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A necessary connection: cholesterol and nicotinic receptors

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It is known that the muscle nicotinic acetylcholine receptor (nAChR) is highly influenced by its lipid environment. It is present in high-density clusters in the muscle cell membrane where it localizes mainly in liquid-ordered (Lo) domains enriched in cholesterol and sphingolipids. Its transmembrane domain forms the ion channel pore and exhibits extensive contacts with the surrounding lipids. In this work we studied the close relationship between nAChR and cholesterol under different experimental conditions in order to enrich with, deplete of, redistribute between both hemilayers, and oxidate cholesterol molecules. These conditions were evaluated either in *T. californica* nAChR-rich membranes, in model membranes containing purified nAChR or in cells expressing nAChR. Cholesterol modifications were confirmed by lipid analysis using thin layer chromatography. Evaluation of a) membrane order perturbations, by Laurdan GP and fluorescence anisotropy, b) increase/decrease of Lo domains, by fluorescence microscopy, c) nAChR-Lo domains correlation, by detergent treatment and SDS-PAGE, and d) nAChR conformation and function, by fluorescence spectroscopy and electrophysiology showed that changes in the amount, distribution or oxidation of cholesterol impacts not only in the size, location and curvature-domain shape of Lo domains and in the nAChR preference for them, but also in nAChR functionality and nAChR structural conformation. A high correlation between the quantitative presence of cholesterol, its transmembrane and lateral asymmetry and nAChR conformation and functionality is postulated.

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