F302 Construction of an expression vector for a filamentous fungus *Neurospora crassa*

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Neurospora crassa, the orange bread mold has become a model system for the study of fungal sporulation, more specifically, developmental gene regulation. To facilitate the molecular studies with *N. crassa*, an expression vector plasmid, pBLX-1 was constructed. The features of the plasmid are: 1) the multiple cloning sites, 2) the inducible *qa-2* promoter sequence and the *qa-4* terminator sequence, 3) a partial *his*-3 gene sequence for homologous integration into the *his*-3 locus and 4) pUC19 base. The multiple cloning sites contain unique restriction endonuclease digestion sequences for *Nru*I, *Nsi*I, *Spe*I, *Mlu*I, *Bam*HI, *Apa*I, *Not*I, and *Sma*I.

F303 Cloning and Sequence Analysis of *ppK* Gene Which Is a Polyphosphate Kinase in *Bacillus* sp.(KL1114)

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세균은 polyphosphate 대사에 관여하는 *ppk*와 *ppx* 유전자를 갖는 것으로 알려져 있다. *Bacillus* sp.(KL1114) 균주에서 이들 유전자를 cloning하기 위하여 이미 염기서열이 밝혀진 세들균의 *ppk*유전자에서 공통적으로 보존된 아미노산 서열로부터 degenerated primer를 제작하였다. KL1114의 genomic DNA를 주형으로 PCR을 수행하여 약 1 Kb 크기의 PCR 산물을 얻은 후 pET22b(+) vector에 cloning하였다. T_7 primer를 사용하여 826개의 염기서열을 분석한 결과 *ppk* 유전자임을 밝혔다. BLASTP을 이용한 아미노산 서열 분석결과 *Pseudomonas aeruginosa*(81%), *Acinetobacter sp.*(52%), *Neisseria meningitidis*(49%), *Mycobaterium leprae*(30%), *Synechocystis* sp.(32%), *Campylobacter coli*(29%), *Helicobacter pylori*(24%), *Escherichia coli*(23%), *Salmonella typhimurium*(24%), *Vibrio cholerae*(22%), *Klebsiella aerogenes*(25%), *Cholrobium tepidum*(41%)등의 Ppk 아미노산 서열과 유사도가 확인되었다. 현재 이 1 Kb의 *ppk* 유전자 DNA를 probe로하여 genomic library로부터 *ppk* 및 *ppx* 유전자를 cloning하고 있다.

This study was supported financially by the MOST & KOSEF though the Research Center for Bio-Medicinial Resources(Bio-Med RRC) in Pai-Chai University, Korea(Project number: 1999-08 RRC)