Summary

Understanding transcriptional regulatory mechanisms has proven to be important in our knowledge of disease states, and several transcription factors are key targets of pharmaceutical drugs. Thus, understanding transcriptional regulatory mechanisms can aide in the development of drugs designed to specifically hit the desired target and minimise severe side effects. In some instances, different subtypes in a nuclear receptor family may regulate the expression of genes involved in opposite pathways, an observation that is quite intriguing in terms of understanding how nuclear receptors regulate transcription. Several studies have demonstrated intricate and elaborate transcription factor cooperativity through direct transcription factor protein-protein interactions. These interactions can be important in mediating the proper transcriptional response. However, the contribution of different domains of the nuclear receptors in mediating the transcriptional response is largely unexplored and poorly understood. This is further complicated by the fact that some nuclear receptor domains are intrinsically disordered, thus complicating crystallographic studies.

The major focus of this thesis has been to understand the molecular mechanisms of subtype-selective gene expression by focusing on the peroxisome proliferator-activated receptors (PPARs).

In Chapter 2, we investigate subtype-selective properties of the PPARs by overexpression in fibroblasts using a wide range of genomics techniques. We identify clear subtype-selective differences between PPARα and PPARγ in terms of both gene expression and chromatin occupancy, as well as identify major differences in the ability of the different subtypes to access closed chromatin regions. Using chimeric PPAR fusion proteins, we identify the intrinsically disordered regions, the A/B-domain and the hinge-region, as key determinants of PPAR subtype-selective transactivation properties.

In Chapter 3, we investigate the contribution of PPARα to the brown adipocyte phenotype especially in the response to β-adrenergic signalling using genome-wide sequencing techniques. We find that PPARα is dispensable for adipogenesis and lipid accumulation, but is important for the transcriptional changes induced by β-adrenergic signalling. Together, these results place PPARα as a central mediator of key transcriptional responses in mature brown adipocytes *in vitro*.

In Chapter 4, we investigate the 3D chromatin organisation in differentiating adipocytes using promoter capture Hi-C in combination with other genome-wide technologies. We find that promoter-anchored interactions change rapidly in response to differentiation, and that interaction points in many cases are putative enhancers.

Together, these results help describe the highly intricate and cooperative efforts that together regulate transcriptional processes whether focusing on specific factors (PPARα-PPARγ, Chapter 2), on a specific stimuli (β-adrenergic signalling, Chapter 3), or on promoter-anchored interactions (PCHi-C, Chapter 4).