

ABSTRAK

Telah dilakukan konstruksi pustaka metagenom dari tanah humus dalam vektor pCC2FOSTM dengan sel inang *E. coli* EPI300-T1^R. DNA metagenom diekstraksi secara langsung dari sampel tanah humus dengan metode lisis berbasis-SDS. Analisis DNA pada setiap tahapan dilakukan dengan elektroforesis agarosa 1%. Konstruksi pustaka metagenom dilakukan sesuai dengan prosedur kit *CopyControlTM Fosmid library production* dengan beberapa tahapan : (1) fragmentasi DNA genom, (2) *end-repair* fragmen DNA, (3) seleksi dan *recovery* fragmen DNA *end-repaired*, (4) ligasi fragmen DNA ke vektor *CopyControl* pCC2FOSTM, (5) *packaging* DNA rekombinan, (6) *plating* dan seleksi pustaka fosmid dengan sel inang *E. coli* EPI300-T1^R. Transforman ditumbuhkan pada LB agar yang mengandung 12,5 µg/ ml khloramfenikol selama semalam. Hasil penelitian menunjukkan bahwa DNA metagenom berhasil diisolasi dari sampel tanah humus dan diklon kedalam sistem fosmid pCC2FOSTM dengan sel inang *E. coli* EPI300-T1^R menghasilkan pustaka metagenom sebanyak 420 klon. Namun demikian, dari pustaka metagenom tersebut belum dapat diketahui ada tidaknya klon yang mengekspresikan enzim-enzim pendegradasi sampah organik. Oleh karena itu, pada penelitian selanjutnya akan dilakukan seleksi untuk enzim-enzim pendegradasi sampah organik, karakterisasi, sequencing dan analisis homologinya dengan data sekuen yang ada di *GeneBank*.

Kata Kunci : Tanah humus, DNA metagenom, pustaka metagenom, pCC2FOSTM,
E. coli EPI300-T1^R

ABSTRACT

The construction of metagenom library has been carried out in the vector pCC2FOSTM/*E. coli* EPI300-T1^R system. Metagenom DNA was extracted directly from humus soil using SDS-based lysis method. The DNA was analyzed by 1% agarose electrophoresis. Metagenom library was constructed in accordance with the procedures of *CopyControlTM Fosmid library production* kit, in several stages: (1) fragmentation of genomic DNA, (2) fragment DNA end-repair, (3) selection and recovery of end-repaired DNA fragments, (4) ligation of fragment DNA into the vector *CopyControl* pCC2FOSTM, (5) recombinant DNA packaging, (6) plating and selection of fosmid library. The transformants were grown overnight on LB agar containing 12.5 µg/ ml chloramphenicol. The results showed that metagenom DNA was isolated from humus soil and cloned into fosmid pCC2FOSTM/*E. coli* EPI300-T1^R system, produced metagenom library of 420 clones. However, it was not yet known whether or not the clones that expressed the organic waste degrading enzymes. Therefore, in further research will be selected for organic waste degrading enzymes, characterization, sequencing, sequence analysis and its homology with the sequences data of *GeneBank*.

Key words : Humus soil, metagenom DNA, genomic library, pCC2FOSTM,
E. coli EPI300-T1^R