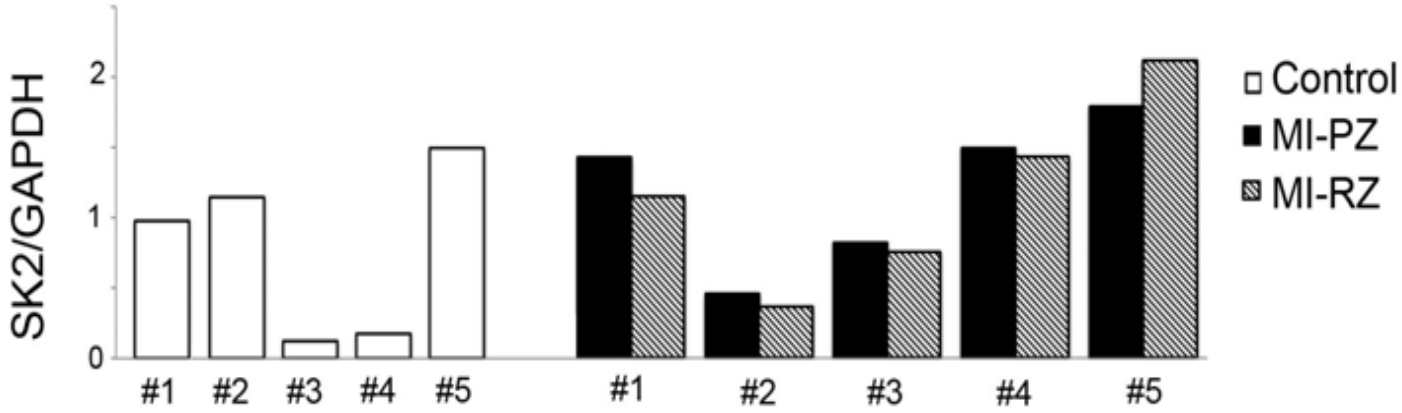


I_{KAS} in MI rabbits



Western blotting of SK2 protein in MI rabbits





Summary (I)

- The APD_{80} and Ca_iTD_{80} in the peri-infarct zone and remote zone were both shorter than the corresponding sites in the normal ventricles.

- Apamin prolonged APD_{80} in normal and MI ventricles, the degree of prolongation was greater in MI than in normal ventricles.

- Apamin did not affect ΔAPD_{80} in normal ventricles, but significantly increased ΔAPD_{80} in MI ventricles.

- There was significant APD_{80} prolongation after apamin, but no additional changes after glibenclamide administration.

Summary (II)

- Mean I_{KAS} density, which is determined as the apamin-sensitive difference current, was significantly larger in peri-infarct zone of MI than in normal ventricular epicardial myocytes.

- Steady-state Ca^{2+} sensitivity of I_{KAS} was leftward-shifted in the MI cells compared to normal cells.

