

Poster presentation
Biofilm

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Morphogene BolA: its role in biofilm formation and respiration of *E. coli* K-12 MG1655

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Objectives: One important and clinically relevant example of *E. coli* adaptation through systematized gene expression is their ability to grow in biofilms in different environmental conditions. Stress response genes are induced whenever a cell needs to survive under adverse growth conditions. One of the possible gene having an influence on this process is morphogene BolA. Therefore, our work aims to understand the stress tolerance response of *E. coli* in relation with BolA gene under different environmental conditions.

Methods: RNA Extraction: Bacterial cells were harvested and resuspended into RNAprotect[®] Bacteria Reagent (Qiagen, UK) and RNA was extracted using Qiagen RNeasy[®] kit. RNA quality was assessed by agarose gel electrophoresis.

Real time RT-PCR: Real time RT-PCR was used to examine the expression level of BolA gene in biofilms and planktonic cells. Total RNA was extracted, converted to cDNA that was processed using real time PCR (ABI PRISM 7500, Applied Biosystems). Reactions were performed in a 25 µl reaction volume. BolA specific primers were developed in house for this purpose.

Metabolic activity-respiratory activity: The respiratory activity of the several biological (biofilm suspensions or suspended cultures) samples was evaluated by measuring oxygen uptake rates in a biological oxygen monitor (BOM) in short-term assays. The assays were performed in a Yellow Springs Instrument BOM (Model 53).

Results: The expression level of BolA gene was found to be higher in wild type strain in biofilm mode when compared with planktonic phase. This was quantified during both planktonic growth and in biofilms by real time RT-PCR during different stress conditions. This is the first study involving respirometric analysis which showed that BolA gene might have a major role in respiration of *E. coli* in both biofilm and planktonic phases.

Conclusion: It has been shown that the growth rate of *E. coli* remained the same without BolA gene in planktonic cells and also that BolA mutant cannot grow as cell aggregates like wild type *E. coli* does which suggests the contribution of BolA gene in biofilm formation. A comparison between wild type and BolA mutant strains also clearly established the ability of BolA gene to respond and adapt to several stress conditions. The involvement of BolA gene in *E. coli* respiration was observed by checking its glucose metabolism and oxygen uptake under various environmental stress conditions in both wild type and mutant strains.