Modulation of ryanodine receptor's activity by homer and a ryanodine receptor C-terminal tail peptide

P. Pouliquin, S. Pace, S.M. Curtis, P.J. Harvey, M.W. Casarotto, A.F. Dulhunty, Division of Molecular Biosciences, John Curtin School of Medical Research-ANU, Canberra, Act, Australia

The ryanodine receptor (RyR) Ca^{2+} release channel is a central player in cytoplasmic Ca^{2+} regulation in many eukaryotic cells. The function of the channel is thought to be maintained by interactions (a) between its 4 subunits which may involve its C-terminal tail and (b) with other regulatory proteins which include the adaptor protein Homer. Here we examine the structure and function of a peptide (CTT19) which corresponds to the extreme 19 C-terminal residues of the skeletal muscle RyR1. Approximately half the residues in CTT19 form an α helix, which is also predicted by sequence alignment with voltage gated K+ channels. The peptide at concentrations of 100nM to 10Î¼M reduced the opening of skeletal RyR1 and cardiac RyR2 channels, and stabilised channel openings to submaximal conductance levels. The long form of Homer (Homer 1B) at a concentration of 12nM activated RyR1 channels and prevented channel inhibition by CCT19. In contrast, 12nM Homer inhibited RyR2 channels and channel activity declined further after addition of CTT19. These data suggest that RyR function depends on inter-domain interactions involving the C-terminal tail, and that these interactions are destabilised by the domain peptide CTT19. The data further suggest that (a) Homer binding to RyR1 stabilizes inter-domain interactions and prevents their disruption by CTT19 and (b) in contrast Homer binding to RyR2 does not stabilize inter-domain interactions as it doesn't prevent their disruption by CTT19. The different effects of Homer binding to RyR1 and RyR2 may be due to differences in Homer binding sites in RyR1 (three predicted binding sites) and RyR2 (two predicted binding sites). The results provide evidence for the role of the C-terminal tail of the RyR in channel function and indicate that adaptive Homer may stabilize the RyR1 channel.