ABSTRACT

DNA from a normal Thai person and a β-thalassemia/HbE patient was cloned into pBR322 vector at EcoRI/PstI site. Two approaches were used to enrich for the DNA fragments containing the β-globin sequence before insertion into the vector and transformation into E.coli C_{600}. Size-fractionation approach involved separation and purification of 3.6 and 0.75 kb EcoRI/PstI restricted fragment of DNA from agarose gel using DEAE-cellulose paper. All of 107 and 3000 clones derived from 0.75 and 3.6 kb fractions, respectively, were screened for β-globin sequence using ^{32}P-βPst probe but no β-globin clone was obtained. Hybridization-selection approach involved hybridization of total human DNA fragments with JW102 β-globin cDNA probe before insertion. Among 399 recombinants, four clones suspected to contain β-globin sequence were obtained. Plasmid DNA from these colonies was isolated and characterized. By analysis of the size and nature of restricted fragments, we concluded that those four clones were derived from the β-cDNA probe rather than genomic β-globin gene.

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