have demonstrated, in fibers internally equilibrated with 70 mM Cl, bathed in Tyrode (4mM K and 155mM Cl; RP~-90mV)? Short fibers isolated from flexor digitorum brevis (FDB) muscles were impaled with 2 microelectrodes and stimulated with step current pulses (~15% of cells). However, periodic Ca2
þ transients consistent with action potentials (APs) in WT cells. ISO (10nM) increased the frequency of APs in the KO cell lineage, but the paced clock'' mechanism for normal pacemaker activity.

To study the underlying pacing mechanism in the KO, we enzymatically isolated SAN cells from WT and NCX1 KO mice. NCX1 KO SAN cells were ~30% longer than occasionally exhibited regular Ca2
þ transients (~15% of cells). However, periodic Ca2
þ waves were often observed in NCX1 KO cells (but rarely in WT). These waves occurred at about the same rate as the spontaneous transients in WT cells. ISO (10mM) increased the frequency of waves by ~50% in KO cells, similar to the increase in the rate of Ca2
þ transients in WT cells.

In summary, periodic Ca2
þ waves persist in NCX1 KO SAN cells. However, only a minority of these cells generate Ca2
þ transients consistent with action potentials. Therefore, the pacing clock can “tick” in the absence of NCX1, but seems no longer coupled to membrane depolarization.

1074-Plat
Atrial Specific Stretch-Dependent Sub-Cellular Ca2
þ Signaling
University of Maryland, Baltimore, MD, USA.
Atrial myocytes undergo stretch during diastole as do ventricular myocytes. We have recently identified a novel mechanism that links cellular stretch in ventricular myocytes to the tuning of Ca2
þ release from the sarcoplasmic reticulum (SR)(Prosser et al. Science 2011:333:1440-5). This mechanism, “X-ROS signaling”, depends on NOX2 (NADPH oxidase) in the sarcolemmal and transverse tubule (TT) membranes to generate reactive oxygen species (ROS), which appears to oxidize the nearby (nanometers) ryadion receptors (SR, Ca2
þ release channels) and increases their sensitivity to [Ca2
þ ]i. Stretch mediates X-ROS signaling through microtubules, which appear to interact with NOX2 to enable it to generate ROS.
We evaluated the effect of acute and repeated stretch on sub-cellular Ca2
þ sparks in muring atrial myocytes (C57/B6; Figure 1A). Ca2
þ sparks were recorded before and after stretch (8% of cell length). Ca2
þ spark frequency increased during

have demonstrated, in fibers internally equilibrated with 70 mM Cl, bathed in TEA-Cl (145mM), and voltage-clamped at ~20mV, that the majority of the Cl currents (ICl; ~1mA/cm2) are contributed by 9-ACA-sensitive chloride (ClC-1) channels expressed in the transverse tubular system (TTS) membranes. Likewise, in fibers internally equilibrated with 150mM K, bathed with 150mM KMeSO4, and clamped at 0mV, the majority of the K-currents arise from inward rectifier K (IKir; ~1mA/cm2) channels in the TTS. Here we address the question: what are the relative contributions of gCl and gKIR to Rm in isolated fibers bathed in Tyrode (4mM K and 155mM Cl; RP~90mV)? Short fibers isolated from flexor digitorum brevis (FDB) muscles were impaled with 2 microelectrodes and stimulated with step current pulses (~15% of cells). However, periodic Ca2
þ transients consistent with action potentials (APs) in WT cells. ISO (10nM) increased the frequency of APs in the KO cell lineage, but the paced clock” mechanism for normal pacemaker activity.

To study the underlying pacing mechanism in the KO, we enzymatically isolated SAN cells from WT and NCX1 KO mice. NCX1 KO SAN cells were ~30% longer than occasionally exhibited regular Ca2
þ transients (~15% of cells). However, periodic Ca2
þ waves were often observed in NCX1 KO cells (but rarely in WT). These waves occurred at about the same rate as the spontaneous transients in WT cells. ISO (10mM) increased the frequency of waves by ~50% in KO cells, similar to the increase in the rate of Ca2
þ transients in WT cells.

In summary, periodic Ca2
þ waves persist in NCX1 KO SAN cells. However, only a minority of these cells generate Ca2
þ transients consistent with action potentials. Therefore, the pacing clock can “tick” in the absence of NCX1, but seems no longer coupled to membrane depolarization.

1074-Plat
Atrial Specific Stretch-Dependent Sub-Cellular Ca2
þ Signaling
University of Maryland, Baltimore, MD, USA.
Atrial myocytes undergo stretch during diastole as do ventricular myocytes. We have recently identified a novel mechanism that links cellular stretch in ventricular myocytes to the tuning of Ca2
þ release from the sarcoplasmic reticulum (SR)(Prosser et al. Science 2011:333:1440-5). This mechanism, “X-ROS signaling”, depends on NOX2 (NADPH oxidase) in the sarcolemmal and transverse tubule (TT) membranes to generate reactive oxygen species (ROS), which appears to oxidize the nearby (nanometers) ryadion receptors (SR, Ca2
þ release channels) and increases their sensitivity to [Ca2
þ ]i. Stretch mediates X-ROS signaling through microtubules, which appear to interact with NOX2 to enable it to generate ROS.
We evaluated the effect of acute and repeated stretch on sub-cellular Ca2
þ sparks in muring atrial myocytes (C57/B6; Figure 1A). Ca2
þ sparks were recorded before and after stretch (8% of cell length). Ca2
þ spark frequency increased during