Transportin-1 and -3 mediated co-regulation of CIRP nuclear import and LLPS

Benjamin Bourgeois[a], Saskia Hutten[b], Sinem Usler[a], Dorothee Dormann[b], Tobias Madl[a]

[a] Institute of Molecular Biology & Biochemistry, Center of Molecular Medicine, Medical University of Graz, 8010 Graz, Austria.
[b] BioMedical Center (BMC), Ludwig-Maximilians-University Munich, 82152 Planegg-Martinsried, Germany.

Chaperoning of proteins by the importins transportin-1/karyopherin-β2 (TNPO1) has been reported very recently by several groups, including ours (1). The chaperoning function of TNPO1 involves inhibition of cytoplasmic liquid-liquid phase separation (LLPS) of its cargos, and this was reported to depend strongly on binding of the importins to the NLS of the respective cargo proteins (1). In case of FUS, it has been shown that TNPO1 not only interacts with the PY-NLS, but also with the LLPS-mediating arginine/arginine-glycine (RG/RGG) repeat region, and that the interaction of TNPO1 with the RG/RGG motif is essential for chaperone function (1-3). Strikingly, mutations of FUS within its PY-NLS and RG/RGG motif are found in neurodegenerative diseases, including amyotrophic lateral sclerosis (ALS) and frontotemporal dementia (FTD) and cause distortion of nucleocytoplasmic shuttling and LLPS (1,3). The molecular basis for TNPO1 interaction with the highly abundant RG/RGG motifs and the interplay with the PY-NLS, however, are enigmatic. Proteins containing both the RG/RGG and the PY-NLS motifs are localizing differently within the cell, which raises the question whether the interplay between these motifs determines nuclear localization.

Here, we show for the first time using as model system the protein CIRBP that a RG/RGG containing protein lacking classical transportins NLS is able to bind efficiently to both transportin-1 and -3 thus regulating its nuclear import and LLPS. Our results on the molecular details of the CIRBP – transportin-1/-3 interaction set the base for understanding nuclear import of RG/RGG motif-containing proteins and its regulation by post-translational modifications and disease-associated mutations found in cancer and neurodegenerative diseases.

[1] Hofweber, M., et al., Cell (2018)

[2] Göbl C et al, Angewandte (2016)

[3] Dormann, D et al., EMBO. J, (2012)