following ischemia-reperfusion, indicating *mg53-/-* mice are more susceptible to CS injury. rhMG53 protein (6 mg/kg) applied intravenously to the wild type mice prior to the onset of CS could protect skeletal muscle injury, as demonstrated by the reduction of CK in serum and Evans Blue positive (injured) muscle fibers. Histochemical studies revealed that rhMG53 treatment could ameliorate pathological changes in mouse skeletal muscle associated with CS. When rhMG53 was examined in a rat model of CS-induced muscle injury, we saw a lesser degree of muscle injury and minimum effect for the exogenous rhMG53 in protection against the development of CS. The lack of effect of rhMG53 in the rat model was likely due to the fact that serum level of endogenous MG53 protein in the rat is more than 20-fold higher than that in the mouse and other animal models. Taken together, our data suggest that rhMG53 can protect ischemia-reperfusion induced muscle injury as a potential therapy for protection against compartment syndrome.

3688-Pos Board B416

The Cathepsin-L Inhibitor CAA0225 Improves Cardiac Function During Ischaemia-Reperfusion

Weihong He, Douglas McCarroll, Elspeth B. Elliott, Christopher M. Loughrey.
University of Glasgow, Glasgow, United Kingdom.

Cathepsins are lysosomal cysteine proteases involved in molecular signaling and protein degradation. Importantly, these proteases can also be secreted into the extracellular space. Cathepsin-L is elevated in the serum of patients with ischaemic heart disease and correlates with disease severity. Our data demonstrate that cathepsin-L levels are increased in the coronary effluent of ex vivo rat hearts after a period of 30 min ischaemia, however the contribution of cathepsin-L to cardiac contractile function remains unknown. Using a cathepsin-L inhibitor (CAA0225), we investigated the hypothesis that cathepsin-L contributes to contractile dysfunction post ischaemia. Langendorff perfused ex vivo adult rat hearts under went 30 min whole heart ischaemia followed by 90 min reperfusion. Left ventricular (LV) function was assessed using a solid-state pressure catheter within a balloon in the LV cavity. In separate cohorts of hearts, CAA0225 or DMSO (control) was applied for 25 min prior to ischaemia. CAA0225 had no effect on LV developed pressure prior to ischaemia. However, during reperfusion post-ischaemia CAA0225 led to a sustained increase of LV developed pressure to 177% of control levels at 90 min reperfusion (25.1 *vs.* 44.5 % developed LV pressure of pre-ischaemia level; DSMO [n=15] *vs.* CAA0225 [n=6]; $P<0.05$). The maximum rate of rise and fall in developed LV pressure were also elevated by CAA0225 by 169% and 151% respectively. Whilst there was no significant difference in the minimum LV pressure during the ischaemic period, this parameter was reduced during reperfusion which after 90 min was 68% of control levels (78.0 *vs.* 53.0 mmHg; DSMO [n=15] *vs.* CAA0225 [n=6]; $P<0.05$). These data demonstrate for the first time that the cathepsin-L inhibitor CAA0225 improves both systolic and diastolic cardiac functional parameters during ischaemia-reperfusion in ex vivo adult rat hearts.

3689-Pos Board B417

Altered Ca Homeostasis and Ca Alternans in Left Atrial Myocytes of Spontaneously Hypertensive Rats

Cornelia F. Pluteanu, Judit Preisenberger, Johannes Heß, Yulia Nikonova, Jelena Plackic, Jens Kockskämper.
Philipps-Universität Marburg, Marburg, Germany.

Chronic hypertension can cause atrial fibrillation. The cellular mechanisms responsible for hypertension-induced atrial tachyarrhythmias, however, are unknown. Here we tested the hypothesis that, in spontaneously hypertensive rats (SHR), altered atrial Ca homeostasis may induce atrial tachyarrhythmias. At 6-8 months of age, SHR had significantly increased systolic blood pressure (185 ± 6 mmHg, N=18; vs WKY: 129 ± 4 mmHg, N=16, $P<0.01$), accompanied by compensatory left ventricular hypertrophy, but left atrial (LA) hypotrophy. At the cellular level, the area of the ventricular myocytes was increased in SHR, while LA myocytes were not different in size. Basal Ca transients (CaTs) of LA myocytes (1 Hz stimulation) showed no major differences between WKY and SHR. Further evaluation, however, revealed increased SR Ca load in SHR (amplitude of caffeine-induced CaT, Fura-2 ratio: 1.07 ± 0.13, n=10; vs WKY: 0.72 ± 0.04, n=18, $P<0.01$), reduced fractional SR Ca release (SHR: 0.20 ± 0.03, n=10; vs WKY: 0.29 ± 0.03, n=18, $P<0.05$) and reduced L-type Ca current (SHR: -4.3 ± 0.2 pA/pF, n=12; vs WKY: -5.2 ± 0.1 pA/pF, n=10, $P<0.05$) accompanied by reduced expression (-33%; $P<0.01$) of the alpha 1C subunit of the L-type calcium channel. Experimental simulation of tachycardia by increasing stimulation frequency (1-2-4 Hz) induced significantly more arrhythmogenic Ca alternans in SHR (2 Hz: 41%, 4 Hz: 72% of cells) than in WKY (2 Hz: 7%, 4 Hz 37% of cells; both $P<0.05$ vs SHR) LA myocytes. These data indicate that, at 6-8 months of age, SHR hearts display morphological changes at the ventricular but not at the atrial level. Subcellular changes in Ca homeostasis occur in LA myocytes at the sarcolemma and the SR, which are associated with a much higher incidence of arrhythmogenic Ca alternans. This may explain the increased propensity of SHR to develop atrial tachyarrhythmias.

