Supplemental Material

Figure Legends

Figure 1S Durations of AP and CaT are tightly correlated during paced rhythm in LQT2

A: Simultaneous Ca_i and V_m tracings from 2 pixels (**a, b**) indicated in the left panel. The basal site has longer APD and 2 secondary CaT elevations, while the more apical site has only one secondary CaT rise. The APD and CaTD are very similar at each site. The maps of $CaTD_{80}$ APD_{80} (**B**) and the corresponding scatterplot (**C**) demonstrate excellent correlation between the two variables.

Figure 2S: Propagated EADs arise in regions of steep voltage gradients

A Ca_i and V_m tracings during EAD from 3 pixels (**a-c**) indicated in the Ca_i and V_m maps. As seen in the map of activation time of the EAD propagation (**AT; right panel**), the earliest V_m upstroke occurs at site **b**, which exhibits steep V_m gradient immediately before the ectopic beat in the V_m map. **B** An example of voltage gradient map (**top panel**; warm colors indicate steep gradient) and activation map (**bottom panel**) of an EAD in another experiment. The dotted lines indicate the EAD focus. The site has a steep voltage gradient before EAD onset. **C** Relative voltage gradients at the site of EAD are plotted for 6 different experiments.

Figure 3S: Low [K201] suppress LQT2-related arrhythmias

A Simultaneous optical recordings of voltage (V_m in blue) and intracellular Ca^{2+} (Ca_i in red) during an arrhythmia elicited by perfusion with dofetilide (0.5 μM) and 50% reduction of KCl and $MgCl_2$.
B Perfusion with 0.5 μM K201 for 5 min suppressed the arrhythmia

Figure 4S Systolic and diastolic Ca_i elevations from the same region of epicardium

Ca_i tracings from 2 pixels (**top**) initiated by electrical pacing during low-resolution optical mapping. Site **a**, but not site **b**, exhibits both systolic and diastolic Ca_i rise. Systolic and diastolic Ca_i maps during paced rhythm 1 and 3 minutes after LQT2 show the overlap between areas of systolic and diastolic Ca^{2+} release (**middle and bottom**). The pixels **a** and **b** from the top panel are indicated in the bottom maps. Note the different color scale in the systolic and diastolic maps.

Figure 5S: During propagated DADs, Ca_i rise precedes V_m rise at the ectopic focus. The top panel shows simultaneous Ca_i and V_m tracing from a low-resolution experiment during ventricular arrhythmia with 3 ectopic beats marked by arrowheads. The zero-phase maps displayed below for each of the beats for both Ca_i and V_m indicate the timing of signal upstroke in each pixel. Note that Ca_i rise precedes V_m rise at the earliest site.

Figure 6S: Intracellular Ca^{2+} ripples

Small-amplitude, spatially poorly synchronized Ca^{2+} waves were occasionally observed on top of prolonged CaT plateau in high-resolution experiments. A large number of these "ripples" are observed to propagate over a distance of a few μm in each myocyte in the field of view. See also **Movie F**.

Movie Files:

Movie A: Subcellular Ca^{2+} dynamics during 2 Hz pacing at baseline. The field of view is a square of 150x150 μm size in this and other high-resolution movies. The signal intensity is normalized in each pixel. At the bottom, Ca^{2+} signal from a single pixel is shown. The position of the moving cursor indicates the current movie frame. The CaT is monophasic and uniform across the field of view.

Movie B: High-resolution movie obtained during slow (1.2 s) pacing in LQT2. Secondary systolic Ca^{2+} oscillation is apparent, which is quite homogenous across the field of view.

Movie C: Diastolic Ca^{2+} waves are seen in this high-resolution movie recorded during slow pacing in LQT2. The secondary systolic Ca^{2+} upstroke takes the form of gradually up-sloping CaT plateau.

