

F009

Isolation and Molecular-Based Characterization of New Carboxydobacteria Capable of Growing on Low Carbon Monoxide Concentration

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Two isolates grown on low carbon monoxide (CO) concentration (400 ppm CO) were obtained from soil. Two isolates were identified as a *Pandoraea* species and a *Pseudonocardia* species by 16S rDNA analysis which genus had not been previously known to oxidize CO. Cell-free extracts from the two isolates grown on both CO and glucose exhibited CO-DH activity. The sequence of 444 amino acids deduced from nucleotide sequence of a putative *cutA* PCR fragment of the *Pandoraea* sp. showed less than 47% identity with those of all reported *cutA* homologues. The *Pseudonocardia* sp. contained two different types of *cutA* homologues, *cutA1* and *cutA2*. The sequences of 420 and 425 amino acids inferred from the *cutA1* and *cutA2* fragments, respectively, were 82 and 90% identical with those of the *Mesorhizobium loti* and *Mycobacterium tuberculosis* H37Rv *cutA* homologues, respectively. Western blot analyses revealed that CO-DH in the *Pandoraea* sp. had no immunogenic epitopes identical to those of the non-mycobacterial and mycobacterial CO-DHs. The analyses revealed that cell extracts prepared from the *Pseudonocardia* sp. cross-reacted with antisera against both types of CO-DHs.

F010

Complete Genome Structure for the Biosynthesis of Macrolide Antibiotic Dihydrochalcocycin (GERI-155) in *Streptomyces* sp. GERI-155Bharat Prasad Jaishy^{1*}, Si-Kyu Lim², Jae-Kyung Sohng³, Jin-Cheol Yoo⁴, and Doo-Hyun Nam^{1,2}¹College of Pharmacy, Yeungnam University, ²Institute of Biotechnology, Yeungnam University, ³Department of Pharmaceutical Engineering, Sunmoon University, ⁴College of Pharmacy, Chosun University

Dihydrochalcocycin (GERI-155) is a 16-membered polyketide macrolide antibiotic with two sugar moieties at C-5 and C-20 positions of a branched lactone ring and has strong antimicrobial activities against Gram-positive bacteria. It is produced by *Streptomyces* sp. GERI-155 isolated from Korean soil. We cloned and sequenced a 75.5-kb region, including polyketide synthase (PKS) region, deoxysugar biosynthesis genes, resistance gene, and other putative regulatory and transporter genes. The 44-kb of PKS genes, consisting of 7 PKS modules arranged in 5 different PKS gene structures, was assumed to be involved in the biosynthesis of 16-membered aglycon based on its chemical structure and modular organization. All the modules showed high sequence similarity with typical type I PKS genes. Post-PKS modification genes for deoxysugar biosynthesis and glycosyltransferase were also found in upstream and downstream of PKS genes. Some genes involved in antibiotic resistance including rRNA methyltransferase and β -glucosidase were also identified. [Supported by grants from KOSEF]

F011

Confirmation of the Biosynthetic Gene Cluster for Macrolide Antibiotic Dihydrochalcocycin (GERI-155) by Gene Disruption in *Streptomyces* sp.Bharat Prasad Jaishy^{1*}, Si-Kyu Lim², Jae-Kyung Sohng³, and Doo-Hyun Nam^{1,2}¹College of Pharmacy, Yeungnam University, ²Institute of Biotechnology, Yeungnam University, ³Department of Pharmaceutical Engineering, Sunmoon University

In 75.5-kb region of a complete genome structure for the biosynthesis of macrolide antibiotic dihydrochalcocycin (GERI-155), a 16-membered polyketide macrolide antibiotic with two sugar moieties at C-5 and C-20 positions of a branched lactone ring, the 44-kb of polyketide synthase (PKS) gene consisting of 7 PKS modules arranged in 5 different PKS gene structures was found. Based on its chemical structure and modular organization, this PKS region was assumed to be involved in the biosynthesis of 16-membered aglycon. To confirm the putative functionality of PKS genes in the cloned gene cluster, the DH domain in module 3 was disrupted by insertional inactivation. The complete loss of antibiotic production was detected in antimicrobial susceptibility test as well as LC-MS analysis of culture broth extract from the disruptant. These results clearly indicate the PKS region of the cloned gene cluster is indeed involved in the formation of macrolide antibiotic, dihydrochalcocycin (GERI-155).

[Supported by grants from KOSEF]

F012

The Modulatory Role of c-FLIP in Adhesion and Motility of Cancer CellsEunsook Shim*, Hyeon Shim, Jinhee Bae, and Dooil Jeoung
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Cellular FLIP (c-FLIP) is an inhibitor of death receptor-induced apoptosis. We employed RNA interference to assess the role of c-FLIP in cellular motility. siRNA directed against c-FLIP inhibited cell motility and adhesion of both SNU387 and HeLa cells. Down regulation of c-FLIP led to increased phosphorylation of p38 MAPK and Akt, but not that of ERK. Inhibition of p38 MAPK or Akt, but not ERK, resulted in downregulation of c-FLIP, inhibition of adhesion to ECM proteins, and motility implying the modulatory role of c-FLIP in motility. FAK, a focal adhesion kinase, has been known to be an essential for cell adhesion to ECM and its downstream targets include Akt. We checked whether downregulation of c-FLIP affected FAK and it indeed significantly decreased phosphorylation suggesting that the modulatory role of c-FLIP in cell adhesion is mediated through its effect on FAK. We also found that downregulation of c-FLIP had a negative effect on adhesion-dependent activation of FAK. Taken together, inhibition of cell motility through down regulation of c-FLIP was accompanied by activation of Akt, p38 MAPK, and inhibition of FAK.

[Supported by grants from KRF, VRC and KNU]