Supporting Online Material

Cell culture and sample preparation

S2R+ cells were obtained from the Drosophila Genome Resource Center and cultured at 25^{o}C and no CO_2 in culturing medium consisting of Schneiders' Drosophila Medium (Gibco #11720) with 10 mM Pen-Strep (Gibco #15070-122) and 10% fetal bovine serum (Sigma #F2442). To transfect with dsRNA, 6.25×10^6 S2R+ cells in 0.4 ml of Schneiders' Drosophila Medium (no serum) were incubated with 15 micrograms of dsRNA and shaken gently for 1 hour at 25°C. Then the cell mixture was plated into a well of a 6-well tissue culture plate, and an additional 1.4 ml of culturing medium with 10% FBS was added to each well. The cells were placed at 25°C for 96 hours before performing assays. To transfect S2R+ cells with cDNA, Fugene HD (Roche) was used according to the manufacturer's protocol.

Preparation of dsRNA

DsRNA was generated by carrying out T7 PCR reactions on a Drosophila genomic library followed by in vitro transcription of the PCR products using the MEGAscript® T7 Kit (Ambion) following the manufacturer guidelines. The dsRNA was cleaned by using an RNeasy filter kit (Qiagen). Supp. Table 3 shows the primer sequences used to make dsRNA.

Measurement of intracellular free calcium concentration

Drosophila S2R+ cells were plated in 96-well plates at 15-20 x 10^5 cells/well. All imaging was carried out on an Axon ImageXpress imaging system (Molecular Devices). For cytosolic Ca2+ measurements, the cells were loaded for 30 minutes at 25°C with 1 μM

FURA2-AM and 0.01% Pluronic (Molecular Probes) in buffer consisting of 5 mM KCl, 125 mM NaCl, 20 mM Hepes, 1.5 mM $MgCl_2$, 1.5 mM $CaCl_2$, and 2.5 mM probenecid (pH 7.4). The cells were then washed once with this buffer and then subsequently imaged at 10X magnification. FURA2 timecourses of more than 200 cells were measured for each cytosolic calcium condition shown in Fig. 3.

To carry out ER calcium measurements, the D1ER FRET probe (1) was cloned into a Drosophila expression vector (pAW obtained from the Drosophila Genomic Resource Center). To make a more sensitive FRET probe, we replaced the CFP in the probe with a much brighter, mono-exponentially decaying variant, mTurquoise (2). Drosophila S2R+ cells were transfected with the pAW- t1ER probe, and two days later when the construct was well-expressed, CFP, YFP, and FRET images were collected at 10X magnification. The images were analyzed using custom-written MATLAB scripts: the YFP images were used to create a mask to define which cells would be analyzed. The FRET ratio (R) for each cell was calculated by ratioing the background-corrected intensities in the FRET and CFP channels, and the well medians of these single-cell FRET ratios were normalized to control wells within each replicate set. The FRET ratios were converted to Ca^{2+} concentration using methodology outlined in (3) and in Supp. Fig. 6. ER Ca2+ levels were also measured indirectly using FURA2-AM, and the methodology and data are described in the Supplementary Materials.

Sample Preparation for Mass Spectrometry Analysis

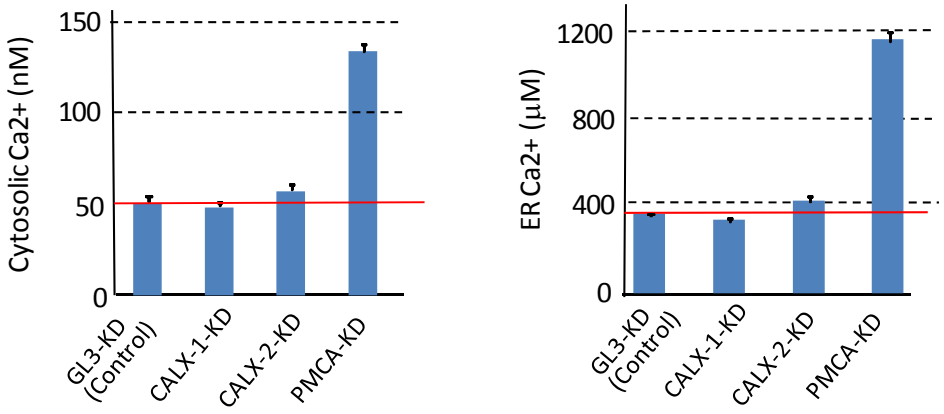
In order to be able to measure small changes in protein concentration between different samples, great pains were taken to make the sample preps as consistent and reproducible as possible. To prevent unwanted, early cleavage of membrane proteins, no trypsin was

used to remove the S2R+ cells from the 6-well dishes. Instead gentle trituration was used. For each sample, 15×10^6 cells were pelleted by a low-speed spin, washed ice-cold PBS, frozen in liquid nitrogen, and resuspended in 500µl of ice-cold lysis buffer (10mM Hepes pH 7.9; 0.5mM MgCl2, 10 mM KCl, 1mM DTT, 0.1% digitonin, 1mM PMSF, and a complete protease inhibitor cocktail (Roche, Mannheim, Germany) at 4°C for 10 min. The cells were broken open by triturating 5 times through a 30-gauge needle and syringe, and the cell lysate was centrifuged at 3,000g for 10 min to remove the nuclear debris. The supernatant was centrifuged at 39,000 RPM (Beckman rotor TLA 120.2) for 1 hour. The pellet (membrane fraction) was resuspended in 500ul of 25mM $NaCO_3$ (pH 11) by pipetting thoroughly. The suspension was left on ice for 60 minutes and then pipetted 25 times with a 200µl pipet tip to further shear and dissolve the membranes. After the addition of 500µl of ammonium bicarbonate (50mM, pH 8), the suspension was centrifuged at 39,000 RPM (Beckman rotor TLA 120.2) for 30 minutes. The pellet was resuspended in 250 µl of ammonium bicarbonate (100mM, pH 8). 60 ul of Rapigest (Waters, 0.1% solution) was used to dissolve the pellet, after which a BCA kit (Pierce, Catalog #23225) was used to measure the concentration of the proteins in each sample.

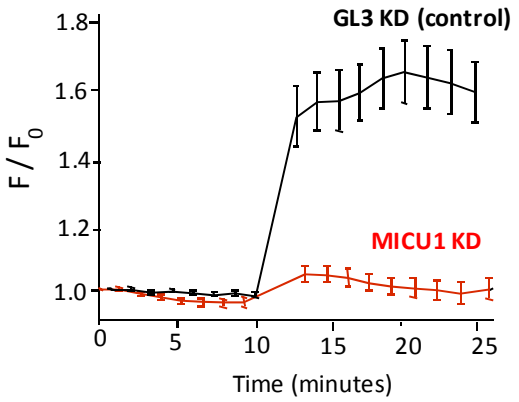
The BCA readings were used to normalize the concentration of protein in and the volumes of each sample at this step in order to make the subsequent addition of heavy peptides, trypsin digestion, and peptide cleanup steps more consistent between samples. The heavy peptide mix was prepared as per Supp. Table 2, and an equal volume of this mix was added to each sample. The solution was then placed in a sonicator for 10 minutes.

