

SUPPLEMENTAL INFORMATION -

TITLE

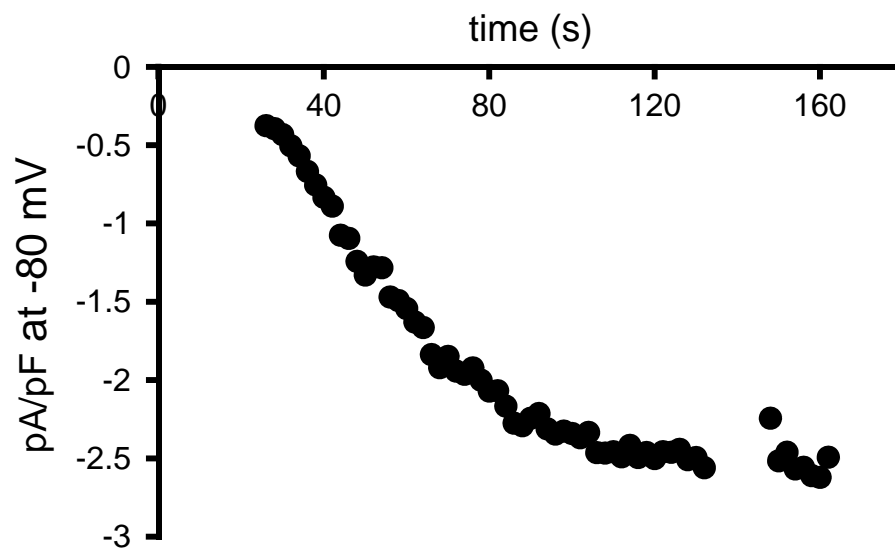
How Many Orai's Does It Take to Make a CRAC Channel?

AUTHORS

Jill L. Thompson and Trevor J. Shuttleworth*

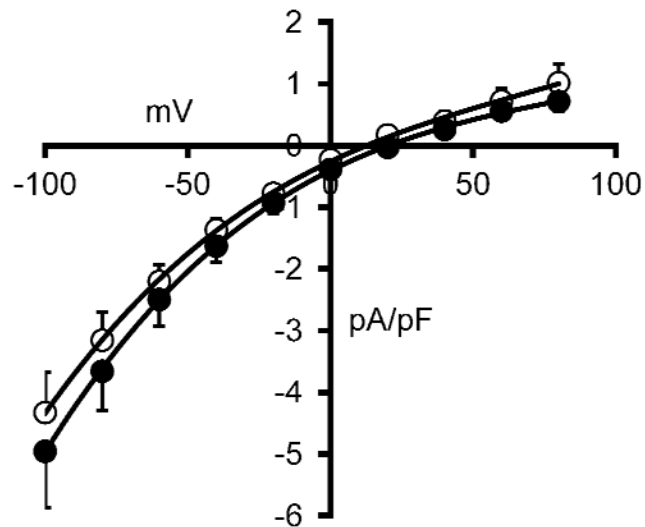
Supplemental Figure 1

Representative trace showing the activation of La^{3+} -sensitive store-operated currents in a HEK cell stably expressing STIM1 and transfected with the concatenated Orai1 hexamer construct. Inward currents measured at -80 mV were progressively activated on achieving whole-cell patch clamp conditions by inclusion of adenophostin A (2 μM) in the pipette solution. Gaps in the trace reflect the times during which current-voltage relationships were recorded.



Supplemental Figure 2

Comparison of the mean (\pm SE) I/V curves for store-operated currents recorded between -100 mV and +80 mV in STIM1-stable cells expressing the concatenated Orai1 hexamer (black symbols, $n = 6$ – data taken from Fig. 1a) and in the same cells transfected with additional STIM1 (white symbols, $n = 5$). The external medium was the standard $\text{Na}^+/\text{Ca}^{2+}$ solution (140 mM Na^+ and 10 mM Ca^{2+}).



Supplemental Figure 3

Representative traces showing the minimal calcium-dependent fast-inactivation (CDI) displayed in cells expressing the concatenated Orai hexamer construct.

Store-operated currents ($2\ \mu\text{M}$ adenophostin A in the pipette) were measured in cells in a sodium-free external solution containing 110 mM calcium (solution

composition, in mM: 110 CaCl_2 , 10 glucose, 1.2 MgCl_2 , 10 Hepes, pH 7.4). Once

maximal activation was achieved, inward currents were measured at a frequency of 5 kHz during 280 ms pulses to $-120\ \text{mV}$ from the holding potential of 0 mV

applied every 2 seconds. All recordings were low-pass filtered with a 1-kHz Bessel filter.

