increase in Ca2+ transient amplitude (84 ± 1.5 %; n=218) that reached steady-state approx. 3 hrs post-treatment. ConT increased the activation of Akt as indicated by phosphorylation of Akt (p-Akt) on Ser473 at 15 min and remained elevated relative to non-treated VMs at 3 hrs. A faster migrating p-Akt immunoreactive band was also identified. This putative Akt-cleavage product was not seen with insulin (10 μM) stimulation of p-Akt and not prevented with inhibitors of caspase activity (Z-DEVD-FMK (40 μM), Boc-D-FMK (50 μM)). This activation was not myocyte specific and conT treatment produced similar results in fibroblasts. The results demonstrate a paracrine mechanism of MSCs on improved cardiomyocyte survival through activation the PI3K/Akt pathway that also triggers remodeling of EC coupling. Stimulation of Akt in fibroblasts presents an additional indirect means by which cardiac repair could be modulated following injury.

2840-Pos

Rac1-Induced Remodelling is Different in the Left and Right Atrium of RacET Mice

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In human atrial fibrillation (AF), increased levels of reactive oxygen species, increased activity of the NADPH oxidase and increased expression of the small GTPase Rac1 have been reported. In a RacET transgenic mouse line, spontaneous AF occurs at ages >12 weeks. Here, we characterise atrial remodelling processes that take place in calcium handling and ion current. We used 4-6 months old male mice and age-matched control mice. Echocardiography indicated mildly enlarged left atria while the right atria displayed several fold dilation. The analysis of whole cell calcium transients revealed that although both cell types displayed reduced calcium transient amplitudes, basal calcium concentration was significantly increased only in cells of the right atrium. Post-rest behaviour was unchanged. While in the left atrium (LA) caffeine responses were unchanged in amplitude but SERCA activity was down by almost 50%, cells from the right atrium (RA) showed significantly decreased caffeine signals (by 40%) and increased Na/Ca exchanger with an unchanged SERCA pump activity. Calcium sparks also displayed striking differences. While in RA their amplitude was reduced, recovery was slowed down and spatial spread was only slightly decreased (by 10%), cells from LA showed an increased amplitude (by 40%), faster recovery (up 20%) and a decreased spatial spread (down by >25 %). Action potentials recorded from LA and RA cells showed a slightly more negative resting membrane potentials in the myocytes from RacET with otherwise unchanged properties. From these data we conclude that Rac1 overexpression and the accompanying increased oxygene radical load results in atrial remodelling that is significantly different between the left and the right atrium.

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2841-Pos

Increased Susceptibility to Ventricular Arrhythmias Relates to Diastolic Ca^{2+} Leak in Rats with Metabolic Syndrome

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Introduction:
Rats genetically selected and bred for low aerobic capacity (LCR) develop characteristics resembling to the metabolic syndrome and animals with heart failure. Rats selected for high aerobic capacity (HCR) develop athletic characteristics. We hypothesized that LCR rats are more susceptible for ventricular arrhythmias and that this relates to increased diastolic SR Ca^{2+} leak.
Methods:
We included 10 LCR and 10 HCR rats. We assessed susceptibility to ventricular fibrillation by burst pacing in Langendorff perfused hearts. In isolated cardiomyocytes we measured Ca^{2+}-handling with FURA2/AM and action potential by di-8-ANEPPS on an inverted epi-fluoresence microscope.
Results:
LCR rats were more susceptible for ventricular fibrillation and monophasic action potential at the left ventricle was prolonged. Prolongation of the action potential was confirmed in single di-8-ANEPPS loaded cardiomyocytes. Cardiomyocyte function was significantly depressed in LCR rats compared to HCR; fractional shortening was 37% lower and time to 50% relenghtening was 53% longer. Ca^{2+}-handling was impaired by elevated diastolic Ca^{2+}, reduced Ca^{2+} amplitude and prolonged time to 50% Ca^{2+} decay. SR Ca^{2+} content was 21% lower and fractional Ca^{2+} release was 10% lower in LCR. During caffeine induced Ca^{2+} transient, we found no difference in Ca^{2+} decay between the two groups, reflecting unaltered NCX function; nor did we find any changes in the plasma membrane Ca^{2+}, assessed by caffeine induced transients in a $0Na^{+}$/$0Ca^{2+}$ solution. Diastolic Ca^{2+} removal is mainly attributed to reduced SERCA function by 15% in LCR rats. By measuring diastolic Ca^{2+} in quiescent myocytes with and without tetracaine we found 46% increased SR Ca^{2+} leak LCR rats.
Conclusion:
Impaired Ca^{2+} handling and increased diastolic SR Ca^{2+} leak together with prolonged action potential duration may explain increased susceptibility to ventricular arrhythmias in LCR rats with metabolic syndrome.

