Title:	Molecular and Functional Insights into the Regulation of d-Galactonate Metabolism by the Transcriptional Regulator DgoR in Escherichia coli
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Keywords:	GntR family Carbohydrate metabolism Dgo operon
Issue Date:	2019
Publisher:	American Society for Microbiology
Citation:	Journal of Bacteriology, 201(4).
Abstract:	d-Galactonate, an aldonic sugar acid, is used as a carbon source by Escherichia coli, and the structural dgo genes involved in its metabolism have previously been investigated. Here, using genetic, biochemical and bioinformatics approaches, we present the first detailed molecular and functional insights into the regulation of d-galactonate metabolism in E. coli K-12 by the transcriptional regulator DgoR. We found that dgoR deletion accelerates the growth of E. coli in d-galactonate concomitant with the strong constitutive expression of dgo genes. In the dgo locus, sequence upstream of dgoR alone harbors the d-galactonate-inducible promoter that likely drives the expression of all dgo genes. DgoR exerts repression on the dgo operon by binding two inverted repeats overlapping the dgo promoter. Binding of d-galactonate induces a conformational change in DgoR to derepress the dgo operon. The findings from our work firmly place DgoR in the GntR family of transcriptional regulators: DgoR binds an operator sequence [5′-TTGTA(G/C)TACA(A/T)-3′] matching the signature of GntR family members that recognize inverted repeats [5′-(N)yGT(N)xAC(N)y-3′, where x and y indicate the number of nucleotides, which varies], and it shares critical protein-DNA contacts. We also identified features in DgoR that are otherwise less conserved in the GntR family. Recently, missense mutations in dgoR were recovered in a natural E. coli isolate adapted to the mammalian gut. Our results show these mutants to be DNA binding defective, emphasizing that mutations in the dgo-regulatory elements are selected in the host to allow simultaneous induction of dgo genes. The present study sets the basis to explore the regulation of dgo genes in additional enterobacterial strains where they have been implicated in host-bacterium interactions.
URI:	https://jb.asm.org/content/201/4/e00281-18 (https://jb.asm.org/content/201/4/e00281-18) http://hdl.handle.net/123456789/2236 (http://hdl.handle.net/123456789/2236)
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