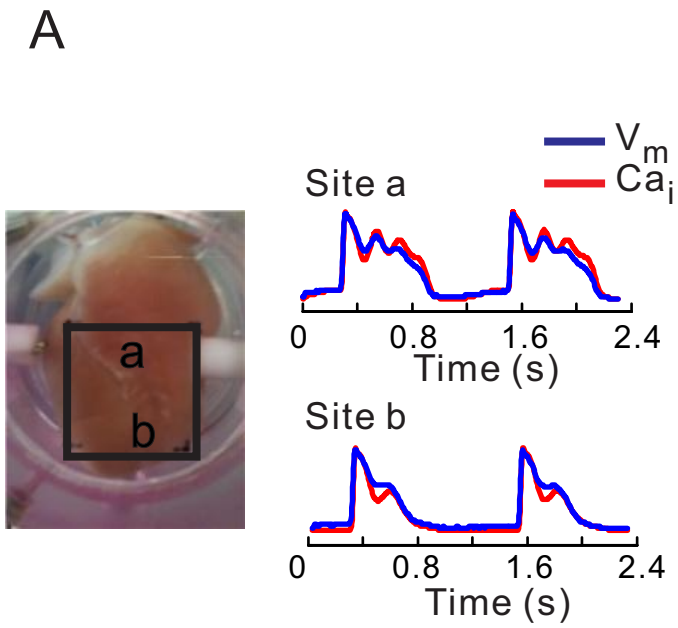
Movie D: During runs of VT in LQT2, multiple CaT upstrokes occur from a depressed CaT plateau in this high-resolution movie. They do not correspond to Ca^{2+} waves; rather, they are cell-synchronous.

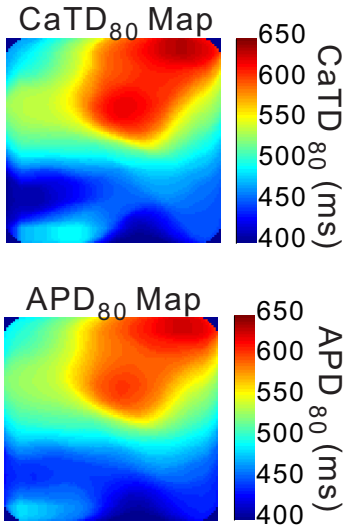
Movie E: An example of simultaneous Ca^{2+} (left) and V_m (right) a low-resolution movie recorded during slow pacing in LQT2. The field of view is a square of 15 mm size. The rapid upstroke is caused by RV pacing. The AP upstroke precedes the CaT upstroke slightly. Two systolic CaT oscillations follow the initial upstroke in this example. Note that SH of CaT exceeds SH of AP, although the signals are well correlated.

Movie F: In this high-resolution movie recorded in LQT2, the initial CaT upstroke is followed by a long, rectangular plateau, on which small-amplitude oscillations, or “ripples” are superimposed. The ripples are not well synchronized within the cell, although they do not propagate across the length of the cell in the same way as diastolic waves.

Movie G: High-resolution movie showing chaotic subcellular Ca^{2+} dynamics in LQT2. Multiple Ca^{2+} waves propagate through the cells, collide and annihilate, and new waves arise continuously. Low baseline Ca^{2+} level is never achieved across the whole cell.

Figure 1S


A
a
b
Site a
Site b
V_m
Ca_i
0
0.8
1.6
2.4
Time (s)


B
$CaTD_{80}$ Map
APD_{80} Map
650
600
550
500
450
400
$CaTD_{80}$ (ms)
APD_{80} (ms)

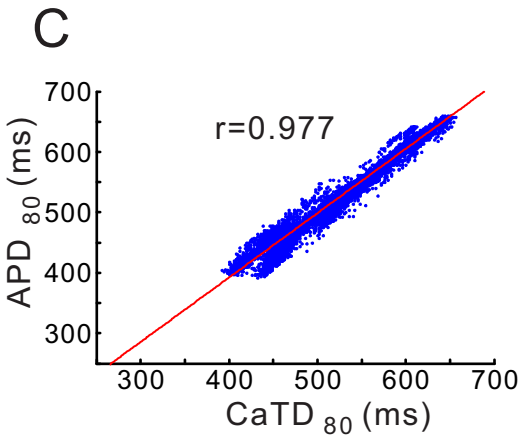

C
r=0.977
700
600
500
400
300
APD_{80} (ms)
$CaTD_{80}$ (ms)

