The PurR regulon in Lactococcus lactis – transcriptional regulation of the purine nucleotide metabolism and translational machinery

Purine nucleotides are either synthesized de novo from 5-phosphoribosyl-1-pyrophosphate (PRPP) or salvaged from the environment. In Lactococcus lactis, transcription of the de novo synthesis operons, purCSQLF and purDEK, has genetically been shown to be activated by the PurR protein when bound to a conserved PurBox motif present on the DNA at a fixed distance from the promoter -10 element. PurR contains a PRPP-binding site, and activation occurs when the intracellular PRPP pool is high as a consequence of low exogenous purine nucleotide pools. By an iterative approach of bioinformatics searches and motif optimization, 21 PurR-regulated genes were identified and used in a redefinition of the PurBox consensus sequence. In the process a new motif, the double-PurBox, which is present in a number of promoters and contains two partly overlapping PurBox motifs, was established. Transcriptional fusions were used to analyse wild-type promoters and promoters with inactivating PurBox mutations to confirm the relevance of the PurBox motifs as PurR-binding sites. The promoters of several operons were shown to be devoid of any -35 sequence, and found to be completely dependent on PurR-mediated activation. This suggests that binding of the PurR protein to the PurBox takes over the role of the -35 sequence. The study has expanded the PurR regulon to include promoters in nucleotide metabolism, C(1) compound metabolism, phosphonate transport, pyrophosphatase activity, (p)ppGpp metabolism, and translation-related functions. Of special interest is the presence of PurBox motifs in rrn promoters, suggesting a novel connection between nucleotide availability and the translational machinery.