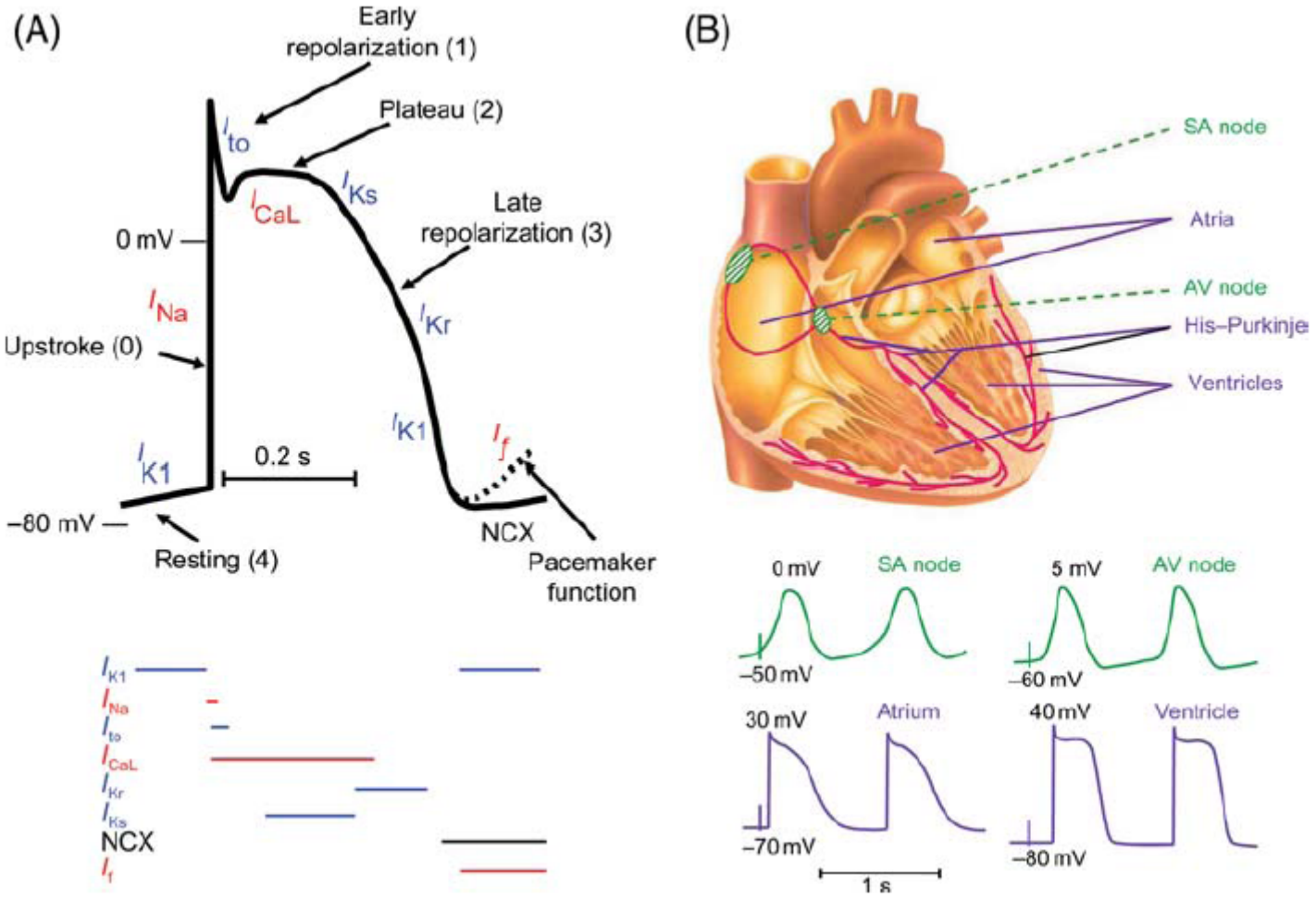
- In terms of SK2 protein, Although there was a trend of increased ratio in peri-infarct zone and remote zone versus control, the difference was not statistically significant.

