내성을 가지고 있었으며, R-Plasmid는 갖고 있지 않았다. 항생물질 발효시 매 24시간마다 인위적으로 오염을 시켜본 결과 초기 2일내에 오염이 되었을 경우에는 항생물질 생산이 거의 이루어지지 않았으나, 발효 3일 이후 즉 항생물질 생성시기 (idiophase)에는 오염이 되었다 하더라도 항생물질 생성에 크게 영향을 못 미쳤다. 또한 초기 오염억제의 방법으로 낮은 농도의 젠타마이신을 발효 초기에 첨가한 결과 항생물질 발효에는 영향을 주지 않고서도 오염을 억제할 수 있었다.

Molecular Cloning of a CMCase Gene from Alkalophilic sp. and Its Expression in *Escherichia coli*

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For isolation of the CMCase gene of the alkalophilic *Bacillus sp.* strain N-4 to analyze their genetic information for the multicomponents of the cellulase, *Bscherichia coli* K12 and plasmid DNA pBR322 was used as host-vector system.

After the digestion of purified chromosomal DNA and plasmid DNA pBR322 with HindIII, these were ligated. The ligated DND were transformed into *Escherichia coli,* and recombinant plasmid 107 carried the gene coding for CMCase was constructed. The CMCase produced by *Escherichia coli* cells containing plasmid DNA pYBC107 was found in the cells as intracellular enzyme and nearly 60% of the total CMCase activity was localized in cellular fraction. Also, the optimum pH for the reaction of CMCase produced by *Escherichia coli* was appeared at pH 8.0 and the enzyme was stable between pH 7.0 and pH 8.0.

Expression of Tunicamycin Resistance in *Bacillus subtilsls* by Several Transfroming Plasmids

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pSp-Si (1.6 kbp) was originally found in pediococcus halophilus to be a cryptic multicopy-plasmid. Hoping that the plasmid can also replicate in Bacillus subtilis, protoplast transformation of strain 207-25 (recE) was performed using pSP-Sl onto which was added the marker of tmrB8 (on 4.9 kbp EcoRI fragment) or tmrB+ (on 0.9 kbp xbaI fragment) gene. Though the tmrB8 gene can expres tunicamycin-resistance at the single copy state, and the tmrB+ gene exerts the resistance only at the multicopy state, we could not confirm the replication of pSP-Sl (tmrB8) or pSP-Sl (tmrB+) in B. subtilis. During the experiment, however, we unexpectedly found that the circularized 0.9 kbp xgaI fragment (tmrB+) itself, which had no replication origin, could transform strain 207-25 to tunicamycin-resistant by protoplast transformation. Southern hybridization analyses with tmrB+ and other probes revealed the integration of the fragment at a single copy state into a position other than the homologous tmrB gene. This recE independent integration of another tmrB+ gene into the chromosome may contribute to the tunicamycinresistance in the transformants.