Characterization of Endogenous and Reduced Promoters for Oxygen-Limited Processes Using Escherichia coli.

Lara AR, Jaén KE, Sigala JC, Mühlmann M, Regestein L, Büchs J (2017) Characterization of Endogenous and Reduced Promoters for Oxygen-Limited Processes Using Escherichia coli. *ACS Synth Biol* 6(2), 344-356.

Abstract

Oxygen limitation can be used as a simple environmental inducer for the expression of target genes. However, there is scarce information on the characteristics of microaerobic promoters potentially useful for cell engineering and synthetic biology applications. Here, we characterized the Vitreoscilla hemoglobin promoter (Pvgb) and a set of microaerobic endogenous promoters in Escherichia coli. Oxygen-limited cultures at different maximum oxygen transfer rates were carried out. The FMN-binding fluorescent protein (FbFP), which is a nonoxygen dependent marker protein, was used as a reporter. Fluorescence and fluorescence emission rates under oxygen-limited conditions were the highest when FbFP was under transcriptional control of PadhE, Ppfl and Pvgb. The lengths of the E. coli endogenous promoters were shortened by 60%, maintaining their key regulatory elements. This resulted in improved promoter activity in most cases, particularly for PadhE, Ppfl and PnarK. Selected promoters were also evaluated using an engineered E. coli strain expressing Vitreoscilla hemoglobin (VHb). The presence of the VHb resulted in a better repression using these promoters under aerobic conditions, and increased the

specific growth and fluorescence emission rates under oxygen-limited conditions. These results are useful for the selection of promoters for specific applications and for the design of modified artificial promoters.

Leibniz-HKI-Authors

Lars Regestein

Details

Identifier

doi: 10.1021/acssynbio.6b00233

PMID: 27715021