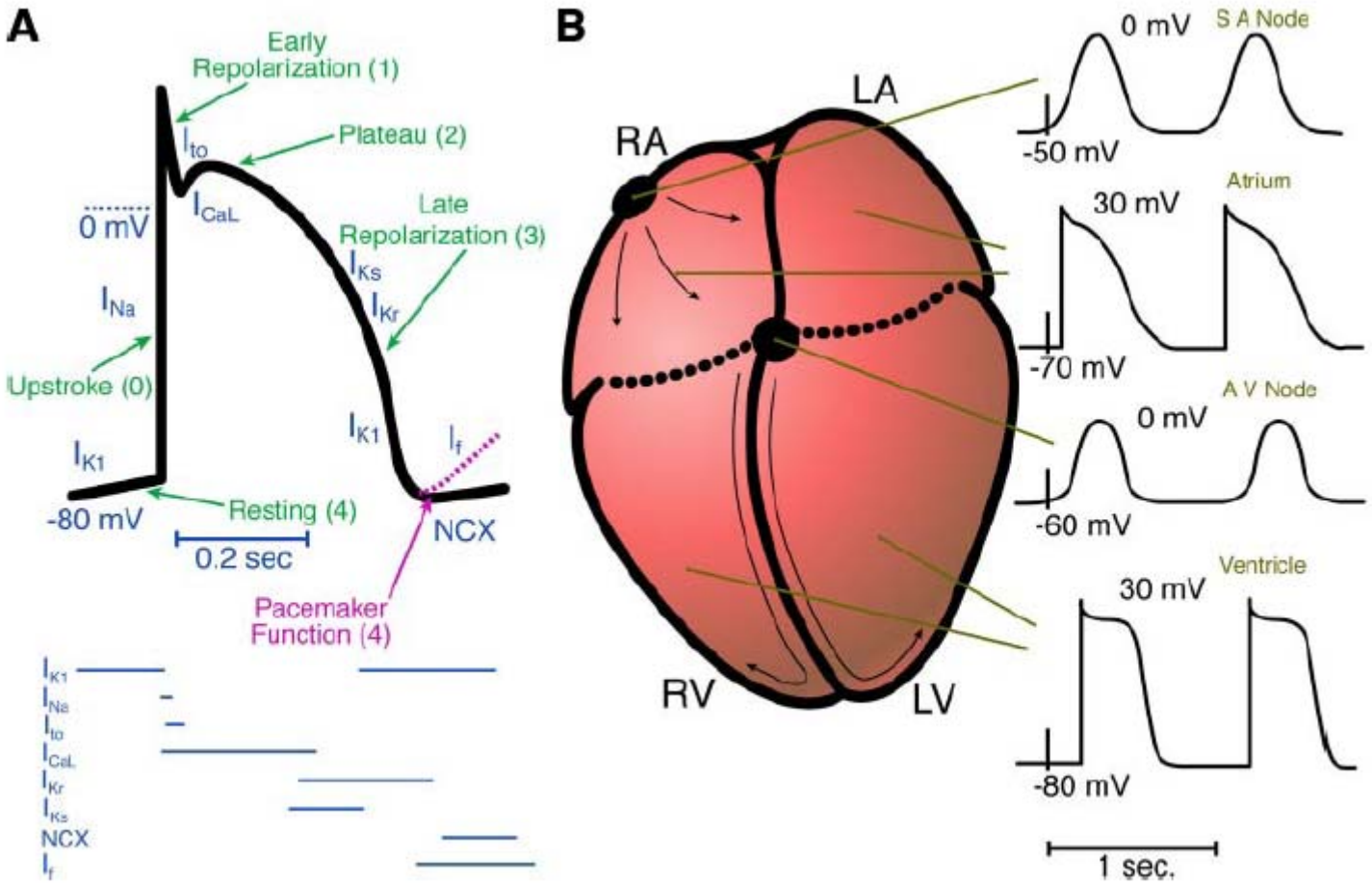
Conclusion

- ✓ Chronic MI is associated with a significantly increased I_{KAS} density and the I_{KAS} sensitivity to intracellular Ca.