Figure 2S

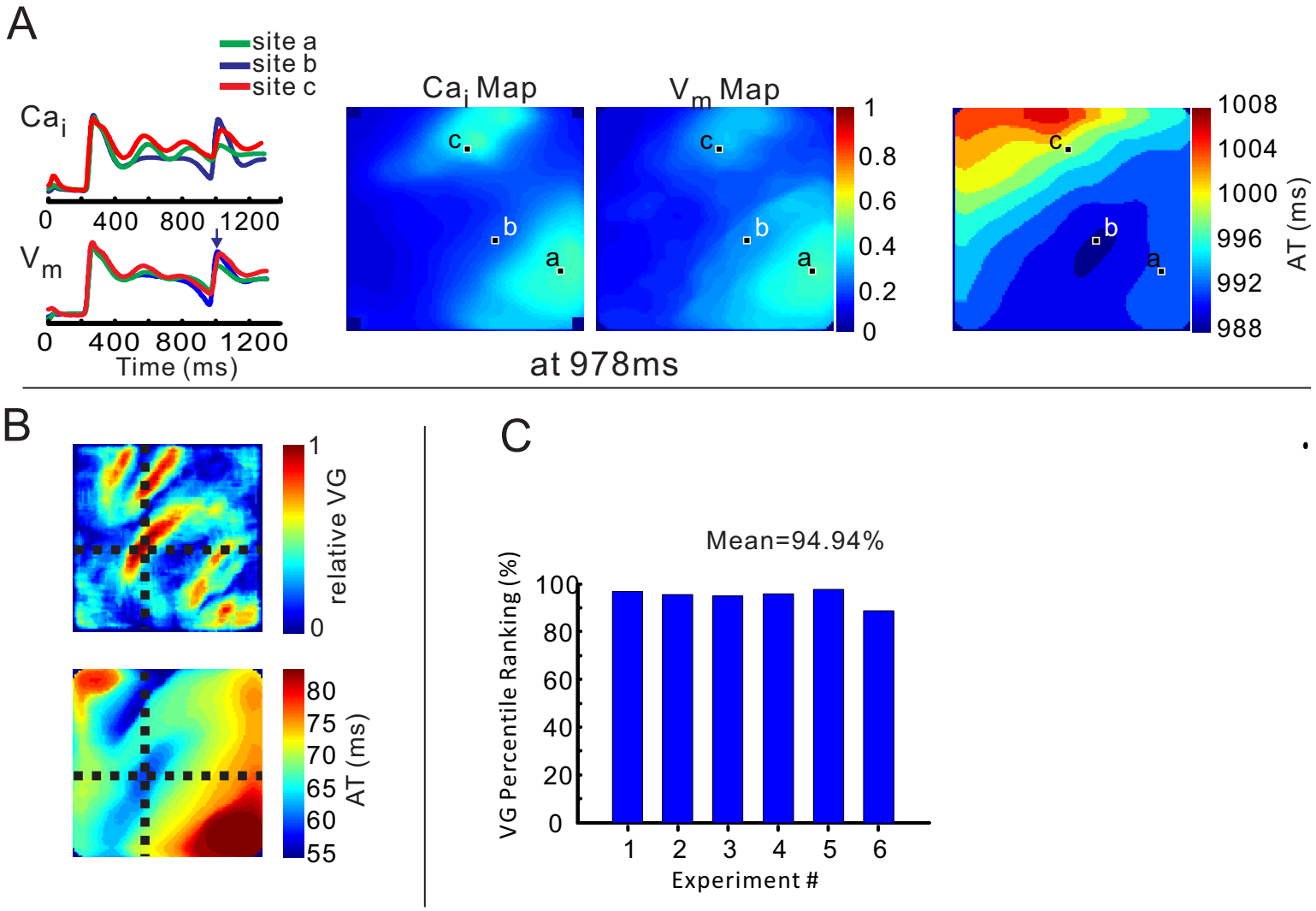

A
site a
site b
site c
Ca_i
0 400 800 1200
V_m
0 400 800 1200
Time (ms)
Ca_i Map
V_m Map
c
b
a
1
0.8
0.6
0.4
0.2
0
1008
1004
1000
996
992
988
AT (ms)
at 978ms
B
1
relative VG
0
80
75
70
65
60
55
AT (ms)
C
Mean=94.94%
VG Percentile Ranking (%)
100
80
60
40
20
0
1 2 3 4 5 6
Experiment #

