Microdrepanocytic Disease in Greece

By C. Choremis and L. Zannos

MICRODREPANOCYTIC disease was first described in Italy by Silvestroni and Bianco in 1945.[1] Since then the disease has been recognized with increasing frequency not only in Italy[2-4] but also in other parts of Europe, South America and the United States.[4, 6,9]

Microdrepanocytic disease—or thalassemia–Hgb S disease in the American literature—is the product of an interaction between two non-allelomorphic genes, i.e., the sickle cell gene and the microcytemia (thalassemia) gene. Being located on different chromosomes, the two genes are transmitted independently of each other. Patients with microdrepanocytic disease may therefore inherit one abnormal gene from each parent or both abnormal genes from one parent,[3] who in the latter case is also suffering from this disease. This possibility is rare, patients with microdrepanocytic disease seldom reaching reproductive age.

This paper deals with the study of microdrepanocytic disease in Greece, and more particularly with its geographical distribution and frequency as compared to the frequency of sickle cell anemia. This is the first report to be published of a large series of Greek patients with microdrepanocytic disease. So far, only sporadic cases have been described in Americans of Greek extraction.

The clinical and routine laboratory findings of our patients have not been included in this study for brevity's sake and in consideration of the fact that—according to our experience, at least—the hematologic picture of microdrepanocytic disease in children is very similar to that of sickle cell anemia. Differential diagnosis between the two diseases seems impossible in the great majority of the cases without examination of the parents. Only in a few patients one may find an unusual splenomegaly or relatively marked bone changes—of the skull particularly (see figure 3 a and b)—which are strongly suggestive of microdrepanocytic disease. The absence of osteoarthritic pains which has been noted in some children is not by itself a valuable criterion in the differential diagnosis as these may also be absent in young patients with sickle cell anemia.

Material and Method

Our material consists of 57 children ranging from 16 months to 15 years, on whom a diagnosis of sickle cell anemia was made on the basis of a positive sickling test, a characteristic clinical picture and the routine laboratory tests. Thirty of them were treated at the Pediatric Clinic of Athens University, the blood of the remaining 27 children and their parents having been sent to us by local physicians from three parts of Greece; namely, Petromagula, Arta and Karditsa.*

The thirty patients from the Athens University Pediatric Clinic included in this study were among many treated for sickle cell anemia during a three year period. The selection of this group was based only on the availability of both parents for study. Otherwise the selection was made at random.

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MAP OF GREECE

The hemoglobin of all 57 children was examined by paper electrophoresis. The instrument used was a German "Elphor" with a veronal buffer of ph 8.6 and ionic strength of 0.05. A current of 1–2 milliampere per paper strip (Whatmen No. 3, diameters: 4 × 28 cm.) at 260–280 volts was delivered for 4–5 hours. The hemoglobin solutions were first prepared in a manner identical to that used for the alkali denaturation technic. A 3 per cent hemoglobin solution was used.

Fetal hemoglobin analysis was done in 19 patients using the alkali denaturation technic as described by Singer.

A sickling test using sodium metabisulfite as reducing agent was performed in all parents. The red cell morphology of the parents with negative sickling tests was examined in dry smears.

Hemoglobin electrophoresis was also carried out in both parents of 30 patients.

Results

Out of the 57 patients examined only 13 were homozygous for the sickling gene, i.e., both parents had a positive sickling test. The electrophoretic behavior of the patients' hemoglobin was that of hemoglobin S.

Determinations of fetal hemoglobin in 7 patients showed values fluctuating between 4.5 and 24 per cent. As shown in figure 1 only one patient had a value as high as 24 per cent which is unusual for sickle anemia (see figure 1).