The disulfide bonds of the proteins and AQUA peptides were reduced by incubation with tris(2-carboxyethyl)phosphine (TCEP) at a final concentration of 5 mM

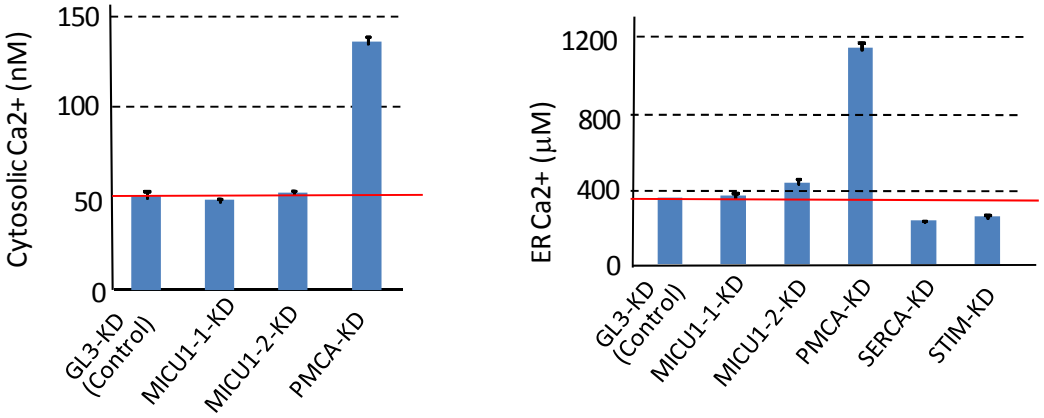
for 30 minutes at 25 °C. The produced free thiols were alkylated with 15 mM iodoacetamide (Sigma) at room temperature for 30 minutes in the dark. The proteins were digested overnight at 37°C with sequencing-grade modified trypsin (Promega, Cat #V5113) added at a ratio of 1 µg per 100µg of protein. The digestion was terminated and the Rapigest degraded by acidifying the samples to pH 2-3 with HCl or formic acid and then incubating for 30 minutes at 37°C. The samples were centrifuged for 10 minutes at 9,300g to remove the Rapigest, and the supernatant, which contained the peptides, was desalted on a C18 Sep-Pak cartridge (Waters, Milford, MA, USA) and dried on a Speedvac. The peptides were resolubilized in 0.2% Acetonitrile with 0.1% formic acid. The concentration of peptides in each sample was readout at 230 nm using a Nanodrop and adjusted to be 0.6 for each sample. Having the same peptide concentration for each sample allowed for more reproducible chromatography, tighter acquisition windows, and thus better signal-to-noise.



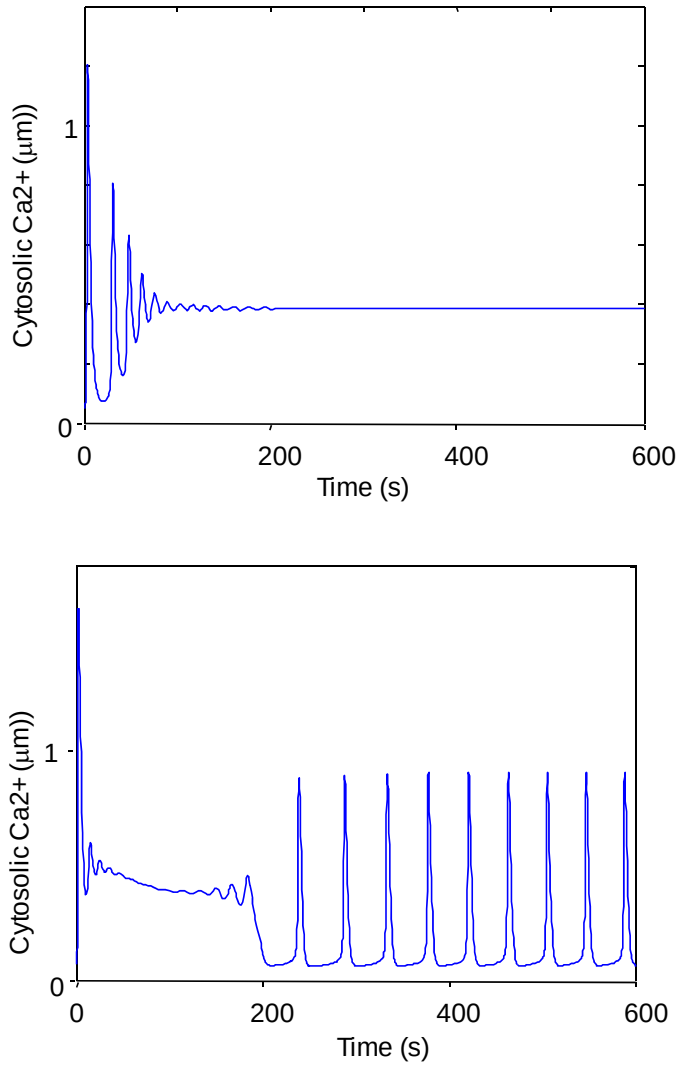
Supp. Figure 1: In comparison to knockdown of the plasma membrane pump PMCA, knockdown of the plasma membrane Na2+/Ca2+ exchanger, CALX, showed no significant effect on basal cytosolic or ER calcium levels in Drosophila S2R+ cells. Experiments were carried out using dsRNA targeted to two different coding regions of CALX (labeled CALX-1 and CALX-2). dsRNA targeting GL3 was used as a control. As described in the Material and Methods section, cytosolic calcium was measured using FURA2-AM, and the ER calcium was measured using the pAW-t1ER ER-targeted FRET probe. Approximately 100 cells were measured for each dsRNA knockdown condition, and the error bars show the standard error.



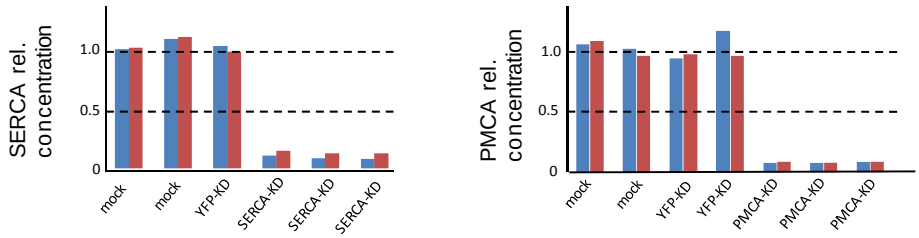
Supp. Figure 2: MICU1 knockdown using dsRNA results in suppression of mitochondrial calcium signals in Drosophila S2R+ cells as measured with a mitochondrially-targeted FRET probe (pAW-4mtD3cpv). To obtain this timecourse, S2R+ cells expressing pAW-4mtD3cpv were placed in extracellular buffer containing thapsagargin (1uM) and EGTA (3mM), final concentrations, for 20 minutes to deplete the ER of calcium, and images (CFP, YFP, FRET) were taken for seven timepoints. Then high calcium (10mM) and thapsagargin (1uM), final concentrations, were added to the cells, and images were taken for another 20 minutes. FRET (F) was calculated for cells by ratioing the intensities in the FRET and the CFP donor channel. F_0 is the baseline FRET calculated as the average FRET value before adding stimulus. To make the pAW-4mtD3cpv probe, we obtained the 4mtD3cpv construct from Dr. Roger Tsien's lab and cloned it into a Drosophila expression vector with a constitutive actin promoter (pAW, obtained from the Drosophila Genome Resource Center). Approximately 50 cells were measured for each dsRNA knockdown condition, and the error bars show standard error. Primers to make the dsRNA are shown in Supp. Table 2 (MICU1-1).



