Molecular mechanisms of redox regulation of Orai1 channels

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Oxidants interacting with reactive cysteine residues can result in altered protein function. Store-operated calcium entry (SOCE) mediated by STIM1 gated Orai1 channels is the major Ca^{2+} entry pathway to activate immune cells and both loss-of-function and gain-of-function mutations lead to immune dysfunction and other pathologies. We have previously shown that pretreatment of the Ca^{2+} selective Orai1 but not its paralogue Orai3 with the oxidant H_2O_2 leads to reduced I_{CRAC} and that C195 is its major ROS sensor [1]. However, the underlying mechanism of inhibition remained elusive. The current work combines patch-clamp analysis, fluorescence microscopy and mutational approaches based on structural information and molecular dynamics simulations with a theoretical reaction diffusion model to explore the molecular mechanisms underlying inhibition of I_{CRAC} by ROS. We show that oxidized Orai1 C195 at the exit of transmembrane domain 3 leads to reduced subunit interaction, slowed diffusion and that either oxidized C195 or its oxidomimetic mutation C195D hinders channel activation by intramolecular interaction with S239 of TM4. Our results reveal the mechanism underlying ROS inhibition of Orai1 and identify a candidate residue for pharmaceutical intervention.

[1] I.Bogeski and C.Kummerow, D.Al-Ansary, E.C.Schwarz, R.Koehler, D.Kozai, N.Takahashi, C.Peinelt, D.Griesemer, M.Bozem, Y.Mori, M.Hoth, B.A Niemeyer. Sci. Sig, 3(115):ra24. doi: 10.1126/scisignal.2000672.