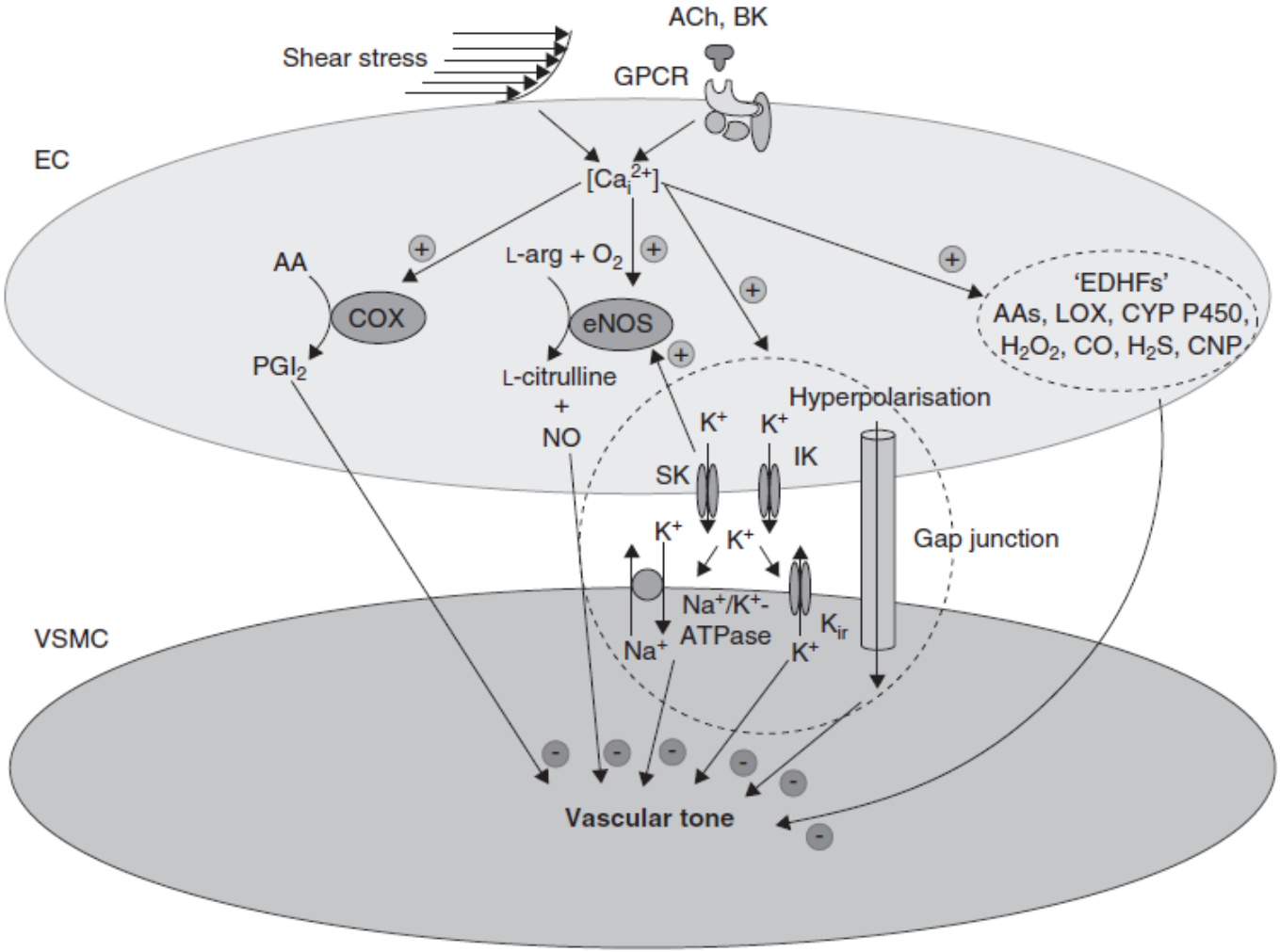
- ✓ I_{KAS} contributes significantly to ventricular repolarization and repolarization reserve in MI ventricles.

Thank you for your attention


(A)
Early repolarization (1)
Plateau (2)
Late repolarization (3)
I_{to}
I_{CaL}
I_{Ks}
I_{Kr}
I_{K1}
I_f
I_{Na}
0 mV
Upstroke (0)
0.2 s
−80 mV
Resting (4)
NCX
Pacemaker function
I_{K1}
I_{Na}
I_{to}
I_{CaL}
I_{Kr}
I_{Ks}
NCX
I_f
(B)
SA node
Atria
AV node
His–Purkinje
Ventricles
0 mV
SA node
5 mV
AV node
−50 mV
−60 mV
30 mV
Atrium
40 mV
Ventricle
−70 mV
−80 mV
1 s


A
Early
Repolarization (1)
Plateau (2)
Late
Repolarization (3)
Upstroke (0)
Resting (4)
0.2 sec
Pacemaker
Function (4)
NCX
0 mV
-80 mV
B
RA
LA
RV
LV
0 mV
S A Node
-50 mV
Atrium
30 mV
-70 mV
A V Node
0 mV
-60 mV
Ventricle
30 mV
-80 mV
1 sec.