Figure 3S

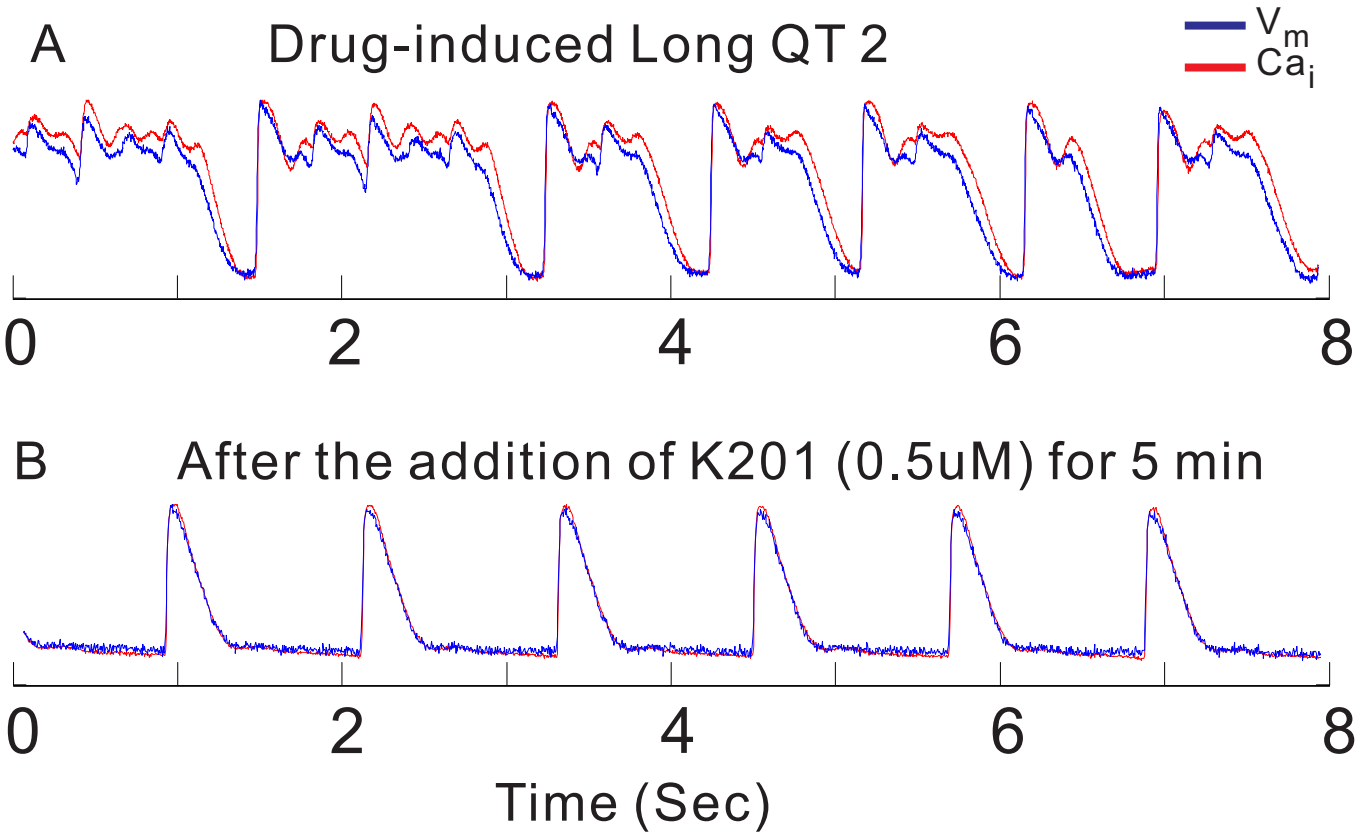

A
Drug-induced Long QT 2
V_m
Ca_i
0
2
4
6
8
B
After the addition of K201 (0.5uM) for 5 min
0
2
4
6
8
Time (Sec)