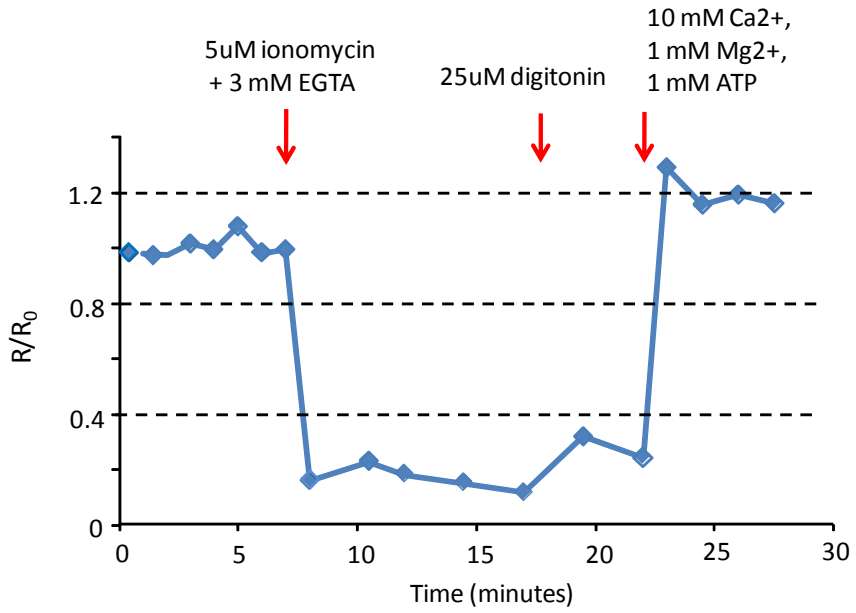
Supp. Figure 3: Knockdown of MICU1 showed no significant effect on basal cytosolic or ER calcium levels in Drosophila S2R+ cells. Experiments were carried out using dsRNA targeted to two different coding regions of MICU1 (labeled MICU1-1 and MICU1-2) . Calcium levels for PMCA, SERCA, and STIM knockdown are shown for comparison. dsRNA targeting GL3 was used as a control. As described in the Material and Methods section, cytosolic calcium was measured using FURA2-AM, and the ER calcium was measured using the pAW-t1ER ER-targeted FRET probe. Approximately 100 cells were measured for each dsRNA knockdown condition, and the error bars show the standard error.



Supp. Figure 4: Sample model output showing complex signaling patterns. The output of the top plot would be categorized as being a "Ca2+ plateau" since the oscillations decay into a stable plateau. The output in the bottom plot would be categorized as "oscillatory" since the plateau phase ends in oscillations.



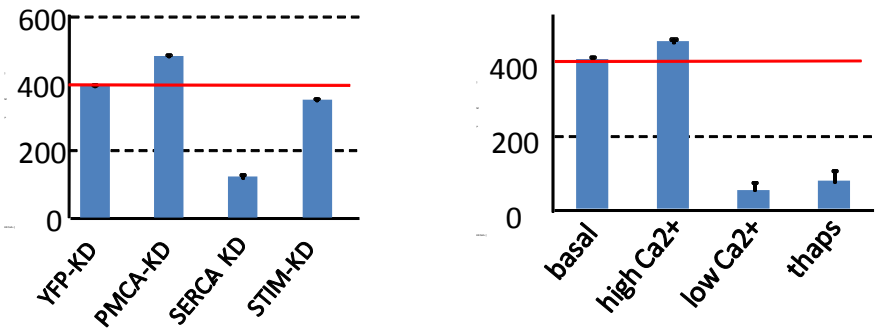
Supp. Figure 5: Knockdown of individual proteins can be measured using SRM mass spectrometry, as shown by use of SERCA and PMCA dsRNA.



Supp. Figure 6: Example of a calibration timecourse for a cell transfected with the pAW-t1ER ER-targeted FRET probe. For each experiment, Drosophila S2R+ cells were transfected with the pAW-t1ER probe and plated into 96-well wells (approx. 100,000 cells per well). The FRET ratio (R) for each cell was calculated by ratioing the background-corrected intensities in the FRET and CFP channels. FRET ratios were converted to Ca^{2+} concentration using the methodology outlined in (3). Cells were imaged for several frames at basal conditions to obtain R_0, the average basal ER calcium concentration. To obtain the minimal possible ER calcium level for each condition (R_{min}), 3 mM EGTA plus 5 uM ionomycin were added to each well, and the cells were subsequently imaged for 10 minutes or until the FRET ratio had reached a stable steady-state. To obtain an R_{max}, cells were treated with 25 uM digitonin and left for 5 minutes in order to allow enough time for the plasma membrane to be permeabilized. Then 10 mM calcium, 1mM ATP, and 1mM Mg^{2+} was added to obtain an immediate rise in ER

Ca^{2+} to its maximal value. To obtain Ca^{2+} concentration for each value of R, the R_{min} and R_{max} were used together with the relevant calibration equation and *in situ* parameters listed in (3).

For the Drosophila cells, the Rmax was found to be nearly the same as the signal measured when we used RNAi to knock down PMCA expression (which raises cytosolic and ER Ca2+), suggesting that we do not have the dynamic range to measure the highest increases in ER Ca2+. Using the calibration procedure outlined in (3), the predicted basal ER Ca2+ level in Drosophila S2R+ cells is 350 μM. This is an approximate measure due to the difficulty in measuring Rmax.



Supp. Figure 7: ER Ca2+ levels were also derived indirectly by measuring the releasable Ca2+ pool by ionomycin addition and monitoring the amplitude of the cytosolic Ca2+ response using FURA2-AM. Timecourses of more than 200 cells were measured for each condition, and the error bars show standard error.

References

1. **Palmer AE & Tsien RY (2006) Measuring calcium signaling using genetically targetable fluorescent indicators. *Nat Protoc* 1(3):1057-1065.**
2. **Goedhart J, *et al.* (2010) Bright cyan fluorescent protein variants identified by fluorescence lifetime screening. *Nat Methods* 7(2):137-139.**
3. **Rudolf R, Magalhaes PJ, & Pozzan T (2006) Direct in vivo monitoring of sarcoplasmic reticulum Ca2+ and cytosolic cAMP dynamics in mouse skeletal muscle. *J Cell Biol* 173(2):187-193.**

Computational Models (with and without adaptive feedback)

The rate of change of cytosolic calcium level is given by:

$$\frac{dCa_I^{2+}}{dt} = J_1 - J_2 + J_3 - J_4$$

where J_1 is the flux of calcium into the cell through the plasma membrane:

$$J_1 = STIM * \left(PMleak + \frac{k_{STIM}^8}{(Ca_S^{2+})^8 + k_{STIM}^8} \right) - PMCA * \frac{(Ca_I^{2+})^2}{(Ca_I^{2+})^2 + (k_{PMCA})^2}$$

J_2 is the flux of calcium into the ER:

$$J_2 = SERCA * \frac{(Ca_I^{2+})^2}{(Ca_I^{2+})^2 + (k_{SERCA})^2}$$

J_3 is the flux of calcium into the cell from the ER (through InsP3R, Ca2+-regulated Ca2+ channel, and leak):

$$J_3 = (1 - DirTransf) * Ca_S^{2+} * \left((1 - g) * InsP3R * \frac{(InsP3)^2}{(InsP3)^2 + (InsP3R)^2} * \frac{(Ca_I^{2+})^2}{(Ca_I^{2+})^2 + (k_{InsP3Rca})^2} + ERleak \right)$$

and J_4 is the flux of calcium into the mitochondria:

$$J_4 = UniPort * \frac{(Ca_I^{2+})^4}{(Ca_I^{2+})^4 + (k_{UniPort})^4} - MitNaCaEx * \left(\frac{Ca_M^{2+}}{Ca_M^{2+} + 0.01} \right)$$

