OVERVIEW

DNA topoisomerases have a dual relationship with the genome. They are essential to solve supercoiling and other topological problems inherent to all DNA transactions, but their intrinsic mechanism of action can result in the formation of DNA breaks, either accidentally during normal cellular metabolism or upon chemotherapy treatment with the so-called topoisomerase poisons. Imbalances in DNA topoisomerase activity can therefore compromise cell survival and genome integrity, entailing serious consequences for human health, such as developmental and degenerative problems and, very importantly, neoplastic transformation processes and their subsequent response to treatment.

We are interested in understanding how DNA topoisomerase activity is regulated to integrate different aspects of genome dynamics, how an imbalance in these processes can lead to the appearance of pathological DNA breaks, and how cells specifically respond to these lesions to maintain genome stability.

“We have uncovered a novel mechanism of transcriptional regulation that allows quick changes in gene expression and has implications in the control of cellular proliferation and cancer progression.”
Accumulation of topological stress in the form of DNA supercoiling is inherent to the advance of RNA polymerase II (Pol II) and needs to be resolved by DNA topoisomerases to sustain productive transcriptional elongation, and therefore the correct expression of genes. Topoisomerases are therefore traditionally considered general positive facilitators of gene expression, especially for long genes in which the load of DNA supercoiling can become particularly burdensome. However, work in our laboratory and others has shown that topoisomerases accumulate at genomic regulatory regions such as enhancers and promoters, suggesting a potential regulatory function for DNA topoisomerases, beyond being mere topological “problem-solvers”.

We unexpectedly found that catalytically inhibiting one of the main cellular DNA topoisomerases (TOP2A) caused a dramatic and acute upregulation of immediate early response genes (FIGURE 1). These genes (IEGs) are normally characterised by quickly responding to different types of cellular stimuli and triggering the subsequent transcriptional waves that control important functions such as neuronal activation or cellular proliferation. Interestingly, the response observed was directly caused by the absence of TOP2A function and not by a response to some kind of topological stress or DNA damage being generated.

Intrigued by these surprising results, pointing to repressive regulatory roles of TOP2A, we developed novel methods to measure topoisomerase activity at specific genomic locations in cells, and combined them with the analysis of supercoiling and genome-wide transcription in cell lines deficient for different DNA topoisomerases engineered with CRISPR-Cas9 technology.

The results obtained allowed us to propose a novel model for transcription regulation based on the control of DNA supercoiling at promoter regions (FIGURE 2). By removing transcription-associated negative supercoiling from promoters, TOP2A ensures the resetting of the topological status after each transcriptional cycle, so transcription occurs in a regulated and controlled manner. When TOP2A activity is limiting or overwhelmed, the accumulated negative supercoiling at the promoters facilitates transcription of subsequent cycles, in a positive feedback loop that results in the typical transcriptional bursts that characterise IEG expression.

These results open up the possibility of modulating topoisomerase activity and DNA supercoiling to regulate IEG expression, and therefore to control cellular proliferation and responses to different types of stimuli, and will need to be taken into account in chemotherapeutic regimens that currently use topoisomerase inhibitors.