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The superoxide dismutase (SOD) gene fragment was amplified by reverse transcription polymerase chain reaction (RT-PCR) using degenerate oligonucleotide primers derived from amino acid sequences conserved in the Fe-SODs. An *N. caninum* cDNA library was screened with the SOD gene fragment as a probe. As a result, a complete gene encoding the Fe-SOD was identified. The gene had an open reading frame of 606 bp and deduced amino acid sequence of 202. Sequence analysis showed that the gene had conserved metal binding residues and conserved amino acid residues which were found in Fe-SODs. Comparison of the deduced amino acid sequence of the enzyme with previously reported Fe-SOD amino acid sequences revealed considerably high homo- logies. The coding region of the *N. caninum* Fe-SOD was cloned and expressed in *E. coli*. The molecular weight of expressed protein was approximately 24 kDa. Staining of native polyacrylamide gel for SOD activity of the expressed protein revealed SOD activity that was inactivated by hydrogen peroxide but not by sodium azide and potassium cyanide. This means that the presence of the recombinant fusion protein is indicative of Fe-SOD.

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Regulation of Histidine Biosynthetic Genes from *Corynebacterium glutamicum*

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The genes of *Corynebacterium glutamicum* involved in histidine biosynthesis were cloned and characterized by complementation of *Escherichia coli* mutants. Complementation and sequence analysis showed the existence of 9 genes which are fragmented in three regions (*hisDCBd*, *hisHAFI*, and *hisEG*). Transcription initiation site of the *C. glutamicum his* genes was determined by primer extension analysis. The start site is 196bp upstream of *hisD* gene. The region corresponding to the untranslated 5′ end of the transcript, named the *his* leader region, displays the typical features of the Gram-positive synthetase gene family, including the terminator and antiterminator. Deletion and mutational analysis of the *his* leader region was performed to identify regions and particular nucleotides important for its function.

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Cloning of the *nahAb,Ac,Ad* Gene Encoding Naphthalene Dioxygenase from *Pseudomonas fluorescens* SMEII

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The *P. fluorescens* SMEII can utilize naphthalene as its sole carbon. Naphthalene dioxygenase (encoded *nahA*), the first step of naphthalene degradation pathway, converts naphthalene to *cis*-dihydrodiol naphthalene. Naphthalene dioxygenase is composed of 4 subunits, reductase (*nahAa*), ferredoxin(*nahAb*), oxygenase large subunit(*nahAc*), and small subunit(*nahAd*) in *Pseudomonas* system. To study the naphthalene dioxygenase, 3kb PCR product was cloned from genomic DNA of *P. fluorescens*. The cloned DNA fragment has restriction sites of *KpnI*, *SalI*, and *PstI*. By means of unidirectional ExoIII deletion and dideoxynucleotide chain termination, the

cloned fragment revealed 3 open reading frames(ORFs); *nahAb*(324bp), *nahAc*(1350bp), and *nahAd*(585bp). Putative amino acid sequences of *nahAb*, *nahAc*, and *nahAd* gene from *P. fluorescens* SMEII show high homology to those from other *Pseudomonas* strains.

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Molecular Cloning of the *nahAa* Gene Encoding Feredoxin Reductase from *Pseudomonas fluorescens* SMEII

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We obtained 4.3kb PCR product from the genomic DNA of *Pseudomonas fluorescens* SMEII, which utilizes naphthalene. This DNA fragment, which carried the *nahAa*, *nahAb*, *nahAc*, and *nahAd* gene for upper naphthalene catabolism, was inserted into pT7Blue(R) vector. This recombinant DNA was subcloned by restriction enzyme *KpnI* to generate 1.8kb DNA fragment (pNA1), which was inserted into pUC19. Restriction endonuclease mapping of 1.8kb insert DNA of the pNA1 was carried out with *EcoRI*, *SphI*, *ApaI*, *AvaI*, and *PstI*. By means of bidirectional subcloning and dideoxynucleotide chain termination, we determined the nucleotide sequence of the *nahAa* gene. The results of sequence analysis, Southern hybridization, and SDS-PAGE showed that the recombinant plasmid pNA1 should be contain the *nahAa* gene.

F315

Control of Motility Development by Early Competence Genes in *Bacillus subtilis*

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Many researchers have studied about the network system that controls spore formation and competence development but the field that controls motility development has received few attentions in connection with these two notable phenomena in *B. subtilis*. So we investigated the effect of mutations of competence-controlling genes on the motility development. Either deletion or over-expression of the *comS* whose gene product antagonizes MecA and thereby enhances competence development didn't affect the expression of *hag-lacZ*, a gene used to monitor the motility development, while the mutation of *codY* which is a known repressor of *srf* promoter that codes *comS* caused a slight increase in *hag-lacZ* expression. These controversing data implicated that the hag expression was not significantly influenced by the amount of ComS itself but rather by some events that were related to the *srf* promoter. Indeed the deletion of early competence genes, the *comQ-X-P-A*, which are known to promote *comS* expression, lowered the level of *hag* expression significantly. The *comA* mutation seemed to be responsible to this defect because its mutation alone lowered the *hag* expression to the same level that was observed when all 4 genes were deleted. On the contrary however, the disruption of *comP*, the sensor histidine kinase of two-component signal transduction systems and phosphorylates its response regulator ComA to ComA-P, resulted in the remarkable increasement of *hag* expression. Likewise, both *phrC* and *spo0K* mutations which are supposed to prevent ComA phosphorylation increased *hag* expression, while the *rapC* mutation which is believed to facilitate ComA phosphorylation lowered the