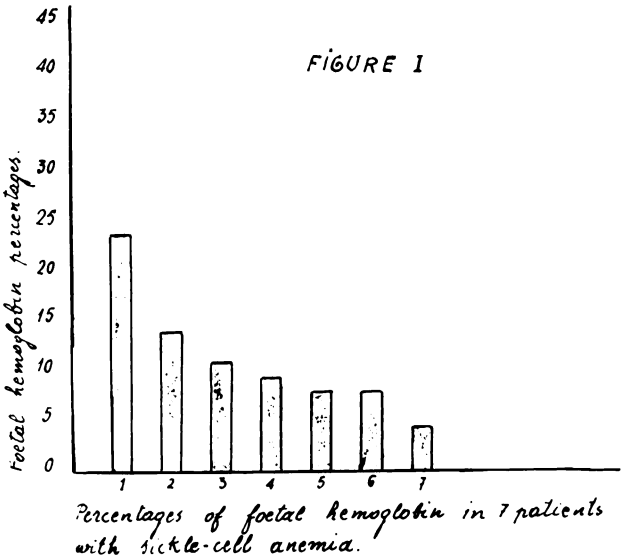
The remaining 44 children proved to be heterozygous for the sickling gene, i.e., the sickling test was positive on one parent, while the other showed the characteristic red cell anomalies of microcytemia (hypochromia, target cells, and a slight anisopoikilocytosis).

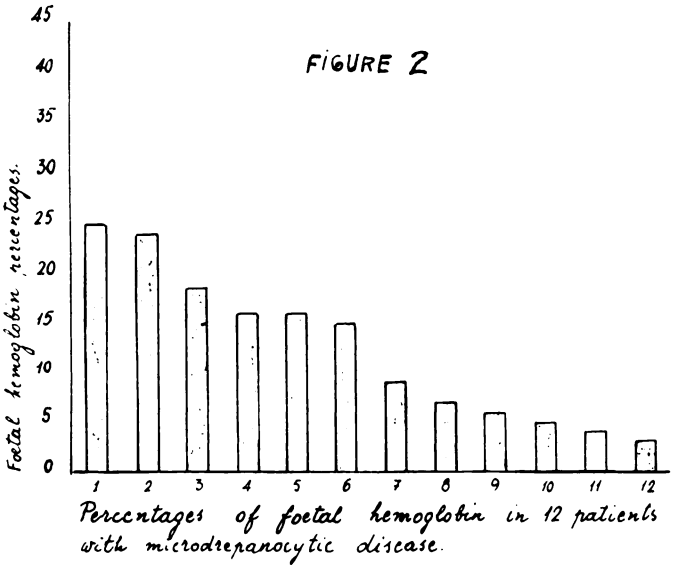
The electrophoretic pattern of the patients' hemoglobin was in most of the cases quite similar to that of the first group. In a few cases the hemoglobin band was slightly wider; however, the difference was not significant enough to serve as a basis for differentiation between the two diseases.

Fetal hemoglobin analysis was performed in 12 patients resulting in values which fluctuated between 3 and 25 per cent. As shown in figure 2 only one patient had a value above 24 per cent.

The electrophoretic pattern of the parents' hemoglobin (20 with the sickle cell trait and 40 with the microcytemia trait) was the expected one. No abnormal pigment other than hemoglobin S could be found.

In conclusion 13 (or 21 per cent) of our patients were suffering from sickle cell anemia and 44 (or 78.9 per cent) from microdrepanocytic disease. These figures probably do not give a true picture of the relative frequencies of the two diseases in Greece as we have not included in this study patients from northern Greece, particularly from the Peninsula of Chalchidiki, where the sickle cell trait has been shown to occur in a very high frequency (up to 32 per cent).[10]





