ABSTRACT

D-galactonate, a widely prevalent hexonate sugar acid, is used as a carbon and energy source by Escherichia coli. Although the structural dgo genes involved in the transport and metabolism of D-galactonate have been investigated, there is limited information of its regulatory aspect. Using various genetic, biochemical and bioinformatics approaches, we investigated the regulation of D-galactonate metabolism in E. coli K-12. In this work, we have presented the first molecular and functional insights into the regulation of D- galactonate metabolism by the transcriptional regulator, DgoR. We find that the dgo operon is transcriptionally repressed by DgoR and induced specifically in the presence of D-galactonate. Deletion of dgoR accelerates the growth of E. coli in medium containing D-galactonate as a carbon source concomitant with the strong constitutive expression of dgo genes. DgoR exerts a strong repression over dgo genes by binding two closely spaced inverted repeats overlapping the putative D-galactonate responsive dgo promoter. D- galactonate itself rather than any of its metabolic intermediates acts as the true effector to relieve the DNA bound by DgoR. Multiple findings from our work firmly place DgoR in the FadR subfamily within the GntR family of transcriptional regulators: DgoR is a majorly α-helical protein with GntR-type N-terminal wHTH domain and a predicted FadR-like all helical C-terminal FCD domain, binds [5'-TTGTA(G/C)TACA(A/T)-3'] operator sequence matching the signature of GntR family members that recognize inverted repeats [5'-(N)yGT(N)xAC(N)y-3'], and shares critical protein-DNA contacts conserved in the GntR family. Additionally, we identified features in DgoR that are otherwise less common in the regulators of GntR family. Multiple reports from the last couple of decades have implicated the physiological significance of D-galactonate metabolic pathway in the interaction of enteric bacteria with their host. Importantly, in a recent in vivo evolutionary study, E. coli adapted to the mammalian gut was found to accumulate multiple missense mutations in dgoR. 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. Considering that DgoR and its binding sites have been predicted in several enterobacterial strains, the present study sets the basis to explore the regulation of D-galactonate metabolic pathway in these strains and its possible role in mediating host-bacterial interactions.