The Sirenids seem to show (in various degrees) traces of a tetraploid condition in their chromosome set: indeed, the peculiar karyotype found in the family becomes comparable to that of other families of *Caudata* only on the hypothesis of its derivation from a numerically lower chromosome set by polyploidy (and further chromosome changes, typical of each species and tending to 'diploidize' the newly arisen tetraploid karyotype). Such mechanisms could have been of importance for achieving gene duplication during the evolution of Vertebrate genomes[5]; however, Sirenids might represent the first example of a whole amphibian family karyologically differentiated by polyploidy[2,6].

Riassunto. I tre Sirenidi viventi hanno 46, 52 e 64 cromosomi; il corredo di *S. lacertina* sembra tetraploide ($4n = 52$). Viene fatta l'ipotesi che anche i corredi delle altre due specie derivino per (allo-)poliploidia da corredi a minor numero cromosomico, come si ritrovano nelle famiglie «superiori» dell'Ordine.

A. MORESCALCHI and E. OLMO[7]

Istituto di Istologia ed Embriologia dell'Università, Via Mezzocannone 8, I-80134 Napoli (Italy),
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Hemoglobin J β Baltimore in a Fourth French Canadian Family

Fourteen families with hemoglobin J β Baltimore have been described, 7 of whom are caucasians of British or Dutch extraction, 3 are caucasians of French-Canadian stock, 3 are negroes and 1 is of mixed (caucasian-negro) parentage[1,2]. We report the discovery of the hemoglobin type in a fourth French Canadian family, thereby adding evidence to the concept that the gene for this rare variant occurs only in certain population groups.

Materials and methods. The hemoglobin type of a healthy, 16-year-old white boy came to our attention during hemoglobin electrophoresis while his basic health status was being evaluated in a health maintenance program for low-income families.

The proband was adolescent, of asthenic build, with no skeletal abnormalities. He was 165 cm tall and weighed 67 kg. Heart, lungs and abdominal findings were normal; the spleen and other lymph glands were not palpable. Physical and mental developments were normal. Previous illnesses were unremarkable; he and other members of the family had been treated at the center for frequent bouts of streptococcal pharyngitis.

Hematologic values were normal. White cell and differential counts were within normal limits; platelets and red cells were of normal morphology. The urinalysis was normal. Electrophoresis of blood was a routine procedure in the evaluation of health status of participants in the health maintenance program.

He was a fraternal twin, the 3rd of 6 children (Figure 1). The paternal line was French Canadian, whose antecedents had emigrated from the Montreal area to northern Maine and eventually to New York State. No relationship was known with the 3 other French Canadian families carrying Hb J β Baltimore, all of whom had also moved to northern New England from the Province of Quebec.

A fast-moving band of hemoglobin was found in the proband's hemolysate during electrophoresis on Titan III cellulose acetate plates in a Zip-Zone electrophoresis chamber, with Supre Heme Buffer of pH 8.4 at a constant voltage of 400 V for 15 min at room temperature (Helena Laboratories, Beaumont, Texas). The variant's mobility was similar to that of other Hb J β Baltimore variants identified in this laboratory. Densitometry indicated that the variant comprised approximately half (52%) of the entire hemolysate volume, a proportion similar to that in hemolysates from patients with hemoglobin J Baltimore[2].

Hemolysates from the father and 2 brothers also contained the fast-moving band in the same proportion (Figure 1).

Structural studies were carried out with 15 ml of the proband's toluene-extracted hemolysate[3], as described previously[1]. The variant hemoglobin was separated and eluted from the other hemoglobins by vertical polyacrylamide gel electrophoresis[4,5], and the eluate concentrated to approximately 10 g per 100 ml by vacuum dialysis[6]. Recombinant products obtained by hybridization were observed on cellulose acetate[7,8].

The mutation site was also localized by a method in which the hemoglobin chains in the dialyzed eluate were cleaved with sodium *p*-hydroxy mercury benzoate (HMB)[9] and separated in horizontal starch gel electro-

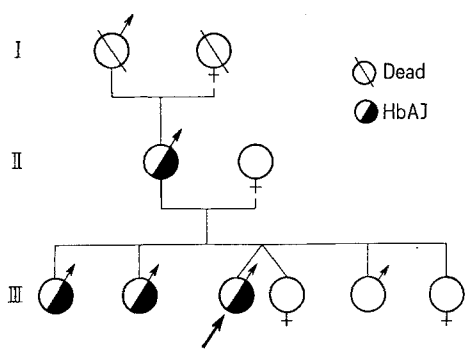

Fig. 1. Pedigree of proband with Hb J β Baltimore.

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phoresis with a gel buffer of *tris*-borate-acid EDTA, 0.036 *M*, pH 8.1 and a bridge buffer of borate, 0.35 *M*, pH 9.0, in a field of 250 V at 0–5 °C for 24 h.

The peptide composition was compared with those of hemoglobin A and hemoglobin J Baltimore by electrophoresis and chromatography[10–12] of trypsin digests[13,10] of the variant's dialyzed eluate.