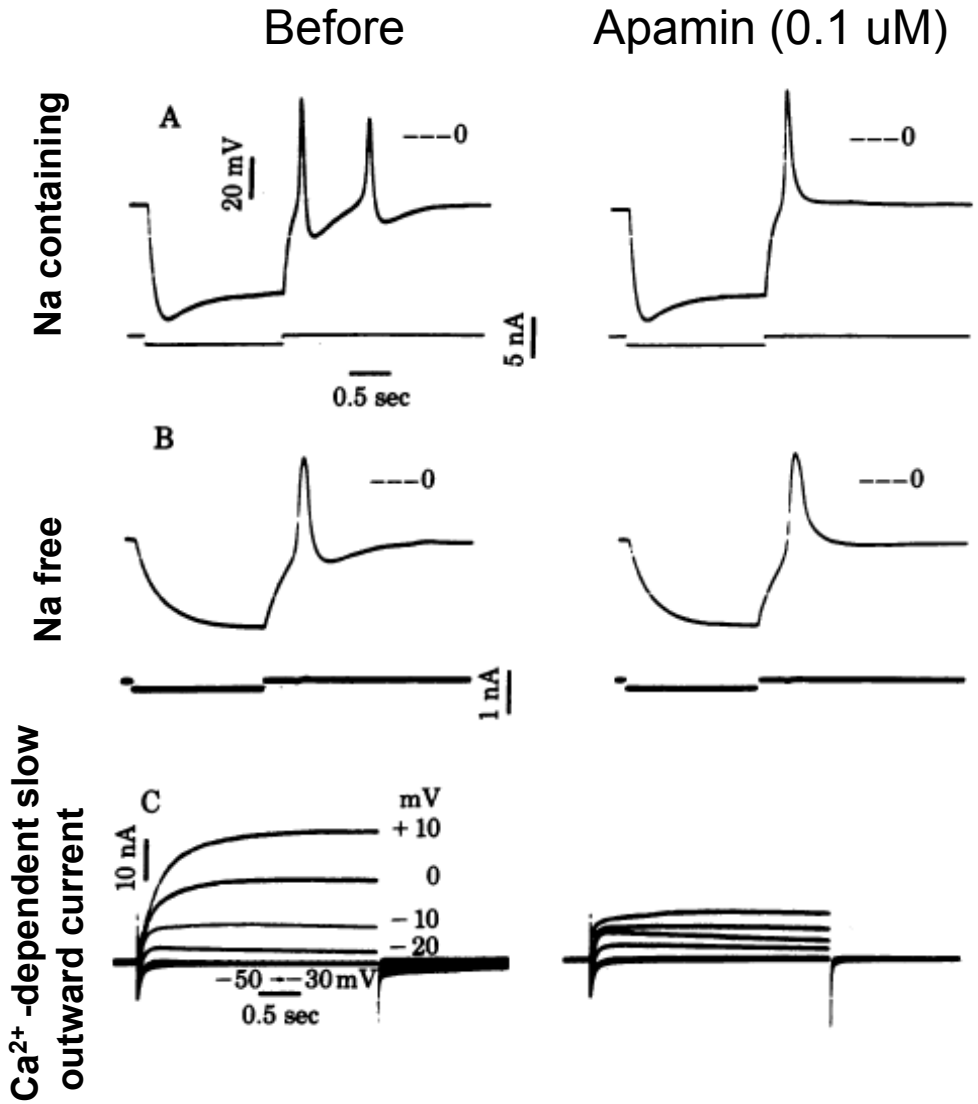
Endothelium-dependent vasodilatation



Blocker and activator of SK, IK and BK channels

Channel	SK			IK	BK
	SK1	SK2	SK3		
Blockers (IC_{50})	Apamin (1 – 12 nM) Leiurotoxin I (325 nM) Tamapin (42 nM) UCL1684 (800 pM) UCL1848 (1 nM)	Apamin (30 – 200 pM) Leiurotoxin I (200 nM) Tamapin (24 pM) UCL1684 (200 pM) UCL1848 (110 pM)	Apamin (1 – 20 nM) Leiurotoxin I (1 nM) Tamapin (2 nM) UCL1684 (10 nM) UCL1848 (2 nM)	Maurotoxin (1 nM) Charybdotoxin (5 nM) TRAM 34 (20 nM) TRAM 39 (60 nM)	Charybdotoxin (5 – 10 nM) Iberiotoxin (2 nM) TEA (200 μM) Paxilline (2 nM)
Activators (IC_{50})	DC-EBIO (25 μM) 1-EBIO (650 μM) NS309 (600 nM) SKA-31 (3 μM) GW542573X (8 μM)	DC-EBIO (27 μM) 1-EBIO (450 μM) NS309 (620 nM) SKA-31 (2 μM) CyPPA (13 μM)	DC-EBIO (12 μM) 1-EBIO (87 μM) NS309 (120 nM) SKA-31 (3 μM) CyPPA (4 μM)	DC-EBIO (750 nM) 1-EBIO (24 – 80 μM) NS309 (27 nM) SKA-31 (260 nM)	NS1608 (4 μM) NS1619 (4 μM) NS11021 (400 nM) BMS204352 (352 nM) BMS223131 BMS191011

Identification of SK channel in Neurons



Hugues, M., et al. Proc. Natl. Acad. Sci. USA, 1982;79:1308-1312.

Ventricular arrhythmia storms in postinfarction patients with implantable defibrillators for primary prevention indications: A MADIT-II substudy

(Heart Rhythm 2007;4:1395–1402)

BACKGROUND Much of prognostic implications of ventricular arrhythmia storms remain unclear.

OBJECTIVE We evaluated the risk associated with electrical storm in patients with defibrillators in the Multicenter Automatic Defibrillator Implantation Trial II (MADIT-II) study.

METHODS Electrical storm was defined as ≥3 episodes of ventricular tachycardia (VT) or ventricular fibrillation (VF) in 24 hours.