The production of InsP3 depends on the catalytic activity of PLC which depends both on the degree of receptor stimulation (R) and on the Ca2+ level:

$$\frac{dInsP3}{dt} = R * Ca_I^{2+} - k_{InsP3deg} * InsP3$$

g represents the inactivation of the InsP3R by cytosolic calcium:

$$\frac{dg}{dt} = InsP3R_{inhibition} * \frac{(Ca_I^{2+})^4}{(k_g)^4 + (k_{InsP3R})^4} * (1 - g) - InsP3R_{recovery} * g$$

The rate of change of ER calcium is given by:

$$\frac{dCa_S^{2+}}{dt} = SERCA * \frac{(Ca_I^{2+})^2}{(Ca_I^{2+})^2 + (k_{SERCA})^2} - 1$$

The rate of change of mitochondrial calcium is given by:

$$\frac{dCa_M^{2+}}{dt} = M_1 - M_2 + M_3$$

where M_1 is the flux of calcium into the mitochondria through the uniporter:

$$M_1 = UniPort * \frac{(Ca_I^{2+})^4}{(Ca_I^{2+})^4 + (k_{UniPort})^4}$$

M_2 is the flux of calcium out of the mitochondria through the mitochondrial Na2+/Ca2+ exchanger:

$$M_2 = MitNaCaEx * \left(\frac{Ca_M^{2+}}{Ca_M^{2+} + 0.01}\right)$$

M_3 is the flux of calcium directly from the ER into the mitochondria:

$$M_3 = DirTransf * Ca_S^{2+} * \left(ERleak + (1 - g) * InsP3R * \frac{(InsP3)^2}{(InsP3)^2 + (InsP3R)^2} M_1 * \frac{(Ca_I^{2+})^2}{(Ca_I^{2+})^2 + (k_{InsP3R})^2}\right)$$

For Model 2 (with adaptive feedback), the following equations were added:

$$\frac{dPMCA}{dt} = PMCA_0 * cr * \left(\frac{(Ca_I^{2+})^4}{(Ca_I^{2+})^4 + (cr-1) * (0.05)^4}\right) - ProtDeg * PMCA$$

$$\frac{dSERCA}{dt} = SERCA_0 * \frac{1}{cr} * \left(\frac{(Ca_S^{2+})^4 + (cr-1) * 2^4}{(Ca_S^{2+})^4}\right) - ProtDeg * SERCA$$

$$\frac{dSTIM}{dt} = STIM_0 * \frac{1}{(cr * cr)} * \left(\frac{(Ca_S^{2+})^2 + (cr-1) * 2^2}{(Ca_S^{2+})^2}\right)$$
$$* \left(\frac{(Ca_I^{2+})^2 + (cr-1) * (0.05)^2}{(Ca_I^{2+})^2}\right) - ProtDeg * STIM$$

where $PMCA_0$, $SERCA_0$, *and* $STIM_0$ are setpoints of unregulated equilibria, and cr = 50 represents a regulatory ceiling.

The fixed parameters used in the models were: InsP3R0=3.0 (rel unit); SERCA0=0.266; InsP3degradation=2.0/sec; InsP3Rinhibition=5.0/sec; InsP3Rrecovery=0.018/sec; kSERCA=0.175 microM; ERleak=0.01/sec; stimulation R=0 to 4; kInsP3R=0.175 micoM; PMleak=0.0346/sec; STIM0=0.02; kInsP3Rca=0.13 microM; kSTIM=1.0; kPMCA=0.2; $PMCA_0$=0.013; kG=1.0; DirTransf=0.03; MitNaCaEx=0.0050; UniPort=0.03; kUniP=0.6.

For Model 2 (with adaptive feedback), the following parameter was added to establish the adaptive feedback loops: ProtDeg=0.00003/sec (protein turnover, same for STIM, PMCA and SERCA).

Matlab Simbiology was used to program and run the model simulations. The models presented in this manuscript, without and with adaptive feedback, will be uploaded to the EMBL-EBI BioModels repository (http://www.ebi.ac.uk/biomodels-main/).

