phenotype in mdx mice. These findings suggest that SLN might be a novel target for DMD therapy.

Session 11: Muscle Development, Regeneration and Disease

S11-1

Expression of truncated obscurins leads to maladaptive responses in the heart

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Obscurin was discovered as binding partner of titin and Novex-3, a titin splice variant. Although their direct binding is known for > 15 years, the physiological relevance of their interaction has been elusive. To assess the effects of the obscurin/titin binding in vivo, we generated a deletion model, Obscn-ΔIg58/59, that carries truncated obscurin lacking the Ig58/Ig59 region that supports binding to both titin and Novex-3. Homozygous Obscn-ΔIg58/59 male mice develop left ventricular (LV) hypertrophy by 6 months, which progresses to LV dilation and severe arrhythmia by 1 year, while female mice present mild arrhythmia. Exertion of pathological and physiological stress in young mice via β-adrenergic stimulation and strenuous exercise, respectively, revealed electrical abnormalities and poorer running ability. Mutations in obscurin and titins, including ones that disrupt their binding, are linked to cardiac and skeletal myopathies. It is thus apparent that the obscurin/titin complex is essential for normal muscle structure and function, and that disruption of their binding is associated with muscle pathogenicity. Our findings using the Obscn-ΔIg58/59 model corroborate this notion.

S11-2

Myocardial overexpression of ANKRD1 affects developmental cardiac remodeling and leads to adult diastolic dysfunction

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Aims: Increased Ankrd1 levels linked to genetic mutations have been correlated to congenital heart disease onset and adult cardiomyopathy occurrence in humans. The link between increased ANKRD1 level and cardiac structural and functional disease onset is not understood. To get insight into this problem, we have generated a ANKRD1 mouse model by overexpressing ANKRD1 in the myocardium.
Methods and Results: We show that ANKRD1 delineates discrete sub-compartments in the developing mouse heart. ANKRD1 transgenic mice present impaired cardiac remodeling, which strongly affects the developing sinoatrial region and leads to sinus venosus defects. Transgenic mice survive to adulthood but develop left atrial enlargement accompanied by severe diastolic dysfunction. Embryonic and neonatal transgenic cardiomyocytes present irregular shape and sarcomeric disorganization, which progresses into sarcomeric loss and mitochondrial damage in adult ventricular but not atrial cardiomyocytes. While isolated embryonic transgenic myofibrils show the same mechanical properties of wild type samples, neonatal transgenic myofibrils present higher passive tension and maximal force compared to wild type. This indicates the presence in ANKRD1 transgenic mice of a faster functional shift towards stiffer and hypercontractile cardiomyocytes, triggered by the increase in workload at birth. At the molecular level, these changes are accompanied by dynamic alterations in titin isoforms ratio. Interestingly, adult wild type and transgenic myofibrils show the same passive tension as transgenic neonatal myofibrils, with adult transgenic myofibrils showing a higher maximal force accompanied at this stage by a marked slowing down of the relaxation phase compatible with the overt diastolic disfunction of adult ANKRD1 transgenic mice.
Conclusions: Our data indicate that genetic mutations leading to increased ANKRD1 levels can lead both to congenital heart disease and adult cardiomyopathy via a common cellular mechanism, with ANKRD1 playing the role of a critical strain sensor-signaling molecule finely modulating cardiomyocyte function during development and postnatal life.

S11-3

Compartmentalization of titin & Novex 3 during sarcomere assembly in regenerating skeletal muscle

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The giant protein titin spans from the Z-disk to the M-line in the sarcomere of striated muscle cells, where it functions as a molecular spring during stretching and relaxation. In this study we used immuno electron microscopy and three-dimensional reconstruction to localize full-length titin and Novex-3 ("tiny titin") at different stages of myofibrillogenesis in regenerating rat soleus muscle after notexin-induced myofibril breakdown. Two days after intoxication with notexin we observed first single thick filaments in the cytosol colocalized with full length titin. In addition, we identified subcellular compartments containing Novex-3 titin as integral elements of emerging Z-bodies. Thick filaments aligned to build first premyofibrils containing titin, myosin and Z-bodies; three days after intoxication we found Z-bodies fusing to Z-disks, forming contracted sarcomeric structures, which later develop into mature myofibrils with I- and A-Band showing the typical striation pattern. Our results support a model in which titin acts as a molecular scaffold for the assembly of Z-discs and thick filaments during skeletal muscle regeneration.

S11-4

Myocardial regeneration: uncommon sense for common problems

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Myocardial regenerative research remains an area of intensive study despite over a decade of frustratingly slow progress and modest clinical efficacy. A fundamental limitation in myocardial regeneration is inherently poor reparative capacity of adult mammalian heart which declines over lifespan. Augmentation of repair requires unnatural solutions to overcome normal adult myocardial biology using Regeneration Associated Cellular Effectors (RACE) to deliver functionally competent therapeutic interventions. The logic and rationale