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LES SÉMINAIRES DE L'INMG

*The endocytic machinery in healthy
and diseased muscle*

Par

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Costameres represent specialized focal adhesion sites of muscle fibres, located between the plasma membrane and sarcomeres, the contractile units of muscle. When disrupted, they directly contribute to the development of several distinct myopathies.

We have shown that the ubiquitous clathrin heavy chain (CHC), well characterized for its role in intracellular membrane traffic and endocytosis from the plasma membrane (PM), forms large plaques connected to α -actinin and actin filaments. Depletion of CHC leads to defective costamere formation and maintenance both *in vitro* and *in vivo* and induces sarcomere disorganization and a loss of contractile force due to the detachment of sarcomeres from the PM. At costameres, CHC is co-expressed with dynamin 2 (DNM2), another key protein of the intracellular membrane trafficking machinery which is mutated in autosomal dominant centronuclear myopathy (CNM). We analyzed the role of DNM2 and several actin binding proteins on clathrin plaque function at costameres *in vitro* by using either siRNA depletion combined to high resolution electron microscopy or *in vivo* by intravital microscopy. We also focused on the possible link between costamere and CNM pathophysiology. Using myoblasts from DNM2-mutated patients and using myoblasts and muscles from a knock-in mouse model of DNM2-related myopathy, we analyzed structure of costameres by biochemical and immunocytochemical approaches, as well as their ultrastructure.

Our results demonstrate a crucial role for the endocytic machinery and the cytoskeleton. Their contribution to the formation and maintenance of the contractile apparatus highlight an unconventional role for clathrin flat lattices in skeletal muscle which may be relevant to pathophysiology of several neuromuscular disorders.

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