Supp. Table 1: List of measured peptides and transitions

m/z Q1	m/z Q3	CE [V]	Sequence	fragment	light,heavy	Peptide Name	Gene Symb	CG#	NP#	Gene Description
472.2640	545.3042	19.4	SPITSSVPR	y5	light	ORAI-2	olf186-F	CG11430	NP_611273.1	CG11430 gene product from transcript CG11430-RB
472.2640	646.3519	19.4	SPITSSVPR	y6	light	ORAI-2	olf186-F	CG11430	NP_611273.1	CG11430 gene product from transcript CG11430-RB
472.2640	759.4359	19.4	SPITSSVPR	y7	light	ORAI-2	olf186-F	CG11430	NP_611273.1	CG11430 gene product from transcript CG11430-RB
477.2640	555.3042	19.4	SPITSSVPR	y5	heavy	ORAI-2	olf186-F	CG11430	NP_611273.1	CG11430 gene product from transcript CG11430-RB
477.2640	656.3519	19.4	SPITSSVPR	y6	heavy	ORAI-2	olf186-F	CG11430	NP_611273.1	CG11430 gene product from transcript CG11430-RB
477.2640	769.4359	19.4	SPITSSVPR	y7	heavy	ORAI-2	olf186-F	CG11430	NP_611273.1	CG11430 gene product from transcript CG11430-RB
512.2771	531.3249	20.7	YEVTVSGIR	y5	light	ORAI-1	olf186-F	CG16944	NP_727448.1 or NP_511109.6	CG11430 gene product from transcript CG11430-RB
512.2771	632.3726	20.7	YEVTVSGIR	y6	light	ORAI-1	olf186-F	CG16944	NP_727448.1 or NP_511109.6	CG11430 gene product from transcript CG11430-RB
512.2771	731.4410	20.7	YEVTVSGIR	y7	light	ORAI-1	olf186-F	CG16944	NP_727448.1 or NP_511109.6	CG11430 gene product from transcript CG11430-RB
515.9254	545.2790	26	QIEVQSTQSGNAQR	y5	light	PRES-1	Psn	CG18803	NP_001137988.1	Presenilin
515.9254	632.3111	26	QIEVQSTQSGNAQR	y6	light	PRES-1	Psn	CG18803	NP_001137988.1	Presenilin
515.9254	760.3696	26	QIEVQSTQSGNAQR	y7	light	PRES-1	Psn	CG18803	NP_001137988.1	Presenilin
517.2771	541.3249	20.7	YEVTVSGIR	y5	heavy	ORAI-1	olf186-F	CG11434	NP_611273.1	CG11430 gene product from transcript CG11430-RB
517.2771	642.3726	20.7	YEVTVSGIR	y6	heavy	ORAI-1	olf186-F	CG11434	NP_611273.1	CG11430 gene product from transcript CG11430-RB
517.2771	741.4410	20.7	YEVTVSGIR	y7	heavy	ORAI-1	olf186-F	CG11434	NP_611273.1	CG11430 gene product from transcript CG11430-RB
519.2587	555.2790	26	QIEVQSTQSGNAQR	y5	heavy	PRES-1	Psn	CG18803	NP_001137988.1	Presenilin
519.2587	642.3111	26	QIEVQSTQSGNAQR	y6	heavy	PRES-1	Psn	CG18803	NP_001137988.1	Presenilin
519.2587	770.3696	26	QIEVQSTQSGNAQR	y7	heavy	PRES-1	Psn	CG18803	NP_001137988.1	Presenilin
570.8004	635.3763	22.7	SAEAFLEFVK	y5	light	ERP44-1	CG9911	CG9911	NP_573111.1 or NP_727949.1	CG9911 gene product from transcript CG9911-RD
570.8004	782.4447	22.7	SAEAFLEFVK	y6	light	ERP44-1	CG9911	CG9911	NP_573111.1 or NP_727949.1	CG9911 gene product from transcript CG9911-RD
570.8004	853.4818	22.7	SAEAFLEFVK	y7	light	ERP44-1	CG9911	CG9911	NP_573111.1 or NP_727949.1	CG9911 gene product from transcript CG9911-RD
570.8004	982.5244	22.7	SAEAFLEFVK	y8	light	ERP44-1	CG9911	CG9911	NP_573111.1 or NP_727949.1	CG9911 gene product from transcript CG9911-RD
574.8004	643.3763	22.7	SAEAFLEFVK	y5	heavy	ERP44-1	CG9911	CG9911	NP_573111.1 or NP_727949.1	CG9911 gene product from transcript CG9911-RD
574.8004	790.4447	22.7	SAEAFLEFVK	y6	heavy	ERP44-1	CG9911	CG9911	NP_573111.1 or NP_727949.1	CG9911 gene product from transcript CG9911-RD
574.8004	861.4818	22.7	SAEAFLEFVK	y7	heavy	ERP44-1	CG9911	CG9911	NP_573111.1 or NP_727949.1	CG9911 gene product from transcript CG9911-RD
574.8004	990.5244	22.7	SAEAFLEFVK	y8	heavy	ERP44-1	CG9911	CG9911	NP_573111.1 or NP_727949.1	CG9911 gene product from transcript CG9911-RD
597.3117	562.3195	23.6	DFAAGGISAAVSK	y6	light	MITCAR-1	sesB	CG16944	NP_727448.1 or NP_511109.3	stress-sensitive B
597.3117	789.4465	23.6	DFAAGGISAAVSK	y9	light	MITCAR-1	sesB	CG16944	NP_727448.1 or NP_511109.2	stress-sensitive B
597.3117	860.4836	23.6	DFAAGGISAAVSK	y10	light	MITCAR-1	sesB	CG16944	NP_727448.1 or NP_511109.1	stress-sensitive B
597.3117	931.5207	23.6	DFAAGGISAAVSK	y11	light	MITCAR-1	sesB	CG16944	NP_727448.1 or NP_511109.1	stress-sensitive B
601.3117	570.3195	23.6	DFAAGGISAAVSK	y6	heavy	MITCAR-1	sesB	CG16944	NP_727448.1 or NP_511109.6	stress-sensitive B
601.3117	797.4465	23.6	DFAAGGISAAVSK	y9	heavy	MITCAR-1	sesB	CG16944	NP_727448.1 or NP_511109.5	stress-sensitive B
601.3117	868.4836	23.6	DFAAGGISAAVSK	y10	heavy	MITCAR-1	sesB	CG16944	NP_727448.1 or NP_511109.4	stress-sensitive B
601.3117	939.5207	23.6	DFAAGGISAAVSK	y11	heavy	MITCAR-1	sesB	CG16944	NP_727448.1 or NP_511109.4	stress-sensitive B
625.3122	646.3519	24.6	DGNGFISAAELR	y6	light	CAM-3	Cam	CG8472	NP_523710.1	Calmodulin
625.3122	759.4359	24.6	DGNGFISAAELR	y7	light	CAM-3	Cam	CG8472	NP_523710.1	Calmodulin
630.3122	656.3519	24.6	DGNGFISAAELR	y6	heavy	CAM-3	Cam	CG8472	NP_523710.1	Calmodulin
630.3122	769.4359	24.6	DGNGFISAAELR	y7	heavy	CAM-3	Cam	CG8472	NP_523710.1	Calmodulin
665.2853	509.2024	32.6	HLDACETMGNATAICSDK	y4	light	PMCA-3	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
665.2853	622.2865	32.6	HLDACETMGNATAICSDK	y5	light	PMCA-3	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
665.2853	693.3236	32.6	HLDACETMGNATAICSDK	y6	light	PMCA-3	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
665.2853	794.3713	32.6	HLDACETMGNATAICSDK	y7	light	PMCA-3	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
667.9519	517.2024	32.6	HLDACETMGNATAICSDK	y4	heavy	PMCA-3	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
667.9519	630.2865	32.6	HLDACETMGNATAICSDK	y5	heavy	PMCA-3	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