3690-Pos Board B418

Alterated Intracellular Calcium Ion Regulation Plays Important Role in High Carbohdyrate Intake Induced Myocardial Remodeling

Esma N. Okatan, Aysegul Toy, **Belma Turan**.
Department of Biophysics, Faculty of Medicine, Ankara University, Ankara, Turkey.

Effects of high-sugar diet on systemic metabolism has been informative, however, myocardialcapacity for fructose uptake and metabolism has not yet been well characterized. Importantly, contractile dysfunction has been observed in sucrose-fed animals both at whole-heart and single cardiomyocyte levels. In this study, we aimed to determine whether the properties of the Ca^{2+} sparks might provide insight into the role of ryanodine receptors, RyR2s, in the altered Ca^{2+} signaling and diastolic dysfunction in resting cardiomyocytes when they were analyzed together with Ca^{2+} transients obtained under electrical stimulation. Our results showed that basal Ca^{2+} level as well as the frequency of Ca^{2+} sparks significantly increased in cardiomyoctes from rat heart with high sucrose-induced metacolic syndrome (MS) compared to aged-matched control rats. Ca^{2+} transients exhibited significantly reduced amplitude and prolonged time courses as well as depressed Ca^{2+} loading of sarcoplasmic reticulum, SR in MS animals. The Spatio-temporal properties of these Ca^{2+} sparks were also significantly changed to being almost parallel to the changes of Ca^{2+} transients. In addition, RyR2s from MS rat heart were hyperphosphorylated and the protein levels of both RyR2s and FKBP12.6 depleted. Taken into consideration the whole-data, MS-induced rat heart exhibits altered local Ca^{2+} signaling with increased basal Ca^{2+} levels which may underline the diastolic dysfunction. Furhermore these functional alterations were supported with ultrastructural changes in SR which due to mostly increased oxidative stress in the heart.
(Supported by grant from TUBITAK SBAG-111S042).

3691-Pos Board B419

nNOS-Deficient Lumbricle Muscles Exhibit Normal Fatigue Resistance and Calcium Handling

Wallace G. Kerrick[1], Justin M. Percival[2].
[1]Physiology and Biophysics, University of Miami, Miami, FL, USA, [2]Cellular and Molecular Pharmacology, University of Miami, Miami, FL, USA.

Murine tibialis anterior skeletal muscles lacking one or more isoforms of neuronal nitric oxide synthase (nNOS) exhibit exaggerated contraction-induced fatigue *in vivo* by mechanisms that remain to be elucidated. To test the hypothesis that depletion of skeletal muscle expressed nNOS was sufficient to cause exaggerated fatigue, we analyzed fatigue resistance in excised lumbricle muscles from nNOS knockout (KN2) and wildtype (WT) mice *in vitro*. Simultaneous measurements of skinned fiber Ca^{2+}-activated force and ATPase activity, and simultaneous measurements of intracellular Ca^{2+} and force in isolated intact lumbricle muscle in the nNOS depleted and WT muscles were performed. Results showed that KN2 mice lumbricle skinned fiber force and ATPase activity were more sensitivity to Ca^{2+} than WT, in contrast to no differences in maximum force and ATPase, or energy cost (ATPase/force) at all levels of force activation. Intact lumbricle muscle showed that fatigue produced by increasing the frequency of repetitive 500 millisecond periods of 100 Hz tetanus stimulation showed the average force and intracellular Ca^{2+} during the tetanus was not statistically different between KN2 and WT lumbricle at all levels of fatigue. The average intracellular Ca^{2+} during the tetanus stimulation showed that fatigue was independent of Ca^{2+} except at the very highest fatigue levels. The rate of fatigue in KN2 mouse muscles was not different from WT and we were unable to observe exaggerated fatigue *in vitro*. These results suggest three possible explanations. First, the regulation of muscle fatigue by nNOS *in vivo* may only be evident under physiological conditions. Second, lumbricle muscle lacks an nNOS muscle-specific fatigue resistant mechanism. Third, that nNOS loss from surrounding tissue compromises fatigue resistance. Regardless, these data suggest that nNOS is dispensable for normal calcium handling, strength and fatigue resistance in lumbricle muscles *in vitro.*

3692-Pos Board B420

Cardiac Selective Modulator of Human Myosin for the Treatment of Genetic Hypertrophic Cardiomyopathy

Haben T. Ghermazien[1], Yonghong Song[2], Christopher P. Willits[1], Arvinder S. Sran[2], Robert L. Anderson[1], Hector M. Rodriguez[1], Johan Oslob[2].
[1]Biology, MyoKardia, South San Francisco, CA, USA, [2]Chemistry, MyoKardia, South San Francisco, CA, USA.

Genetic hypertrophic cardiomyopathy (HCM) results from mutations in the cardiac sarcomere, including β-cardiac myosin, with HCM afflicting about