A new zinc modulation site on glycine receptor alpha 1 subunit is associated with human hyperekplexia

The divalent cation zinc modulates several types of synaptic receptors, including glycine receptors, $GABA_A$ receptors, and NMDA receptors. In physiological condition, zinc also modulates synaptic transmission. The transgenic mice with mutant glycine alpha1 receptors lacking sensitivity to zinc potentiation develop startle responses that resembles human hyperekplexia. However, it is still not clear whether zinc modulation on synaptic receptors is involved in human diseases. Here, we report that a single mutation (W170S) in glycine receptor alpha1 subunit, which was recently identified from Omani families with hyperekplexia and mild mental retardation, caused almost complete loss of zinc-mediated potentiation and enhanced zinc-mediated inhibition. Whole-cell patch clamp recordings in HEK293 cells transfected either wild type or W170S-containing glycine receptors revealed that the potentiation effect of zinc (0.1 to 10 micromolars) was impaired in both alpha1W^{170S} and alpha1^{W170S}/beta compositions of glycine receptors. Inhibitory effects by zinc (10 to 50 micromolars) were observed in wild type receptors, and this inhibition was enhanced in W170S receptors. The impairment of zinc-mediated potentiation was observed in glycine receptor currents mediated by three different endogenous agonists, including glycine, taurine, and beta-alanine. To temporally differentiate between the potentiating and the inhibitory effects of zinc at higher concentrations (>10 micromolar) on glycine alpha1 receptors, we applied zinc after the agonist-binding site has already been activated by glycine. We found that 10-50 micromolar zinc induced potentiation on wild type glycine currents, but caused inhibition in W170S receptors. To further unmask the potentiating effect of higher concentrations of zinc, we generated W170S mutation on the background of H107N, which is a previously reported mutation lacking zinc inhibition. W170S/H107N double mutation strongly attenuated zinc-mediated potentiation to less than 10% of H107N single mutation receptors. Moreover, we have confirmed that the alpha1^{W170S}/beta receptors had no significant effect on other electrophysiological properties of glycine receptors, including the glycine-induced maximum responses, dose-response curves of glycine, taurine, and beta-alanine, strychnine-mediated inhibition, and current-voltage curves compared to wild type receptors. Taken together, our study has discovered a new zinc potentiation site on glycine alpha 1 receptors and also revealed a strong link between synaptic zinc modulation and human disease.