667.9519	701.3236	32.6	HLDACETMGNATAICSDK	y6	heavy	PMCA-3	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
667.9519	802.3713	32.6	HLDACETMGNATAICSDK	y7	heavy	PMCA-3	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
675.3163	663.3130	26.3	LTDEEVDEMIR	y5	light	CAM-1	Cam	CG8472	NP_523710.1	Calmodulin
675.3163	762.3815	26.3	LTDEEVDEMIR	y6	light	CAM-1	Cam	CG8472	NP_523710.1	Calmodulin
675.3163	1135.4936	26.3	LTDEEVDEMIR	y9	light	CAM-1	Cam	CG8472	NP_523710.1	Calmodulin
676.8019	638.3144	26.3	ASEDPQSASFDAK	y6	light	InsP3R-1	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
676.8019	725.3464	26.3	ASEDPQSASFDAK	y7	light	InsP3R-1	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
676.8019	950.4578	26.3	ASEDPQSASFDAK	y9	light	InsP3R-1	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
680.3163	673.3130	26.3	LTDEEVDEMIR	y5	heavy	CAM-1	Cam	CG8472	NP_523710.1	Calmodulin
680.3163	772.3815	26.3	LTDEEVDEMIR	y6	heavy	CAM-1	Cam	CG8472	NP_523710.1	Calmodulin
680.3163	1145.4936	26.3	LTDEEVDEMIR	y9	heavy	CAM-1	Cam	CG8472	NP_523710.1	Calmodulin
680.8019	646.3144	26.3	ASEDPQSASFDAK	y6	heavy	InsP3R-1	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
680.8019	733.3464	26.3	ASEDPQSASFDAK	y7	heavy	InsP3R-1	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
680.8019	958.4578	26.3	ASEDPQSASFDAK	y9	heavy	InsP3R-1	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
726.7041	665.3770	35.3	LVANNGLNPVYNEDPFVFR	y5	light	PLCNORP-1	norpA	CG362	NP_995604.1	no receptor potential A
726.7041	1023.4894	35.3	LVANNGLNPVYNEDPFVFR	y8	light	PLCNORP-1	norpA	CG11430	NP_611273.1	no receptor potential A
726.7041	1186.5528	35.3	LVANNGLNPVYNEDPFVFR	y9	light	PLCNORP-1	norpA	CG11430	NP_611273.1	no receptor potential A
729.8628	432.2201	28.1	NALGDVTNELQER	y3	light	STIM-2	Stim	CG9126	NP_996470.1	Stromal interaction molecule
729.8628	545.3042	28.1	NALGDVTNELQER	y4	light	STIM-2	Stim	CG9126	NP_996470.1	Stromal interaction molecule
729.8628	788.3897	28.1	NALGDVTNELQER	y6	light	STIM-2	Stim	CG9126	NP_996470.1	Stromal interaction molecule
729.8628	889.4374	28.1	NALGDVTNELQER	y7	light	STIM-2	Stim	CG9126	NP_996470.1	Stromal interaction molecule
730.0375	675.3770	35.3	LVANNGLNPVYNEDPFVFR	y5	heavy	PLCNORP-1	norpA	CG362	NP_995604.1	no receptor potential A
730.0375	1033.4894	35.3	LVANNGLNPVYNEDPFVFR	y8	heavy	PLCNORP-1	norpA	CG11430	NP_611273.1	no receptor potential A
730.0375	1196.5528	35.3	LVANNGLNPVYNEDPFVFR	y9	heavy	PLCNORP-1	norpA	CG11430	NP_611273.1	no receptor potential A
734.8628	442.2201	28.1	NALGDVTNELQER	y3	heavy	STIM-2	Stim	CG9126	NP_996470.1	Stromal interaction molecule
734.8628	555.3042	28.1	NALGDVTNELQER	y4	heavy	STIM-2	Stim	CG9126	NP_996470.1	Stromal interaction molecule
734.8628	798.3897	28.1	NALGDVTNELQER	y6	heavy	STIM-2	Stim	CG9126	NP_996470.1	Stromal interaction molecule
734.8628	899.4374	28.1	NALGDVTNELQER	y7	heavy	STIM-2	Stim	CG9126	NP_996470.1	Stromal interaction molecule
766.8594	581.3293	29.4	FEGSSESDLSTFVK	y5	light	DISIS-1	ERp60	CG8983	NP_524211.1	CG8983 gene product from transcript CG8983-RA
766.8594	694.4134	29.4	FEGSSESDLSTFVK	y6	light	DISIS-1	ERp60	CG8983	NP_524211.1	CG8983 gene product from transcript CG8983-RA
766.8594	896.4724	29.4	FEGSSESDLSTFVK	y8	light	DISIS-1	ERp60	CG8983	NP_524211.1	CG8983 gene product from transcript CG8983-RA
766.8594	1112.5470	29.4	FEGSSESDLSTFVK	y10	light	DISIS-1	ERp60	CG8983	NP_524211.1	CG8983 gene product from transcript CG8983-RA
770.8594	589.3293	29.4	FEGSSESDLSTFVK	y5	heavy	DISIS-1	ERp60	CG8983	NP_524211.1	CG8983 gene product from transcript CG8983-RA
770.8594	702.4134	29.4	FEGSSESDLSTFVK	y6	heavy	DISIS-1	ERp60	CG8983	NP_524211.1	CG8983 gene product from transcript CG8983-RA
770.8594	904.4724	29.4	FEGSSESDLSTFVK	y8	heavy	DISIS-1	ERp60	CG8983	NP_524211.1	CG8983 gene product from transcript CG8983-RA
770.8594	1120.5470	29.4	FEGSSESDLSTFVK	y10	heavy	DISIS-1	ERp60	CG8983	NP_524211.1	CG8983 gene product from transcript CG8983-RA
772.4325	460.2766	29.6	VGEATETALIVLAEK	y4	light	SERCA-2	Ca-P60A	CG3725	NP_476832.1	Calcium ATPase at 60A
772.4325	559.3450	29.6	VGEATETALIVLAEK	y5	light	SERCA-2	Ca-P60A	CG3725	NP_476832.1	Calcium ATPase at 60A
772.4325	785.5131	29.6	VGEATETALIVLAEK	y7	light	SERCA-2	Ca-P60A	CG3725	NP_476832.1	Calcium ATPase at 60A
772.4325	957.5979	29.6	VGEATETALIVLAEK	y9	light	SERCA-2	Ca-P60A	CG3725	NP_476832.1	Calcium ATPase at 60A
776.4325	468.2766	29.6	VGEATETALIVLAEK	y4	heavy	SERCA-2	Ca-P60A	CG3725	NP_476832.1	Calcium ATPase at 60A
776.4325	567.3450	29.6	VGEATETALIVLAEK	y5	heavy	SERCA-2	Ca-P60A	CG3725	NP_476832.1	Calcium ATPase at 60A
776.4325	793.5131	29.6	VGEATETALIVLAEK	y7	heavy	SERCA-2	Ca-P60A	CG3725	NP_476832.1	Calcium ATPase at 60A
776.4325	965.5979	29.6	VGEATETALIVLAEK	y9	heavy	SERCA-2	Ca-P60A	CG3725	NP_476832.1	Calcium ATPase at 60A
778.4143	505.2617	29.8	EIVPGDLVEVSVGDK	y5	light	SERCA-3	Ca-P60A	CG3725	NP_476832.1	Calcium ATPase at 60A
778.4143	604.3301	29.8	EIVPGDLVEVSVGDK	y6	light	SERCA-3	Ca-P60A	CG3725	NP_476832.1	Calcium ATPase at 60A
778.4143	733.3727	29.8	EIVPGDLVEVSVGDK	y7	light	SERCA-3	Ca-P60A	CG3725	NP_476832.1	Calcium ATPase at 60A
778.4143	832.4411	29.8	EIVPGDLVEVSVGDK	y8	light	SERCA-3	Ca-P60A	CG3725	NP_476832.1	Calcium ATPase at 60A

