Abstract

The journey of an eukaryotic protein typically begins in a ribosome and ends in a proteasome. What happens in between is a great diversity of functions that makes life possible, but the fate of most proteins is to be eventually degraded. A key components in this process are ubiquitin ligases. This thesis aims to provide more insight into the molecular basis of regulation of one such protein, the Nedd4-2.

Nedd4-2 is a member of the Nedd4 subfamily within the HECT family of ubiquitin ligases. Its most famous ubiquitination target is the Epithelial sodium channel, ENaC. It was reported that 14-3-3 protein and calcium modulate this and other interactions of Nedd4-2, but many questions remain to be answered about the structural basis of this regulation. Aiming to bring more insight at the molecular level, we performed measurements of small-angle x-ray scattering, analytical ultracentrifugation and hydrogen-deuterium exchange. Our conclusions bring new understanding in the area of ubiquitin ligases and for example can be applied in the new promising field of PROTAC drug design.

Keywords: Nedd4-2, 14-3-3 protein, calcium, protein interactions