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RNA-mediated gene expression regulation

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Akademisk avhandling

som med vederbörligt tillstånd av Rektor vid Umeå universitet för avläggande av filosofie doktorsexamen framläggs till offentligt försvar i Major Groove, Department of Molecular Biology, University hospital area, building 6L.

Fredag den 03 Mai, kl. 09:00.
Avhandlingen kommer att försvaras på engelsk.

Fakultetsopponent: Professor Henrik Nielsen,
Department of Cellular and Molecular Medicine, University of Copenhagen.

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Abstract

The regulation of gene expression is a key mechanism that underlies all biological processes, from embryonic development to the onset and progression of various diseases, including cancer. A growing body of evidence places RNA molecules at the center of critical regulatory steps in gene expression. They serve not only as intermediate molecules between DNA and proteins but also act as regulators of processes such as alternative splicing (AS) and translation, among others. This thesis focuses on the role of RNA in gene expression regulation. Specifically, it addresses how intrinsic properties of RNA, RNA chemical modifications, and RNA binding proteins (RBPs) can control gene expression regulatory processes.

The first part tackles specific aspects of AS in neurodifferentiation. **Paper I** shows how RBPs affect AS in mouse embryonic stem cells (ESCs). Within this work, we identified ZFP207, a known transcription factor (TF), as an RBP with a crucial role in modulating the AS of key transcripts for neurodifferentiation. Depletion of ZFP207 in mouse ESCs led to abnormal AS patterns and a differentiated cell phenotype.

The second part (Papers II-IV) focuses on the role of RNA modifications in disease. In **Paper II**, the publicly available literature linking deregulations of RNA modifications and their regulatory proteins with different diseases was curated. The obtained information was integrated into the 2021 update of the MODOMICS database, the most extensive RNA modifications database to date. Papers III and IV exemplify how two different RNA marks contribute to breast cancer. **Paper III** shows how METTL3, the enzyme responsible for N6-methyladenosine (m^6A) deposition on messenger RNA (mRNA), affects tumorigenesis by modulating AS. METTL3-mediated AS regulation can be done either by depositing m^6A at the intron-exon junctions of specific transcripts or on transcripts encoding for splicing and transcription factors, such as MYC. Changes in RNA modifications of ribosomal RNA (rRNA) affect stability, folding, and interactions with other molecules, leading to perturbed translation efficiency (TE). In **Paper IV**, we focused on the role of 2'-O-methylation, the most abundant rRNA modification, and its catalytic enzyme, fibrillarin (FBL), in triple-negative breast cancer (TNBC). We discovered that certain proto-oncogenes associated with breast cancer displayed a reduction in TE upon FBL depletion. Additionally, we identified 7 2'-O-methylation sites that might mediate TE regulation in a TNBC cellular model. Moreover, our study uncovered alterations in the ribosomal protein composition within the ribosomes of FBL-depleted cells. Our results support the pivotal role of 2'-O-methylation in controlling the translational capabilities of ribosomes in TNBC cells.

Overall, this work encompasses multiple aspects of gene expression regulation and describes how RNA modifications and RBPs modulate the fate of specific transcripts by controlling AS or translation.

Keywords: RNA modifications, fibrillarin, 2'-O-methylation, translation, alternative splicing, METTL3, m^6A, ZFP207, breast cancer, mouse ESCs.

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