Supp. Table 1: List of measured peptides and transitions (continued)

m/z Q1	m/z Q3	CE [V]	Sequence	fragment	light,heavy	Peptide Name	Gene Symb	CG#	NP#	Gene Description
794.9433	446.2973	30.3	IDQSILTGESVSVIK	y4	light	SERCA-1	Ca-P60A	CG18803	NP_001137988.1	Calcium ATPase at 60A
794.9433	818.4618	30.3	IDQSILTGESVSVIK	y8	light	SERCA-1	Ca-P60A	CG18803	NP_001137988.1	Calcium ATPase at 60A
794.9433	919.5095	30.3	IDQSILTGESVSVIK	y9	light	SERCA-1	Ca-P60A	CG18803	NP_001137988.1	Calcium ATPase at 60A
794.9433	1032.5936	30.3	IDQSILTGESVSVIK	y10	light	SERCA-1	Ca-P60A	CG18803	NP_001137988.1	Calcium ATPase at 60A
798.9433	454.2973	30.3	IDQSILTGESVSVIK	y4	heavy	SERCA-1	Ca-P60A	CG18803	NP_001137988.1	Calcium ATPase at 60A
798.9433	826.4618	30.3	IDQSILTGESVSVIK	y8	heavy	SERCA-1	Ca-P60A	CG18803	NP_001137988.1	Calcium ATPase at 60A
798.9433	927.5095	30.3	IDQSILTGESVSVIK	y9	heavy	SERCA-1	Ca-P60A	CG18803	NP_001137988.1	Calcium ATPase at 60A
798.9433	1040.5936	30.3	IDQSILTGESVSVIK	y10	heavy	SERCA-1	Ca-P60A	CG18803	NP_001137988.1	Calcium ATPase at 60A
804.9082	560.3191	30.7	EICTLGFDLWQVK	y4	light	CALRET-1	Crc	CG9429	NP_524293.2	Calreticulin
804.9082	788.4301	30.7	EICTLGFDLWQVK	y6	light	CALRET-1	Crc	CG9429	NP_524293.2	Calreticulin
804.9082	992.5200	30.7	EICTLGFDLWQVK	y8	light	CALRET-1	Crc	CG9429	NP_524293.2	Calreticulin
808.9082	568.3191	30.7	EICTLGFDLWQVK	y4	heavy	CALRET-1	Crc	CG9429	NP_524293.2	Calreticulin
808.9082	796.4301	30.7	EICTLGFDLWQVK	y6	heavy	CALRET-1	Crc	CG9429	NP_524293.2	Calreticulin
808.9082	1000.5200	30.7	EICTLGFDLWQVK	y8	heavy	CALRET-1	Crc	CG9429	NP_524293.2	Calreticulin
821.9178	428.2867	31.3	EVVAVTGDGTNDGPALK	y4	light	PMCA-1	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
821.9178	485.3082	31.3	EVVAVTGDGTNDGPALK	y5	light	PMCA-1	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
821.9178	872.4472	31.3	EVVAVTGDGTNDGPALK	y9	light	PMCA-1	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
821.9178	1044.4956	31.3	EVVAVTGDGTNDGPALK	y11	light	PMCA-1	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
821.9178	1145.5433	31.3	EVVAVTGDGTNDGPALK	y12	light	PMCA-1	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
825.9178	436.2867	31.3	EVVAVTGDGTNDGPALK	y4	heavy	PMCA-1	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
825.9178	493.3082	31.3	EVVAVTGDGTNDGPALK	y5	heavy	PMCA-1	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
825.9178	880.4472	31.3	EVVAVTGDGTNDGPALK	y9	heavy	PMCA-1	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
825.9178	1052.4956	31.3	EVVAVTGDGTNDGPALK	y11	heavy	PMCA-1	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
825.9178	1153.5433	31.3	EVVAVTGDGTNDGPALK	y12	heavy	PMCA-1	PMCA	, CG34036, C	NP_001014687.3	plasma membrane calcium ATPase
827.0262	466.2330	39.7	EADIDGDGQVNYEEFVTMM	y4	light	CAM-2	Cam	CG8472	NP_523710.1	Calmodulin
827.0262	597.2735	39.7	EADIDGDGQVNYEEFVTMM	y5	light	CAM-2	Cam	CG8472	NP_523710.1	Calmodulin
827.0262	698.3212	39.7	EADIDGDGQVNYEEFVTMM	y6	light	CAM-2	Cam	CG8472	NP_523710.1	Calmodulin
827.0262	797.3896	39.7	EADIDGDGQVNYEEFVTMM	y7	light	CAM-2	Cam	CG8472	NP_523710.1	Calmodulin
829.6928	474.2330	39.7	EADIDGDGQVNYEEFVTMM	y4	heavy	CAM-2	Cam	CG8472	NP_523710.1	Calmodulin
829.6928	605.2735	39.7	EADIDGDGQVNYEEFVTMM	y5	heavy	CAM-2	Cam	CG8472	NP_523710.1	Calmodulin
829.6928	706.3212	39.7	EADIDGDGQVNYEEFVTMM	y6	heavy	CAM-2	Cam	CG8472	NP_523710.1	Calmodulin
829.6928	805.3896	39.7	EADIDGDGQVNYEEFVTMM	y7	heavy	CAM-2	Cam	CG8472	NP_523710.1	Calmodulin
847.0889	460.2766	40.6	NIHSGEESSAISVNSPLEDILA	y4	light	InsP3R-2	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
847.0889	573.3606	40.6	NIHSGEESSAISVNSPLEDILA	y5	light	InsP3R-2	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
847.0889	688.3876	40.6	NIHSGEESSAISVNSPLEDILA	y6	light	InsP3R-2	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
847.0889	817.4302	40.6	NIHSGEESSAISVNSPLEDILA	y7	light	InsP3R-2	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
848.4089	448.2402	32.2	TMAISSELTNAESDVK	y4	light	InR-1	InR	CG18402	NP_524436.2	Insulin-like receptor
848.4089	762.3628	32.2	TMAISSELTNAESDVK	y7	light	InR-1	InR	CG18402	NP_524436.2	Insulin-like receptor
848.4089	863.4105	32.2	TMAISSELTNAESDVK	y8	light	InR-1	InR	CG18402	NP_524436.2	Insulin-like receptor
848.4089	976.4946	32.2	TMAISSELTNAESDVK	y9	light	InR-1	InR	CG18402	NP_524436.2	Insulin-like receptor
849.7556	468.2766	40.6	NIHSGEESSAISVNSPLEDILA	y4	heavy	InsP3R-2	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
849.7556	581.3606	40.6	NIHSGEESSAISVNSPLEDILA	y5	heavy	InsP3R-2	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
849.7556	696.3876	40.6	NIHSGEESSAISVNSPLEDILA	y6	heavy	InsP3R-2	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
849.7556	825.4302	40.6	NIHSGEESSAISVNSPLEDILA	y7	heavy	InsP3R-2	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
852.4089	456.2402	32.2	TMAISSELTNAESDVK	y4	heavy	InR-1	InR	CG18402	NP_524436.2	Insulin-like receptor
852.4089	770.3628	32.2	TMAISSELTNAESDVK	y7	heavy	InR-1	InR	CG18402	NP_524436.2	Insulin-like receptor
852.4089	871.4105	32.2	TMAISSELTNAESDVK	y8	heavy	InR-1	InR	CG18402	NP_524436.2	Insulin-like receptor
852.4089	984.4946	32.2	TMAISSELTNAESDVK	y9	heavy	InR-1	InR	CG18402	NP_524436.2	Insulin-like receptor
871.3780	742.2825	32.9	TLWDDAGIQECYDR	y5	light	GAQ-1	CG9911	CG17759	NP_523718.1	G protein alpha49B
871.3780	870.3410	32.9	TLWDDAGIQECYDR	y6	light	GAQ-1	CG9911	CG17759	NP_523718.1	G protein alpha49B
871.3780	1040.4466	32.9	TLWDDAGIQECYDR	y8	light	GAQ-1	CG9911	CG17759	NP_523718.1	G protein alpha49B
876.3780	752.2825	32.9	TLWDDAGIQECYDR	y5	heavy	GAQ-1	CG9911	CG17759	NP_523718.1	G protein alpha49B
876.3780	880.3410	32.9	TLWDDAGIQECYDR	y6	heavy	GAQ-1	CG9911	CG17759	NP_523718.1	G protein alpha49B
876.3780	1050.4466	32.9	TLWDDAGIQECYDR	y8	heavy	GAQ-1	CG9911	CG17759	NP_523718.1	G protein alpha49B
882.9103	532.3089	33.3	ITNSTEDLDDESIQGK	y5	light	STIM-1	Stim	CG9126	NP_996470.1	Stromal interaction molecule
882.9103	776.3785	33.3	ITNSTEDLDDESIQGK	y7	light	STIM-1	Stim	CG9126	NP_996470.1	Stromal interaction molecule
882.9103	891.4054	33.3	ITNSTEDLDDESIQGK	y8	light	STIM-1	Stim	CG9126	NP_996470.1	Stromal interaction molecule
886.9103	540.3089	33.3	ITNSTEDLDDESIQGK	y5	heavy	STIM-1	Stim	CG9126	NP_996470.1	Stromal interaction molecule
886.9103	784.3785	33.3	ITNSTEDLDDESIQGK	y7	heavy	STIM-1	Stim	CG9126	NP_996470.1	Stromal interaction molecule
886.9103	899.4054	33.3	ITNSTEDLDDESIQGK	y8	heavy	STIM-1	Stim	CG9126	NP_996470.1	Stromal interaction molecule
894.9435	530.3297	33.7	AMSLAAVDADGEQIELR	y4	light	InsP3R-3	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
894.9435	658.3883	33.7	AMSLAAVDADGEQIELR	y5	light	InsP3R-3	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
894.9435	844.4523	33.7	AMSLAAVDADGEQIELR	y7	light	InsP3R-3	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
894.9435	959.4793	33.7	AMSLAAVDADGEQIELR	y8	light	InsP3R-3	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
899.9435	540.3297	33.7	AMSLAAVDADGEQIELR	y4	heavy	InsP3R-3	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
899.9435	668.3883	33.7	AMSLAAVDADGEQIELR	y5	heavy	InsP3R-3	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
899.9435	854.4523	33.7	AMSLAAVDADGEQIELR	y7	heavy	InsP3R-3	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
899.9435	969.4793	33.7	AMSLAAVDADGEQIELR	y8	heavy	InsP3R-3	Itp-r83A	CG1063	NP_730941.1	Inositol 1,4,5,-tris-phosphate receptor
973.4893	516.3140	36.4	EAPTLSSSNSDLEVQQLK	y4	light	STIM-3	Stim	CG9126	NP_996470.1	Stromal interaction molecule
973.4893	615.3824	36.4	EAPTLSSSNSDLEVQQLK	y5	light	STIM-3	Stim	CG9126	NP_996470.1	Stromal interaction molecule
973.4893	744.4250	36.4	EAPTLSSSNSDLEVQQLK	y6	light	STIM-3	Stim	CG9126	NP_996470.1	Stromal interaction molecule
973.4893	857.5091	36.4	EAPTLSSSNSDLEVQQLK	y7	light	STIM-3	Stim	CG9126	NP_996470.1	Stromal interaction molecule
977.4893	524.3140	36.4	EAPTLSSSNSDLEVQQLK	y4	heavy	STIM-3	Stim	CG9126	NP_996470.1	Stromal interaction molecule
977.4893	623.3824	36.4	EAPTLSSSNSDLEVQQLK	y5	heavy	STIM-3	Stim	CG9126	NP_996470.1	Stromal interaction molecule
977.4893	752.4250	36.4	EAPTLSSSNSDLEVQQLK	y6	heavy	STIM-3	Stim	CG9126	NP_996470.1	Stromal interaction molecule
536.3241	845.4727	21.5	IILLGDSGVGK	y9	light	CG32679	Rab9D	CG32678	NP_727432.1	Rab GTPase 9D
546.7878	879.4571	21.9	VNFTVDEIR	y7	light	CG2238	Ef2b	CG2238	NP_525105.2	Elongation factor 2b
547.2875	638.3297	21.9	LNDLFGTWK	y5	light	CG5502	RpL4	CG5502	NP_524538.2	Ribosomal Protein L4
611.8213	1024.4986	24.1	VVELFDEFPK	y8	light	CG7490	RpLP0	CG7490	NP_524211.1	Ribosomal protein LP0
611.8613	796.4676	24.1	VLVDGPLTGVPR	y8	light	CG6253	RpL14	CG6253	NP_523975.1	Ribosomal protein L14
638.8666	818.4367	25	VFIVTGANTGIGK	y9	light	CG2064	CG2064	CG2064	NP_610310.2	CG2064 gene product from transcript CG2064-RA
644.8375	774.4104	25.2	GLCAIAQAESLR	y7	light	CG6779	RpS3	CG6779	NP_476632.1	Ribosomal protein S3
649.8512	803.4258	25.4	DIPGLTDTTIPR	y7	light	CG10944	RpS6	CG10944	NP_511073.1	Ribosomal protein S6
714.3239	794.3648	27.6	FDSGNLCMITGGR	y7	light	CG11276	RpS4	CG11276	NP_729871.1	Ribosomal protein S4
715.8767	805.3873	27.7	NIVWIAECVAQK	y7	light	CG2934	VhaAC39	CG2934	NP_570080.1	CG2934 gene product from transcript CG2934-RA
716.8619	503.2824	27.7	EAGEDTTLIELNK	y4	light	CG13887	CG13887	CG13887	NP_612062.1	CG13887 gene product from transcript CG13887-RC
739.8779	1022.5153	28.5	TAANVEEAFINTAK	y9	light	CG3269	Rab2	CG3269	NP_477090.1	Rab-protein 2
743.3808	685.3991	28.6	GVVDSDDLPLNVSR	y6	light	CG5520	Gp93	CG5520	NP_651601.1	Glycoprotein 93
780.3999	919.4381	29.8	GLQLTQPNTNNFGR	y8	light	CG3922	RpS17	CG4326	NP_524002.1	Ribosomal protein S17
865.4282	872.4724	32.7	EAAAGEDITPLADESIK	y8	light	CG13388	Akap200	CG13388	NP_477459.1	A kinase anchor protein 200

