Linking superinhibitory PLN mutations to CaMKII activation: a new arrhythmogenic mechanism in genetic DCM?

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This editorial refers to 'A novel human R25C-phospholamban mutation is associated with super-inhibition of calcium cycling and ventricular arrhythmia' by G.-S. Liu et al., pp. 164–174.

Genetically caused dilated cardiomyopathy (DCM) is typically diagnosed between 40 and 60 years and patients often present with heart failure or advanced arrhythmia, which may possibly lead to stroke or sudden cardiac death.1 While potentially lethal arrhythmias are commonly found in end-stage heart failure independent of its aetiology, there is growing evidence that in certain cases of DCM, arrhythmia and sudden cardiac death are present already in early stages and may precede the development of overt heart failure. So far, this subset of cases has been linked to mutations in LMNA, coding for lamin A/C, and SCN5A, coding for the cardiac fast Na+ channel α-subunit Na1.5.1 In the recent years, also a mutation in PLN, coding for phospholamban (PLB), R14del, has been connected to this subset of DCM.2–4

PLB is the main regulator of SERCA activity and thus a critical regulator of cardiac Ca2+ cycling. The association with PLB inhibits SERCA by reducing its apparent Ca2+ affinity. During β-adrenergic stress, PLB is phosphorylated by protein kinase A and dissociates from SERCA, resulting in SERCA disinhibition. This mechanism accelerates and increases the diastolic filling of the sarcoplasmic reticulum (SR), resulting in the lusitropic and contributing to the inotropic effects of β-receptor activation.5 On the other hand, diminished phosphorylation of PLB and a reduction in SERCA activity are the hallmarks of failing hearts.6 R14del increases the inhibition of SERCA by PLB, leading to a ‘superinhibition’.6

In an elegant study, Liu et al.7 report another human PLN mutation, R25C, that exhibits superinhibition of SERCA and causes DCM with prominent arrhythmias in patients, similar to the R14del mutation. Most importantly, they describe a molecular mechanism, by which superinhibition of SERCA may lead to cardiac arrhythmia (Figure 1). By overexpressing the PLN R25C in rat adult ventricular myocytes, they not only demonstrate a larger depression of contractility, systolic Ca2+ transients, and SR Ca2+ uptake compared with wild type, but also demonstrate a marked elevation of diastolic Ca2+ levels. This, in turn, is suggested to activate the Ca2+ and calmodulin-dependent protein kinase II (CaMKII), leading to increased phosphorylation of the S2814 site of the ryanodine receptor 2 (RyR), resulting in an increased Ca2+ leak from the SR and via activation of Na+/Ca2+ exchanger (NCX) leading to increased Ca2+ wave and spark frequency. The result of this are triggered aftercontractions, which may underlie the arrhythmogenic phenotype of this mutation in humans. Interestingly, by activation of the β-adrenergic signalling pathway, the inhibition of SERCA by the mutated PLB can be abrogated to the same extent as in wild type, indicating that phosphorylated R25C PLB dissociates from SERCA as well as wild type.

The study by Liu et al. is the first to suggest a molecular mechanism by which superinhibition of SERCA may lead to cardiac arrhythmia in genetic DCM. However, several factors indicate that in patients, the situation may be more complex than described. Interestingly, the increase in intracellular Ca2+ levels by superinhibition of SERCA seems sufficient to activate CaMKII, but not to directly activate NCX sufficiently to induce arrhythmia. This could be due to different sensitivities to Ca2+, but it is also possible that the increases in Ca2+ concentration by SERCA inhibition and by increased RyR leak take place in different Ca2+ microdomains that could be connected by CaMKII. Besides CaMKII, increased diastolic Ca2+ levels can also potentially activate the phosphatase calcineurin (CaN). CaN is involved in several signalling pathways, including the dephosphorylation of nuclear factors of activated T-cells (NFATs), which may contribute to the observed hypertrophy in the affected patients.8 Moreover, CaN, by dephosphorylating and thereby deactivating the protein phosphatase (PP) inhibitor 1 (I-1), can disinhibit PP1 and thus reduce phosphorylation of PLB and further increase the inhibition of SERCA.9 Notably, in the study of Liu et al.,7 there seems to be no obvious reduction in PLB phosphorylation in the transfected rat cardiomyocytes and it will be interesting to investigate whether the above-mentioned pathway may be active in animal models of R25C PLN and/or affected patients. The most puzzling finding is, however, that the described propensity for arrhythmia was present already 24 h post infection of the cardiomyocytes with adenovirus, whereas in patients it takes decades until they present with arrhythmic events.2 Maybe the first arrhythmic events are mild and ignored by the patients for years until severe arrhythmia develops. But most likely, a number of additional factors are involved in the arrhythmia development.

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pathophysiological mechanisms and modifying gene variants contribute to the pathogenesis of DCM and arrhythmia in patients suffering from the R25C mutation.

Can the mechanistic insights provided in the study be a first step towards a genotype-specific treatment for DCM caused by PLN mutations? β-Adrenoceptor blockers have been shown to improve outcomes in heart failure and are widely used in genetic DCM. By reducing SR Ca\(^{2+}\) load and Ca\(^{2+}\) leak, they exert a solid antiarrhythmic effect. However, in the case of R25C PLN, the molecular mechanism suggests potential deleterious effects of β-blockers in these patients, as β-adrenoceptor blockade could, in theory, aggravate the phenotype by reducing PLB phosphorylation. Proper surveillance and close monitoring of affected patients under β-adrenoceptor blockers seems warranted.

References