2842-Pos

Effects of Membrane Calcium Flux Localizations and Realistic T-Tubule Geometry on Cardiac Excitation-Contraction Coupling

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The sub-cellular anatomy and ionic flux distributions are considered important in calcium (Ca) regulation and their variations over time may hold clues to the progression of heart diseases. To investigate how these coupled processes may affect the cardiac cell function we developed 3D continuum model of Ca-signaling, buffering and diffusion in rat ventricular myocytes. The model geometry was derived from high definition light and electron microscopy images in rodent cardiac cells through a process called tomographic reconstruction [1, 2]. The current model includes: the 3D geometry of a single t-tubule and its surrounding half-sarcomeres; spatially distributed Ca handling proteins along the t-tubule and surface membrane; stationary and mobile Ca buffers (ATP, calmodulin, fluo-3, troponin C). A finite element software package SMOL-SubCell was used to solve the PDE system on cluster of computers [3]. In agreement with experiment [4,5], model suggests that the rat t-tubule anatomy and the heterogeneous distribution of Ca fluxes along the cell membrane might be important mechanisms for maintaining uniform Ca concentration in presence of 100 microM fluo-3 and sarcoplasmic reticulum inhibited. In the absence of fluo-3, model predicts that the overall Ca distribution can not be maintained uniform when the membrane Ca fluxes were heterogeneously distributed.

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2843-Pos

The Generation of a Novel Animal Model of Inducible Hypertrophy: Overexpression of NCX1 in the Murine Heart Using the Doxycycline-Dependent Promoter

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The Na^{+}/Ca^{2+} exchanger (NCX1) is essential regulator of Ca^{2+} homeostasis in cardiomyocytes. In normal cardiac muscle, the primary role of NCX1 is to extrude cytoplasmic Ca^{2+} during myocyte repolarization and diastole. In hypertrophic and failing hearts, sarcolemmal NCX1 expression has often been shown to be elevated, which could be compensatory for the reduced ability of the sarcoplasmic reticulum to maintain low diastolic $[Ca^{2+}]_i$ under these pathological conditions. However, whether increased NCX1 expression invariably leads to enhanced function under disease conditions is not clear. In this study, we have produced a line of the transgenic mice in which expression of the canine cardiac NCX1 transgene was induced using the doxycycline (DOX) dependent promoter. After the injection of 100 μL of DOX (2 mg/mL in normal saline solution) into the abdominal cavity per 1 day during 2 weeks, the expression of canine NCX1 protein in the hearts of the DOX-treated transgenic mice was 3-fold higher, but the protein levels involved in excitation-contraction coupling, sarcoplasmic reticulum Ca^{2+}-ATPase, phospholamban, ryanodine receptor, and L-type Ca^{2+} channel were unchanged compared with those of wild-type (WT) and the non-induced transgenic mice. The total heart-to-body weight ratio was increased by 45% in the DOX-treated transgenic mice compared with WT or non-induced mice. The DOX-treated transgenic mice developed cardiac hypertrophy accompanied by elevation of multiple hypertrophic markers, but not by the any signs of other typical heart failures. We suggest that the overexpression of NCX1 is sufficient to develop cardiac hypertrophy and these mice would be useful as the animal model to study on the pathological role of NCX1 in cardiac hypertrophy and progression of cardiac dysfunction on heart failure.