Supp. Table 2: Heavy Peptide Mix

Heavy Peptides	Concentration (pmol/ul)	ul / Mix	Final volume of stock solution	Final concentration in stock (pmol/ul)
CalRet	5.0	50.0	2050	0.1220
CAM1	5.0	50.0	2050	0.1220
CAM2	5.0	50.0	2050	0.1220
CAM3	5.0	25.0	2050	0.0610
DISIS1	3.4	50.0	2050	0.0829
ERP44	2.9	16.6	2050	0.0235
GAQ1	4.3	25.0	2050	0.0524
InR1	6.8	16.6	2050	0.0551
InsP3R-1	5.0	5.0	2050	0.0122
InsP3R-2	5.0	16.6	2050	0.0405
InsP3R-3	5.0	50.0	2050	0.1220
MITCAR1	5.0	50.0	2050	0.1220
ORAI1	5.0	16.6	2050	0.0405
ORAi2	5.0	5.0	2050	0.0122
ORAi3	5.0	5.0	2050	0.0122
PLCNORP	8.1	16.6	2050	0.0656
PMCA1	5.0	25.0	2050	0.0610
PMCA2	5.0	50.0	2050	0.1220
PMCA3	5.0	50.0	2050	0.1220
PRESEN1	5.7	75.0	2050	0.2085
SERCA1	5.0	50.0	2050	0.1220
SERCA2	5.0	50.0	2050	0.1220
SERCA3	5.0	25.0	2050	0.0610
STIM1	5.0	25.0	2050	0.0610
STIM2	5.0	50.0	2050	0.1220
STIM3	5.0	25.0	2050	0.0610

Supp. Table 3: Primers used to make dsRNA

Primer Name	**Primer Sequence**
STIM_forward	TAATACGACTCACTATAGGGCAGAACATTCTCCAAGTAGGGC
STIM_reverse	TAATACGACTCACTATAGGGAAAAACTAGATTTGGAGCGTCG
PMCA_forward	TAATACGACTCACTATAGGGAATGCTTCACCAAGTTATTGGC
PMCA_reverse	TAATACGACTCACTATAGGGATGGTCGATCGCAAATAAAGG
SERCA_forward	TAATACGACTCACTATAGGGCTGCTGACTACGATACCCTGC
SERCA_reverse	TAATACGACTCACTATAGGGGACAATGGAATCGAAAACTTCC
YFP_forward	GCGTAATACGACTCACTATAGGCATCCTGGTCGAGCTGGAC
YFP_reverse	GCGTAATACGACTCACTATAGGCGTTGGGGTCTTTGCTCAG
GL3_forward	GCGTAATACGACTCACTATAGGGCGGTCGGTAAAGTTGTTC
GL3_reverse	GCGTAATACGACTCACTATAGGTCTTGCGTCGAGTTTTCCG
MICU1-1_forward	TAA TAC GAC TCA CTA TAG GGA CTC GGC TTT GAT CAC ATA TTT C
MICU1-1_reverse	TAA TAC GAC TCA CTA TAG GGT TCT CAT CAA AGA TGG TAA AGA CC
MICU1-2_forward	TAA TAC GAC TCA CTA TAG GGT GCA GAA CTC CTA CTG GCC TAC
MICU1-2_reverse	TAA TAC GAC TCA CTA TAG GGT TCT CAT CAA AGA TGG TAA AGA CC
CALX-1_forward	TAA TAC GAC TCA CTA TAG GGT GTA GCG AGG GTC TTG TCC
CALX-1_reverse	TAA TAC GAC TCA CTA TAG GGG CGG TAA CGA AGA AGA CTC
CALX-2_forward	TAA TAC GAC TCA CTA TAG GGT ACG TGA GCC ACT TCG TC
CALX-2_reverse	TAA TAC GAC TCA CTA TAG GGC ATG CTG GCG AAT GTA TC