

Controlling gene expression in bacteria

Gene expression enables cells to implement the instructions encoded within the genes on their DNA. Being able to control gene expression is therefore key to this most fundamental biological process, so determining cell fate and behaviour. Cells can fine-tune this process allowing an optimal response to internal and external requirements. Fine-tuning, or gene regulation, mostly occurs at initiation of transcription, the first step of gene expression.

Our laboratory studies transcription initiation using the gut bacterium *Escherichia coli*. Rapid growth rates and the availability of extensive molecular tools and genetic information make this organism an ideal model for studying basic cell biology at the molecular level.

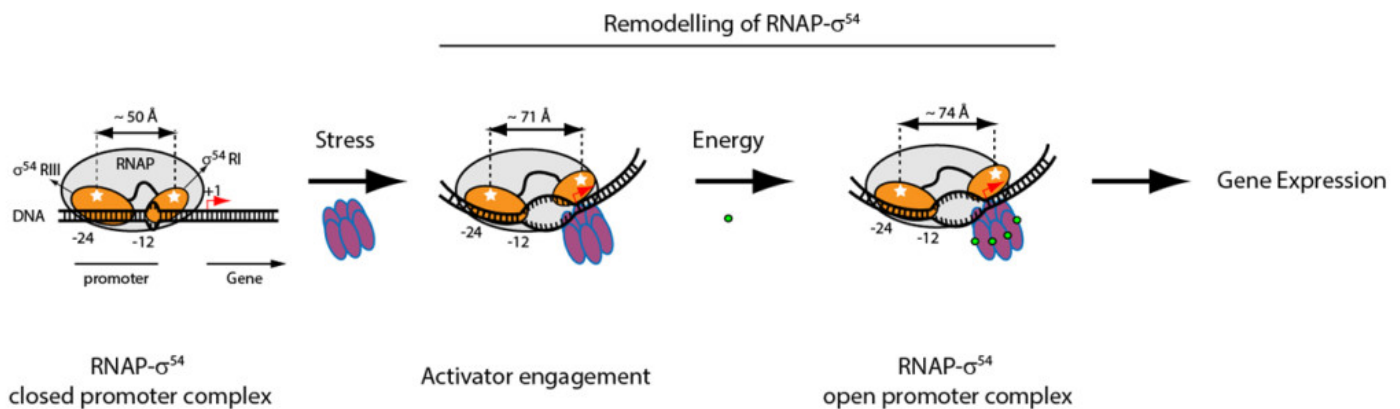


Fig. 1. Regulation of σ^{54} -dependent genes. The σ^{54} factor enables RNAP to interact with the promoter upstream of genes whose expression are required under specific stress conditions. Initially, RNAP and σ^{54} form a transcriptionally inactive closed promoter complex. Here, Region I of σ^{54} (σ^{54} RI) blocks RNAP from accessing the DNA. Upon cell stress, an activator protein remodels the RNAP- σ^{54} closed promoter complex in an energy-consuming process thereby opening up the promoter, removing σ^{54} RI and allowing RNAP access to the DNA for gene expression to begin. Remodelling is indicated by changes in distance between the subunits of σ^{54} Region I (RI) and III (RIII); Å: unit of length, 10 Å = 1 nm. RNAP: grey; σ^{54} : orange; activator: purple; -24 / -12: promoter; start site of gene expression: red arrow.

During transcription initiation in bacteria, RNA polymerase (RNAP, the enzyme that carries out transcription) is directed to the regulatory DNA region of a gene (promoter) via a specificity (sigma, σ) factor. The majority of bacteria contain 2 differing classes of sigma factors: i) σ^{70} directs RNAP to promoters of house-keeping genes whose expression is required for normal cell function; ii) σ^{54} directs RNAP to promoters of genes whose products are involved in specific responses to cell stress including processes which can be exploited for biotechnological applications.

Initially, RNAP and σ form a transcriptionally inactive closed complex (Fig. 1.) at the promoter upstream of the gene. Subsequent opening up of the promoter DNA allows RNAP to access the DNA for transcription and thus gene expression to begin. For expression of σ^{70} -dependent genes this process of open promoter complex formation occurs spontaneously. In contrast, σ^{54} acts as a physical road block preventing RNAP from accessing the DNA thereby inhibiting gene expression in absence of cell stress. For gene expression to occur, the RNAP- σ^{54} complex needs to be remodelled in an energy-consuming process via activator proteins (Fig. 1.) removing the sub-unit of σ^{54} (Region I) which prevents RNAP from accessing the DNA. This additional factor-dependent step in gene expression enables tighter control of σ^{54} -dependent than σ^{70} -dependent genes and is only active under specific cell stress conditions.

Activator-dependency can be bypassed when using a σ^{54} variant lacking Region I in transcription initiation experiments outside the cell with purified components. In our current study we investigated whether such activator bypass transcription events can also be observed in living cells. Activator bypass could be a source of new control networks emerging during evolution to regulate gene expression and thus create novel ways of how cells adapt to their environment.

Our work establishes for the first time that i) in the cell only low level activator bypass is observable arguing that it is strongly constrained; ii) this constriction appears to arise from the structure of DNA at the promoter and iii) σ^{54} can not only act as initiator of σ^{54} -dependent but also as repressor of σ^{70} -dependent gene expression. Our findings therefore suggest that in the cell loss of σ^{54} rather than the creation of activator bypass variants could yield new gene expression patterns during evolution.

Publication

[Genome wide interactions of wild-type and activator bypass forms of \$\sigma^{54}\$.](#)

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