Stretch Differentially Affects Ventricular Epicardial and Endocardial Action Potential Duration in the Rat

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INTRODUCTION

TREK-1 (KCNK2 or $K_{2P}2.1$) is a mechano-sensitive member of the 4T2P (4 transmembrane domain 2 pore) family of K^+ channels. Previously we showed by real time PCR that TREK-1 is differentially expressed in left ventricular epicardial and endocardial regions of the rat heart and hypothesised that it may play a role in dispersion of repolarisation.

AIM

In these experiments we aimed to determine the transmural effect of stretch on monophasic action potential (MAP) duration.

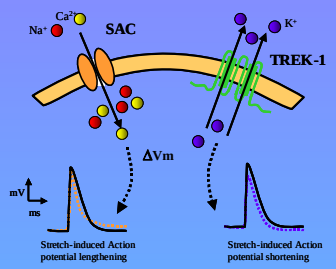

Figure 1

Overview of pathways involved in mechano-electric feedback (MEF) in the cardiomyocyte

MATERIALS AND METHODS

LANGENDORFF PERFUSION:

Seven male Sprague-Dawley rats (466 ± 7 g) were anaesthetized (i.p., Ketamine hydrochloride (75 mg/kg), medatomadine hydrochloride (0.5 mg/kg) and the hearts rapidly excised into ice cold HEPES buffer. Hearts were cannulated on an 18 gauge needle and retrogradely perfused at a constant perfusion pressure of 60 mmHg with bicarbonate buffered solution. Hearts were paced through the right atrium at twice threshold (5 Hz) with 1ms bipolar pulses. A fluid-filled latex balloon was introduced into the left ventricle through the incised left atrium. Left ventricular diastolic and systolic pressure from which left ventricular dP/dt was continuously derived via a pressure transducer. Following stabilization and baseline recordings, the balloon was inflated from baseline (0-5 mm.Hg; no stretch) with water until the desired diastolic pressures were obtained (20-25 mm.Hg; moderate stretch and 50-55 mm.Hg; elevated stretch).

SOLUTION DETAILS

BICARBONATE PERFUSATE was equilibrated with 95% O2 and 5% CO_2 and consisted of (in mmol/L) $CaCl_2$ 1.5, NaCl 111.0, KCl 4.0, $MgCl_2$ 0.6, $NaHCO_3$ 23.9, NaH_2PO_4 1.2, D-glucose 12.0 at 37ºC

Hepes Buffer contained (in mmol/L) NaCl 133.5, NaH2PO4 1.2, KCl 4, HEPES 10.0, MgCl2 1.2, and D-glucose 11 (pH 7.4 0 with NaOH).

MATERIALS AND METHODS Contd.

VENTRICULAR MONOPHASIC ACTION POTENTIAL RECORDINGS:

MAP recordings were made via transmural electrodes consisting of stainless steel acupuncture needles (0.3mm in diameter, Brand, Aust.) insulated (except for the tip) relative to an indifferent (small AgCl ball ~1mm in diameter) placed on the surface of the heart **Figure 2**. Following each stepwise change in diastolic pressure, the MAP electrode was reinserted and held against the heart using a micromanipulator. MAP electrograms were amplified, digitised at a rate of 2 kHz, displayed and analysed using Chart V and peak parameter software (ADInstruments, Australia) **Figure 3**. Action potential durations at 20, 50 and 80% repolarisation were calculated.

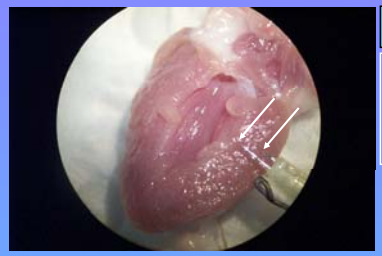

Figure 2

The MAP recording electrodes against the thickness of the left ventricular free wall of the rat heart.

RESULTS & STATISTICAL ANALYSES

Action potential durations at 20, 50 and 80% repolarisation were analysed using general linear model analysis of variance (GLM-ANOVA) followed post hoc by Tukey's pair-wise comparisons.

• APDs did not differ between no stretch and moderate stretch for APD_{20} and APD_{80} (p=0.09 and 0.35 respectively) however at APD_{50} during moderate stretch were significantly shorter than control (no stretch) **Figure 4**.

• Expressed relative to control levels elevated stretch generated action potentials that were significantly shorter for both epi- and endocardium APD_{50}: 92% and 69%, APD_{80}: 85% and 58% respectively **Figure 5**.

• The shortening was significantly greater for endocardium (p<0.001 and p<0.001 for APD_{50} and APD_{80} respectively).

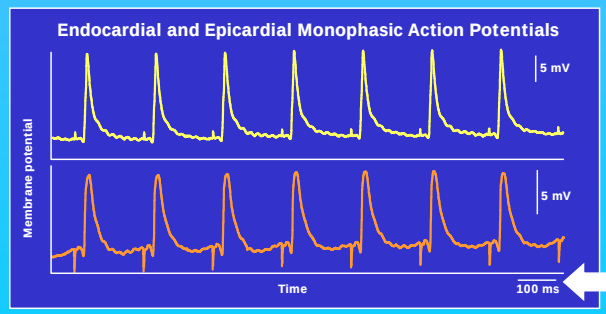

Figure 3

RESULTS & STATISTICAL ANALYSES Contd.

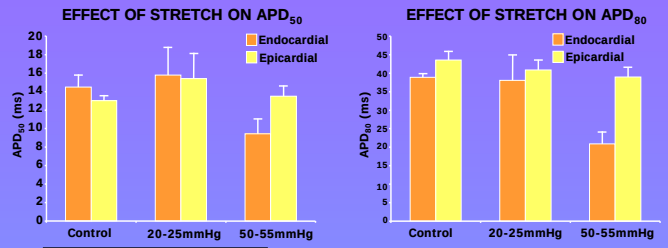

Figure 4

Figure 4: shows the relationships between epicardial and endocardial under conditions of no stretch, moderate stretch and elevated stretch at 50 and 80% repolarisation.

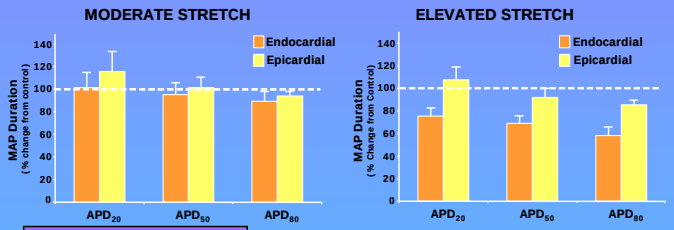

Figure 5

Figure 5: shows the relationship between epicardial and endocardial at 20, 50 and 80% repolarisation relative to control under conditions of moderate stretch and elevated stretch. Horizontal dotted line represents control, no stretch.

CONCLUSION

These results provide support for stretch activated channels being differentially distributed in epicardium and endocardium. The further suggest a potential role for these channels in inducing compensatory changes following altered haemodynamics in the heart.

References

Tan JH, Liu W and Saint DA. Differential expression of the mechanosensitive potassium channel TREK-1 in epicardial and endocardial myocytes in rat ventricle. (2004) *Exp Physiol.* 89(3):237-42.

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