Results. The mutation site was localized to the β chain by the hybridization experiment, in which bands with mobilities of hybrids with $\beta_2{}^A$-chains were absent from the electrophoretogram, as in canine-variant hybrids of hemoglobin β J Baltimore described previously[1]. The chain electrophoresis data confirmed the interpretation as the mobility of the variant hemoglobin's α-chains was similar to those of Hb A, while its β-chains moved faster than those of Hb A (Figure 2).

The peptide map was that of Hb J β Baltimore[12,14], in that β^A TpII was missing and β^A TpIII was diminished; the identity of the peptides involved was clarified by differential staining for tryptophane and arginine, respectively, in the appropriate positions in peptide maps of hemoglobin A. In addition, the map revealed 2 peptides with mobilities and staining properties similar to the aberrant peptides of Hb J β Baltimore – a neutral one, resembling β^J TpII in position and tryptophane content, and an anodally-migrating peptide, resembling the combination peptide, β^J TpII-III, in position and tryptophane/arginine content.

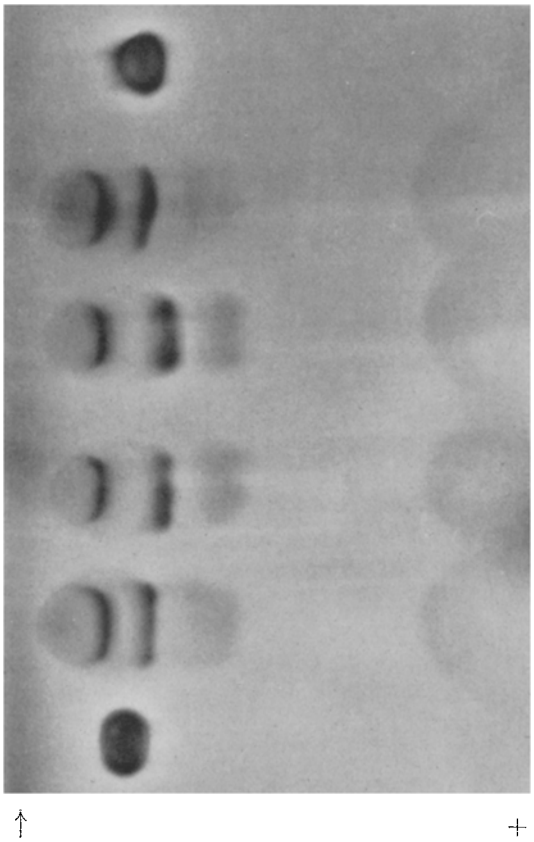

Fig. 2. Electrophoretogram of α- and β-chains of variant hemoglobin and Hb A. Arrow indicates origin; + indicates anode. Reading from top to bottom: line 1, uncleaved variant hemoglobin; lines 2 and 5: α- and β-chains, respectively, of HMB-cleaved Hb A; lines 3 and 4: α- and β-chains, respectively, of HMB-cleaved variant hemoglobin; line 6, uncleaved Hb A.

Molar ratios of glycine and aspartic acid in the combination peptide, B^J TpII-III, were similar to those in the equivalent peptide of hemoglobin J β Baltimore, $\alpha_2{}^A\beta_2{}^{16\,ASP}$, described by Baglioni and Weatherall[14].

Discussion. In the absence of screening data or population studies, reports of individual cases or families with inherited traits are used to illustrate the distribution of rare mutant genes. Thus, our discovery of hemoglobin J β Baltimore in a fourth unrelated French-Canadian family, the 11th caucasian family, in a total of 15 known to carry the trait, suggests a limited distribution to certain population groups. Furthermore, all the caucasian families live in or emigrated from Western Europe's coastal regions-either the British Isles or Holland, including a Danish family, in which the variant is of Dutch origin[2]. The variant has been reported in only 4 other families, 3 of which are of African negro stock, and 1, of mixed negro-caucasian parentage (the variant of which was traced to the caucasian line). Indeed, if the French Canadian is considered Mediterranean (K. Winterhalder, personal communication), rather than coastal West European, then J Baltimore may, in this group, bridge the gene distribution gap between African and West European populations!

The lack of reports of other hemoglobinopathies in French Canadians suggests that J β Baltimore is the structural variant most likely to be found in this population group, as hemoglobins S and C so clearly are in negroes. Conversely, the few reports of J Baltimore in other populations, even those at high risk for hemoglobinopathies and, therefore, likely to be screened electrophoretically, also indicate its rarity and limited distribution.

Résumé. On a découvert l'hémoglobine J Baltimore dans une nouvelle famille canadienne française, ce qui porte à quatre le nombre de familles de cette ethnie, sans lien de parenté, où cette hémoglobine variante est connue. Cette découverte confirme le concept d'une distribution réstreinte au point de vue ethnologique, de cette mutation.

S. Kelly, L. Desjardins, A. Onukagu and L. Peddada

Birth Defects Institute,
New York State Department of Health,
120 New Scotland Avenue,
Albany (New York 12201, USA),
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