

F312 Expression and regulation of *koarsR* in *Escherichia coli* - Regulatory gene of the arsenic resistance operon in pMH12 from *Klebsiella oxytoca*, D12

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The arsenic resistance operon which consisted of *arsR*, *arsD*, *arsA*, *arsB*, and *arsC* was identified in plasmid R773 from *E. coli*. Our *koars* operon also consisted of same genes and same order. ArsR in R773 binds its own promoter to repress transcription and inducer-binding to ArsR separated it from promoter to start transcription. *koarsR* was highly homologous with *arsR* at 76% and ArsR protein is well conserved in *koars* operon in pMH12. So we researched the function of *koarsR*. Promoter-excluded *koarsR* was obtained by PCR process and cloned into T-tailing vector at first and it was cloned into the vector containing T7 RNA polymerase promoter and expressed. And promoter-included *koarsR* was cloned into promoterless *cat* gene fusion vector and observed its promoter binding abilities. The result demonstrated that KoarsR is a 13kDa protein and is a trans-acting regulatory protein which controls its own expression.

F314 Characterization of Nickel Resistance from *Hafnia alvei* 5-5

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Hafnia alvei is a new highly nickel-resistant bacterium. It was isolated after enrichment culture selective for *Escherichia coli* type bacteria from a soil-litter mixture underneath the canopy of the nickel-hyperaccumulating tree *Sebertia acuminata* in New Caledonia. Two plasmids were identified, one of the size of pULB113 (about 70kb) and another is very small (about 3kb). Curing mutants which have lost the two big plasmids, have not been obtained when the cells grown in the presence of mitomycin C, novobiocin or rifampicin. Only a few mutants were isolated, which had lost the small plasmid (M 650, 651). To examine the nickel resistance gene is located on plasmids, transformation was performed. The transformant KEY10 was obtained, growth curve was measured in nickel media, and DNA-DNA hybridization was tried. The results showed that nickel resistance gene is located on this plasmid.