Study on Hemoglobinopathies in Hubei Province: Report of Hemoglobin G\textit{Coushatta} Homozygotes and Hemoglobin G\textit{Taipei} Heterozygotes

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Summary: This paper presents the results of a survey of hemoglobinopathies in 1137 persons in Hubei Province. Among them 4 cases of abnormal hemoglobin were found and the frequency was 3.52%. According to the clinical and laboratory data and especially the structural analysis of the hemoglobin variant, 3 cases were proved to be HbE, HbG\textit{Taipei} and HbG\textit{Coushatta} respectively. In the family with HbG\textit{Coushatta} 2 members were found to be homozygotes. HbG\textit{Coushatta} and HbG\textit{Taipei} are described in detail in this report.

Key words: hemoglobinopathy, hemoglobin, homozygote, fingerprinting, chromatography, DABITC/PITC double coupling method, HbG\textit{Coushatta}, HbG\textit{Taipei}.

Between April and December 1980, a survey of hemoglobinopathies was carried out in 1137 persons in Hubei Province. Of these, 959 (84.84%) were pure natives of Hubei (group 1) while 178 (15.65%) were “mixed Hubei natives” (group 2) in which only one of the parents of each individual examined was from Hubei Province. 4 cases in group 1 were found to have abnormal hemoglobin and in 3 of them, structural analysis was performed. It was shown that one had HbE ($\alpha_2\beta_2^{28Glu\rightarrow Ala}$), and the other two carried HbG\textit{Coushatta} ($\alpha_2\beta_2^{22Glu\rightarrow Ala}$) and HbG\textit{Taipei} ($\alpha_2\beta_2^{22Glu\rightarrow Lys}$), respectively. The study on HbG\textit{Coushatta} and HbG\textit{Taipei} is reported in this paper.

HEMOGLOBIN G\textit{Coushatta} ($\alpha_2\beta_2^{22Glu\rightarrow Ala}$)

It was demonstrated that HbG\textit{Coushatta} is a slow-migrating abnormal hemoglobin in electrophoresis. Structural analysis of the hemoglobin revealed that glutamic acid is replaced by alanine at position 22 of the $\beta$-chain. In a family of Huangpi County of Hubei Province, 4 members were proved to have HbG\textit{Coushatta} and 2 of them were homozygotes. The first case of homozygote of HbG\textit{Coushatta} was reported by Beissel in 1979 and the cases presented in this paper come second in the world literature.

1. Pedigree investigation

(1) Cellulose acetate membrane electrophoresis of the hemolysate in borate buffer (pH 8.6):
Electrophoretic analysis of the propositus revealed a slow-migrating abnormal hemoglobin and the same type of hemoglobin variant was also found in the hemolysate of the elder sister, mother and aunt.
of the propositus. No HbA, however, of the mother and aunt was demonstrated (fig. 1).

![Cellulose acetate membrane electrophoresis pattern](image)

Fig. 1. Cellulose acetate membrane electrophoresis pattern of the hemolysate of 4 cases of Hb G Cushatia (pH 8.6 B.B.).

(2) Pedigree pattern (fig. 2).

(3) Clinical and laboratory data (table 1).

(4) Other laboratory findings of the mother of the propositus:

The erythrocyte sickling test was negative. No H inclusion bodies were found. The rate of the formation of Heinz bodies was 22.25% (within normal range).

2. Structural analysis

(1) Dissociation of peptide chains:

The hemoglobin of the propositus was first dissociated in 8 M urea and then subjected to cellulose acetate membrane electrophoresis at pH 8.6, in which a slow-migrating abnormal band of β-chain was identified (fig. 3).

(2) Fingerprinting analysis:

The AE (aminoethylated)-β-normal chain and AE-βG C Cushatia were digested by trypsin, and the products analysed by fingerprinting. It was shown in fig. 4 that βG C Cushatia Tp3 (tryptic peptide 3 of βG C Cushatia chain) migrated at a slower speed toward the anode than did the βA Tp3 in high pressure electrophoresis, while in chromatography, the βG C Cushatia Tp3 moved more rapidly (fig. 4).

(3) Analysis of the amino acid composition of βTp3 segment:

The amino acid composition of the hydrolysate of both βA Tp3 and βG C Cushatia Tp3 segments was analysed, and it was found that in βG C Cushatia Tp3 segment, 1 mol of glutamic acid was replaced by 1 mol of alanine (table 2).

(4) Sequence analysis of amino acids of the βTp3 segment:

The amino acid sequence of the βA Tp3 segment was found to be -Val-Asp-Val-Asp-Glu-Val-Gly-Gly-Glu-Ala-Leu-Gly-Arg-, with two glutamic acid residues at

![Pedigree pattern](image)

Fig. 2. Pedigree pattern of the family with Hb G Cushatia.