Pol I gets repressed

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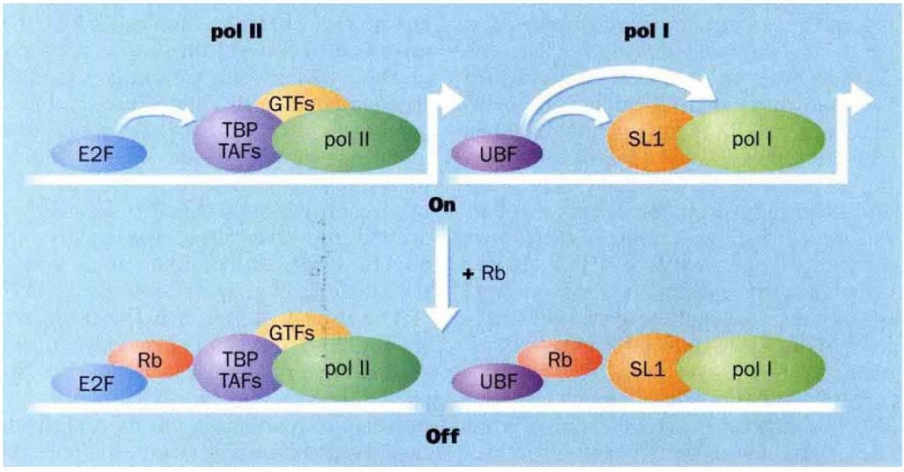
A KEY player in the study of the regulation of transcription during the cell cycle has been the retinoblastoma gene product Rb, the first tumour suppressor to be identified. The prevailing wisdom has been that Rb suppresses cell growth by preventing the expression of those genes, transcribed by RNA polymerase II (pol II), that are necessary for proliferation. The work of Cavanaugh *et al.*, described on page 177 of this issue[1], now suggests an additional mechanism for Rb-mediated growth suppression, in which Rb inhibits activation of transcription by RNA polymerase I (pol I) as well.



Models for Rb repression of transcription from promoters for RNA polymerases II and I, illustrating similarities of mechanism. Rb can repress pol II transactivation mediated by E2F (curved arrow) or other upstream activators that interact with the basal transcriptional machinery, including TFIID (composed of TBP and TAFs) or other general transcription factors (GTFs). Repression of UBF-stimulated transcription from the pol I promoter by Rb requires binding to UBF, which may block interactions between this activator and SL1 or pol I, or both. In both cases, the end result is transcriptional repression.

Previous studies examining the mechanism of growth suppression have focused on the ability of Rb to repress transactivation by the transcription factor E2F, a factor implicated in the expression of genes required for entry into the DNA-synthesis (S) phase of the cell cycle (reviewed in ref. 2). This repression is abrogated by phosphorylation of Rb, which renders the protein incapable of associating with important targets. There are many cellular proteins apart from E2F that can bind to unphosphorylated or hypophosphorylated Rb, and most of them are sequence-specific transcription factors that are associated with a proliferative signal.

It might be expected that a global repression of genes would be necessary to arrest cell growth, and the work of Cavanaugh *et al.* suggests another mechanism for Rb repression which could readily effect such global changes. As growing cells depend on the steady synthesis of ribosomal RNA by pol I, shutting down the synthesis of these genes could be a most effective way for Rb to inhibit cell proliferation.

At least three components are necessary for maximal transcription of rDNA genes by pol I, including the promoter selectivity factor SL1, the upstream-binding factor UBF, and the polymerase itself[3–5]. UBF binds to the pol I promoter and, once bound, can interact directly with the polymerase[6] and probably with SL1 as well. Cavanaugh *et al.* show that Rb specifically blocks activation of pol I transcription by UBF and that this inhibition requires an interaction (presumably direct) between UBF and Rb. The authors demonstrate this association in two ways *in vitro* and furthermore show that the UBF–Rb complex is also present in cell extracts.

These results are satisfyingly consistent with previous findings[7], in which a cDNA encoding UBF was isolated on the basis of an expression library screen using purified Rb protein as probe. Moreover, the biochemical data of Cavanaugh *et al.* are supported by studies at the cellular level: in these experiments, the monocyte-like cell line, U937, can be induced to differentiate by the phorbol ester TPA, which causes an accumulation of Rb in nucleoli, the site of rDNA transcription. Concomitant with this accumulation, there was a marked decrease in rRNA synthesis, while the amount of Rb associated with UBF increased.

The new work points to further experiments that should extend our understanding of the threads that tie together transcriptional activation of different polymerases, and provide further insight into how repressors such as Rb function (see figure). Interesting analogies already spring to mind between the ways in which Rb might repress transcription by pol I and pol II. For example, stimulation of pol II promoters by upstream activators, such as E2F, may be blocked by preventing this factor from interacting with components of the basal transcription machinery, such as the TATA-box-binding protein (TBP) and TBP-associated factors (TAFs). Similarly, association of Rb with UBF may block the effect of this activator on pol I or SL1, itself composed of TBP and pol-I-specific TAFs[8].

The results of Cavanaugh *et al.* open many avenues for exploration. For example, we should find out whether repression of pol I transcription by Rb is a result of an inability of UBF–Rb complexes to bind DNA or of interference with interactions between UBF and pol I (and possibly SL1). A mutational analysis will be required to determine whether the association between UBF and Rb is mediated through a region of UBF overlapping the transcriptional activation sequence, or through the segment in the third HMG-box repeat containing an LxCxE motif, a sequence found in several viral and cellular proteins that interact with Rb. Also, the effect of phosphorylation of Rb on the repression of UBF can be readily tested. It will be interesting to see if Rb can mediate repression of transcription by RNA polymerase III as well, especially in light of the finding that UBF can associate not only with pol I but also with a subunit of yeast pol III (ref. 6).

Despite the pace of progress over the past five years, a number of questions remain regarding Rb function. Many of these can be addressed using rigorous biochemistry now that several important transcriptional targets of Rb have been identified. Finally, another finding of Cavanaugh *et al.* that merits further investigation is the possible role of Rb in the differentiation of haematopoietic cells. □

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