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Caveolae associated proteins and how they effect caveolae dynamics

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Akademisk avhandling

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Abstract
The caveolae is a type of invaginated membrane domain that has been shown to be involved in several disease states, like lipodystrophy, muscular dystrophys and cancer. Several of the diseases are caused by lack of caveolae or caveolae-related signalling deficiencies in the tissues where caveolae domain are abundant such as lung, fat, muscle and their related endothelial cells. Caveolae are formed through the assembly of the transmembrane protein caveolin, cholesterol and the recently described family of cavin proteins, which together form the caveolae coat. The work in this thesis focuses on understanding the protein components and mechanisms that control the biogenesis and dynamics of caveolae.

We have found that the protein EHD2 is an important regulator and stabilizer of caveolae at the cell membrane. EHD2 is a dimeric ATPase known to oligomerise into ring-like structures around lipid membranes to control their shape. We have characterised the domain interactions involved in the specific targeting and assembly of this protein at caveolae. We propose a stringent regulatory mechanism for the assembly of EHD2 involving ATP binding and switching of the EH domain position to release the N-terminus and facilitate oligomerisation in the presence of membrane species. We show that loss of EHD2 in cells results in hyper-dynamic caveolae and that caveolae stability at the membrane can be restored by reintroducing EHD2 into these cells.

In a study of the protein cavin-3, which is known to be an integral component of the caveolae coat, we showed that this protein is targeted to caveolae via direct binding to the caveolae core protein caveolin1. Furthermore, we show that cavin-3 is enriched at deeply invaginated caveolae and regulate the duration time of caveolae at the cell surface .

In combination with a biochemical and cellbiological approach, the advanced fluorescence microscopy techniques, like Fluorescence Recovery After Photobleaching (FRAP), Total Internal Reflection microscopy (TIRF), combined with correlative Atomic Force Microscopy (AFM) have allowed us to characterise distinct caveolae-associated proteins and their respective functions at caveolae .

Keywords
caveolae, caveolin, EHD2, cavin, microdomain, microscopy, TIRF, AFM

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