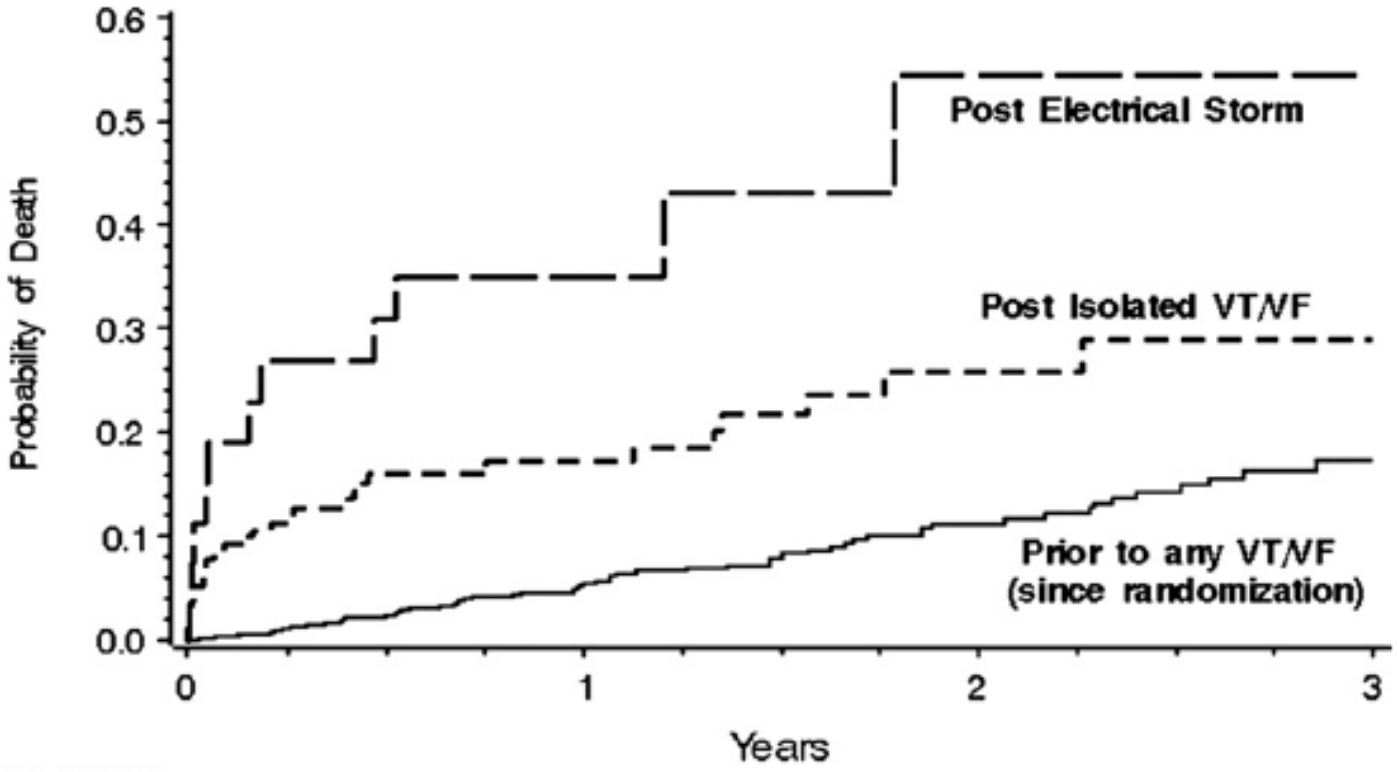
RESULTS Of the 719 patients who received internal cardiac defibrillator (ICD) implants and had follow-up in the MADIT-II, 27 patients (4%) had electrical storm, 142 (20%) had isolated episodes of VT/VF, and the remaining 550 patients had no ICD-recorded VT events. Baseline clinical characteristics among the groups were similar. Patients who experienced electrical storm had a significantly higher risk of death. After adjustments for relevant clinical covariates, the hazard ratio (HR) for death in the first 3 months after the storm event was 17.8 (95% confidence interval [CI] 8.0 to 39.5, $P < .01$) in comparison with those with no VT/VF. This risk continued even after 3 months for those with electrical storm (HR of 3.5, 95% CI 1.2 to 9.8, $P = .02$). Study patients with isolated VT/VF episodes also were at an increased risk of dying (HR = 2.5, 95% CI 1.5 to 4.0, $P < .01$) when compared with patients without VT/VF episodes. Statistically significant predictors of electrical storm were interim postenrollment coronary events (myocardial infarction or angina) HR 3.1 (95% CI 1.2 to 8.1, $P = .02$) and isolated VT or VF HR 9.2 (95% CI 4.0 to 20.9, $P < .01$).

CONCLUSION Postinfarction patients with severe left ventricular dysfunction in whom electrical storm developed have significantly higher mortality than patients with only isolated VT/VF as well as those without any episodes of VT/VF. Patients who experienced postenrollment ventricular arrhythmias and/or interim coronary events during follow-up were at higher risk for VT/VF storms.

KEYWORDS Ventricular tachycardia; Ventricular fibrillation; Coronary artery disease; Congestive heart failure; Implantable defibrillators

(Heart Rhythm 2007;4:1395–1402)

Survival in post-MI



PATIENTS AT RISK				
Electrical Storm	27	9 (0.35)	3 (0.55)	2 (0.55)
Isolated VT/VF	156	67 (0.17)	28 (0.26)	6 (0.29)
Prior to any VT/VF	719	419 (0.06)	206 (0.11)	75 (0.17)