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Environmental regulation of the *pgaABCD* locus in *Escherichia coli* Nuno Cerca<sup>1</sup>, Joana Azeredo<sup>2</sup>, Rosario Oliveira<sup>2</sup>, Gerald B. Pier<sup>1</sup> and Kimberly K. Jefferson<sup>1</sup>

**Background:** The staphylococci are a major cause of catheter-related infections, due, in part, to their ability to form a biofilm. The major constituent of the biofilm matrix is the polysaccharide PNAG, synthesized by the proteins encoded in the *icaADBC* locus. Recently, it has been demonstrated that the *Escherichia coli* locus *ycdSRQP* (re-named *pgaABCD*) has homology to the staphylococcal *icaADBC* locus, and also encodes proteins that direct the synthesis of a polysaccharide that is biochemically identical to PNAG. Furthermore, PNAG promotes biofilm formation in *E. coli*. The aim of this work was to determine environmental factors that regulate transcription of the *pgaABCD* locus and the production of PNAG.

**Methods:** We cloned the promoter of the locus *pgaABCD* into a green fluorescent protein (GFP) reporter plasmid, and transformed the plasmid into a competent *E. coli* strain. The transformed cells were grown in TSB supplemented with a variety of factors, including glucose, sucrose, mannose, magnesium, manganese and ethanol. The production of GFP was detected with a fluorimeter. Factors that augmented the production of GFP were further tested for their influence on PNAG production and biofilm formation in 18 *E. coli* urinary tract clinical isolates by immunoblot using IgG raised against staphylococcal PNAG and by microtiter well biofilm assay, respectively.

**Results:** Transcription of *pgaABCD* was augmented by glucose, manganese, potassium, and ethanol. PNAG production, as detected by immunoblot, was also increased by the same factors. However, glucose was the only factor to significantly increase biofilm formation.

**Conclusions:** Environmental regulation of transcription of the *E. coli pgaABCD* locus, as determined by reporter gene activation was correlated with the production of PNAG in *E. coli* clinical isolates. However, biofilm formation by *E.coli* may not be entirely dependent on PNAG production.

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