

# MOLECULAR MECHANISM OF VARIOUS α-THALASSEMIA SYNDROMES

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## Abstract

α-thalassemia, β-thalassemia, hemoglobin(Hb) E and Hb Constant Spring (Hb CS) are prevalent in Thailand. Different combinations of these genes resulted in various thalassemic syndromes including AE Bart's disease, EF Bart's disease and αβ-thalassemia. These syndromes have been previously characterized by clinic, hematologic and genetic studies. Recently, DNA mapping technique has been used to elucidate the molecular basis of thalassemic disease particularly α-thalassemia.

DNA was extracted from the peripheral blood leucocytes of 54 subjects with different α-thalassemic syndromes (9 cases of AE Bart's disease, 18 cases of CSAE Bart's disease, 7 cases of EF Bart's disease, 2 cases of CSE Bart's disease, 8 cases of αβ-thalassemia and 10 cases of homozygous Hb CS. In addition 38 subjects carried α-thalassemia gene or/and Hb CS gene were also extracted by phenol-chloroform extraction. DNA was digested with restriction enzyme Eco RI and hybridized to α- and ζ-specific probes. DNA mapping by Eco RI enzyme can detect all α-thalassemia with α-gene deletion i.e. α-thalassemia 1 and α-thalassemia 2. Types of α-thalassemia 2 were further investigated by digestion with Bgl II enzyme and hybridized to α-specific probe.

In AE Bart's disease, the results showed that all cases of CSAE Bart's had the genotype of α-thalassemia 1/Hb CS with heterozygous Hb E ($--/\alpha^{CS}\alpha$, $\beta^A/\beta^E$). But among 9 cases with hemoglobin types of A+E+Bart's only 4 cases had the genotype of α-thalassemia 1/α-thalassemia 2 with heterozygous Hb E ($--/-\alpha$, $\beta^A/\beta^E$). The remaining could not be demonstrated α-thalassemia 2 haplotype. These AE Bart's patients may result from the interaction of α-thalassemia 1/nondeletion α-thalassemia with

heterozygous Hb E ($--/\alpha\alpha^{T}$, $\beta^{A}/\beta^{E}$) or α-thalassemia 1/Hb CS with heterozygous Hb E ($--/\alpha^{CS}\alpha$, $\beta^{A}/\beta^{E}$).

In EF Bart's disease, all cases had hemoglobin type of CS+E+F+Bart's. DNA analysis and family studies indicated that only one case had the genotype of α-thalassemia 1/Hb CS with $\beta^{0}$-thalassemia/Hb E ($--/\alpha^{CS}\alpha$, $\beta^{0}/\beta^{E}$). The remaining had the genotype of α-thalassemia 1/Hb CS with homozygous Hb E ($--/\alpha^{CS}\alpha$, $\beta^{E}/\beta^{E}$) which is the same as two cases of patients with CS+E+Bart's.

In homozygous Hb CS, the result confirmed that all cases with the hemoglobin type of CS+A or CS+A+Bart's and the amount of Hb CS more than 4.8 % are homozygous Hb CS ($\alpha^{CS}\alpha/\alpha^{CS}\alpha$) because α-thalassemia gene deletion could not be demonstrated.

In αβ-thalassemia, all 8 cases were heterozygous β-thalassemia (Hb $A_2$ > 3.5 %). DNA mapping demonstrated that only one case had the genotype of α-thalassemia 1 and 5 out of 8 cases had the genotype of α-thalassemia 2. α-globin gene deletion could not be demonstrated in the remaining two cases. However, family studies indicated that they carried Hb CS gene.

DNA mapping and family studied classified 38 subjects carried α-thalassemia genes or/and Hb CS genes into 5 groups i.e. 11 cases of heterozygous α-thalassemia 1, 3 cases of homozygous α-thalassemia 2, 5 cases of double heterozygous α-thalassemia 2 and Hb CS, 7 cases of heterozygous α-thalassemia 2 and 12 cases of heterozygous Hb CS. Without DNA mapping, heterozygous α-thalassemia 1 can not be distinquished from homozygous α-thalassemia 2 and heterozygous α-thalassemia 2 can not be distinquished from the normal individual. Among 15 subjects

carrying α-thalassemia 2, 11 were available to characterize the type of α-thalassemia 2. The result showed that α-thalassemia 2 was the rightward deletion type ($-\alpha^{3.7}/$). This study revealed that deletion of α-globin gene is the major molecular defects in various α-thalassemia syndromes.