Abstract

Transcription has turned out to be a discontinuous process when imaged at a single cell level. This observation is referred to as transcriptional bursting or pulsing and has been detected in a variety of organisms ranging from bacteria to mammalian cells. The dynamics of transcriptional pulsing are influenced by the properties intrinsic to the transcriptional process, as well as by upstream factors: chromatin environment, signalling molecules, cell cycle stage etc. In the first part of this thesis, we focused on the regulation of transcriptional pulsing in the nucleolus. Using imaging of living cells, we detected pulsatile transcription of a transgene with nucleolar localization whose expression was mediated by RNA polymerase II. In the second part of the thesis, we investigated the relationship between chromatin decondensation and transcriptional dynamics. We used hyperosmotic medium to induce global condensation of chromatin and revealed that upon chromatin decondensation, a transient spike in transcriptional intensity occurs in induvial living cells. Next, we analysed expression of TFRC and POLR2A genes in several cell cycle stages using single molecule RNA FISH. We detected increase in both frequency and size of transcriptional pulses during a limited time window which coincided with chromatin decondensation during telophase/early G1. This upregulation of transcription was controlled by processes distinct from the interphase. We suggest that transcriptional output is largely affected by chromatin decondensation through specific and unspecific mechanisms.

Key words: cell nucleus, nucleolus, transcription, transcriptional pulsing, microscopy, chromatin, chromatin decondensation, mitosis