The dominant-negative effect of Loss of function Nav 1.5 variants is not a critical determinant of phenotype severity

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Introduction: Loss-of-function (LOF) variants of SCN5A cause various arrhythmic diseases. Recent studies have demonstrated that some LOF-SCN5A variants can exert a dominant-negative (DN) effect through channel dimerization, resulting in higher penetrance of disease phenotypes. However, the association between the DN effect and phenotype severity remains unclear. In this study, we identified a proband with compound heterozygous SCN5A variants, p.G833R and p.T1396P, in a family with bradyarrhythmia. We analyzed the electrophysiological function of these variants and their association with channel-dimerization to elucidate their effects on phenotype severity.

Purpose: The purpose of this study is to investigate the effect of sodium channel dimerization on the phenotype severity in a family with bradyarrhythmia.

Methods: We performed functional analysis of the mutant Nav1.5s induced in HEK cells utilizing a whole-cell patch-clamp method. The DN effect was also assessed by co-expressing WT and mutants in each heterozygous combinations, with or without difopein, a peptide working as a channel-coupling canceler. Cell surface expression of mutated channels were measured by flowcytometry.

Results: We identified SCN5A p.T1396P and p.G833R variants segregating in a family suffering from severe bradyarrhythmia. The proband carried both variants in a compound heterozygous manner, while her mother carried only p.T1396P. The clinical phenotype was more severe and had early onset in the proband; the compound heterozygous case.

In the patch-clamp study, HEK 293 cells solely expressing T1396P did not show any measurable sodium current, while those with G833R had comparative current with that of WT channel. Cells expressing WT/T1396P showed significant reduction and delay of inactivation decay, which was normalized by co-expression with difopein. The results implied that loss-of-function variant T1396P manifested DN-effect via channel dimerization.

Cells co-expressing T1396P and G833R did not demonstrate a DN-effect, suggesting that Nav1.5-G833R did not form a dimer. Considering more severe phenotype of compound heterozygous member in the family, the DN-cancelation by monomerized Nav1.5-G833R was not associated with a mild phenotype.

Conclusion: Our result supports the current Nav1.5-dimerization concept and consequent DN-effect by LOF-SCN5A variants. However, the phenotype severity was not simply explainable by the presence of a dominant negative effect. Further multiplanar analyses including assays for channel clustering, localization, and gating modification would be necessary.