Figure 4S

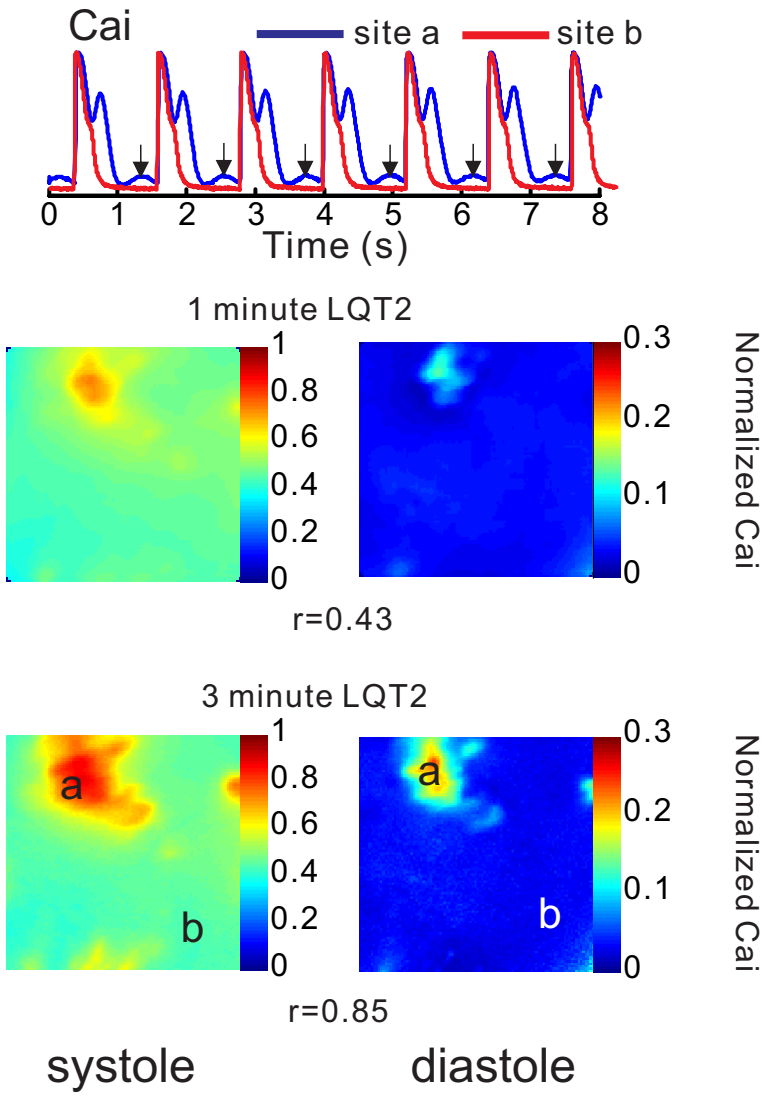

Cai
site a
site b
0 1 2 3 4 5 6 7 8
Time (s)
1 minute LQT2
1
0.8
0.6
0.4
0.2
0
0.3
0.2
0.1
0
Normalized Cai
r=0.43
3 minute LQT2
a
b
a
b
r=0.85
systole
diastole

Figure 5S

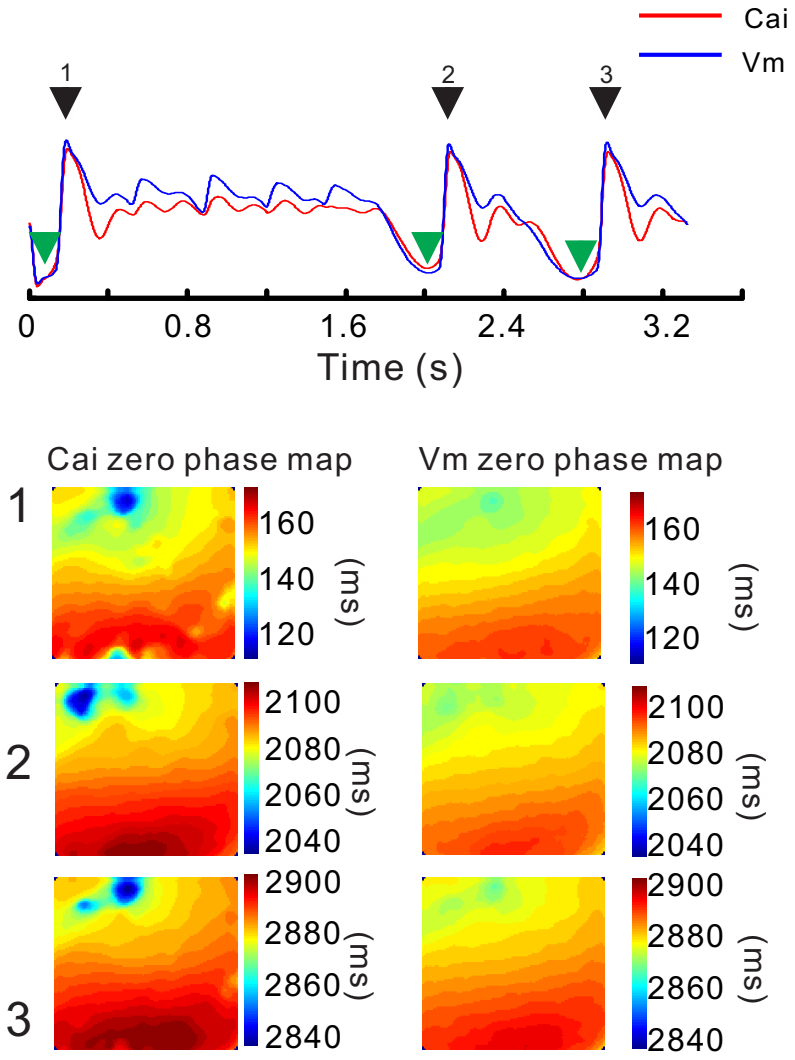

Cai
Vm
1
2
3
0
0.8
1.6
2.4
3.2
Time (s)
Cai zero phase map
Vm zero phase map
1
2
3
160
140
120
(ms)
2100
2080
2060
2040
2900
2880
2860
2840

Figure 6S