Interactions of human profilin-1 and phosphatidylinositol 4,5-bisphosphate in giant unilamellar vesicles

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Human Profilin-I is a small actin binding proteins which also interact with polyphosphoinositides (PPI) (Lassing & Lindberg, 1985) and proline rich motif containing proteins. Profilin is involved in the signaling pathway linking the receptors in the cell membrane to the microfilament system. Profilin is thought to play critical roles in this signaling pathway through its interaction with phosphatidylinositol 4,5-bisphosphate $[PI(4,5)P_2]$ (Lu *et al.*, 1996). So far, profilin's interaction with PPI has only been studied in micelles or small vesicles. Profilin binds with high affinity to small clusters of $PI(4,5)P_2$ molecules (Goldschmidt-Clermont *et al.*, 1991). In cells, the organization of PPI lipids is different from micelles, therefore the interaction with profilin might be quite different.

We have used giant unilamellar vesicles (GUV) as a membrane model system to investigate the interactions between $PI(4,5)P_2$ and profilin. BODIPY® TMR $PI(4,5)P_2C16$ (Rhodamine labelled 16-carbon fatty acid containing Phosphatidylinositol 4, 5-bisphosphate) was incorporated into GUV membrane. Confocal images of GUVs were obtained in presence and absence of profilin. These images were analysed using SimFCS software (Prof. Enrico Gratton, Laboratory for Fluorescence Dynamics, Irvine, USA). The diffusion coefficient and the aggregation of PIP2 in the membrane were determined and profilin had a clear effect upon the diffusion coefficient and aggregation of PIP2.

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