A transcription factor from the cryptic *Escherichia coli* Rac prophage controls both phage and host operons

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Background

Prophage-derived transcription factors (TF) play a crucial role in bacterial gene regulation and stress response, often influencing cellular physiology and survival. The cryptic prophage Rac in *Escherichia coli* harbors a cluster of genes, including *ydaS* and *ydaT*, which are co-transcribed from a shared promoter and are tightly regulated due to their toxic potential. While previous studies have expolred the repressive role of RacR on *ydaS* expression, however, the broader regulatory network involving YdaT, a putative transcription factor, remains poorly defined. Understanding how YdaT modulates gene expression and interacts with other regulatory elements is essential for decoding prophage-host interactions.

Objective

This study aims to elucidate the regulatory network coordinated by YdaT in *E. coli*, focusing on its DNA-binding specificity, gene targets, mode of operon control, and the physiological impact.

Results

First, the authors demonstrated that *ydaS* and *ydaT* are co-transcribed from a single strong promoter, P_{ydaST}. To identify the potential repressors responsible for this promoter, they detect P_{ydaST} activity by reporter assay. They identified that TFs RacR and YdaS together with P_{racR4} showed inhibition to *ydaT* expression. Biochemical assays further revealed that YdaT binds DNA at two sites: the intergenic region containing P_{ydaST} and a distinct YdaT-box overlapping the YdaT start codon. These interactions repress P_{ydaST} activity, indicating an autoregulation that limits YdaT expression. In addition to RacR, which strongly represses P_{ydaST}, the host factor OxyR was also found to influence *ydaT* transcription. Beyond autoregulation, YdaT was shown to activate the host gene *rcsA*, linking prophage regulation to changes in biofilm formation.

Conclusion

n summary, this study demonstrates that YdaT is a prophage-encoded transcription factor whose expression is tightly regulated to avoid toxicity, primarily through repression by RacR and its own autoregulatory binding to the P_{ydaST} promoter and YdaT-box. Although normally silent, once YdaT expressed, it not only limits its own transcription but also alters host physiology by activating *rcsA* and influencing biofilm formation. These findings provide mechanistic insights into the hidden regulatory potential of cryptic prophages in bacterial genomes.