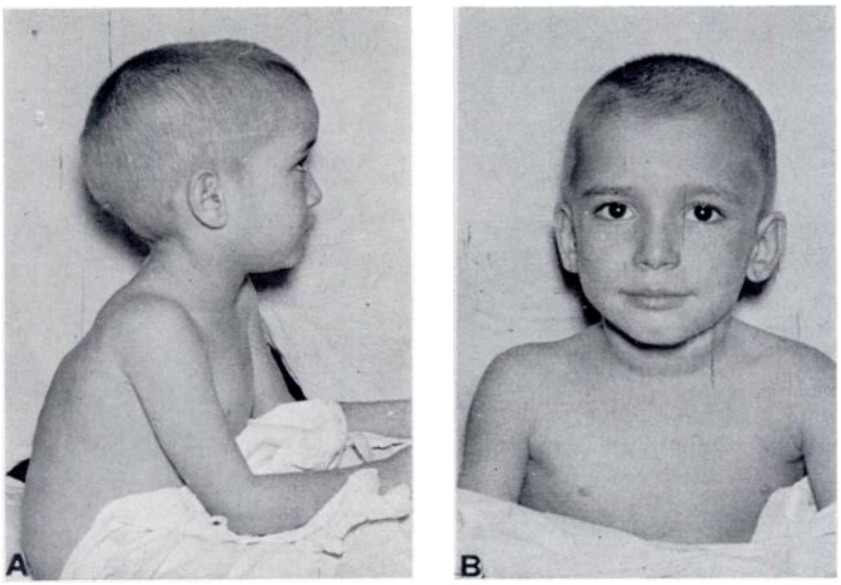


FIG. 3.—T. K., 7 year old male with microdrepanocytic disease. Note marked dolichocephalic skull due to prominence of frontal and occipital bones. A mongoloid facies is absent.

Table 1 shows the geographical distribution of the sickling trait in parents of the 13 patients with sickle cell anemia. The great majority of the parents originated from Petromagula and the neighboring areas (Levadia, Atalanti) where the sickle cell trait is known to occur in a frequency of 15.8 per cent.[11, 12] The relatively frequent mating of two sickle cell trait carriers is, therefore, to be expected.

Table 2 shows the geographical distribution of the sickle cell trait and the thalassemia trait of parents of the 44 children with microdrepanocytic disease. As can be seen both traits are scattered very widely over the country.

TABLE 1.—*Geographical Distribution of the Sickle Cell Trait in the Parents of 13 Patients with Sickle Cell Anemia*

	Number of parents
Petromagula	8
Atalanti	6
Levadia	7
Arta	2
Istiaea (Euboea)	2
Pyrgos	1
Total	26

TABLE 2.—*Geographical Distribution of the Sickle Cell Trait and Microcytemia in the Parents of 44 Children with Microdrepanocytic Disease*

	Number of parents	
	sickle cell trait	microcytemia
Petromagula	2	2
Karditsa	7	7
Sparta	3	4
Arta	13	14
Atalanti	1	1
Levadia	3	3
Gastouni	1	—
Lamia	2	3
Locris	2	2
Limnos	1	2
Gortynia	1	1
Magnesia (Asia Minor)	—	1
Hydra	1	—
Herakleion (Kreta)	—	2
Pholegandros	1	—
Prevesa	1	1
Chalcis (Euboea)	1	1
Mandouthi (Euboea)	2	1
Istiaea (Euboea)	2	—
Total	44	44

DISCUSSION

The results indicate that microdrepanocytic disease in Greece is much more common than sickle cell anemia. This finding is in agreement with the prevailing impression in our country that the frequency of the microcytemia trait is much higher than that of the sickle cell trait. So far, however, studies of the frequency of the microcytemia trait have not been undertaken in Greece. Personally, we base the above impression on the fact that the number of patients with thalassemia admitted to the Pediatric Clinic is much higher (about double) than that of sickle cell anemia and microdrepanocytic disease patients combined. Thalassemia patients in Greece are, as our investigations indicate, true homozygotes for the microcytemia trait. We have examined the blood of 30 patients with thalassemia of the major and intermediate type by paper electro-

phoresis and alkali denaturation, and have found no abnormal hemoglobin—only fetal hemoglobin in high values (unpublished data).

We admit, however, that hospital admissions are not a very reliable criterion in Greece, and thalassemia in reality might not be more frequent than the other anemias. The severe anemia, the mongoloid facies, and the protruding abdomen in thalassemia are symptoms that compel the parents to bring their sick child to Athens even from remote areas of the country. The abdominal and osteoarthritic pains do not frighten the parents and are often misdiagnosed as rheumatic fever and treated by the local doctors.

The present study also shows that the patients with sickle cell anemia originated from areas where focuses of the sickle cell trait are known to have existed for many years. Microdrepanocytic disease on the contrary is much more widely distributed. This observation is also in agreement with an impression of ours based again on hospital admissions; namely, that the microcytemia trait is more scattered over the country than the sickle cell trait. Our thalassemia patients originate from all parts of Greece.* Moreover there are certain islands such as Cyprus, Corfu, Creta and those along the Asia Minor coast where thalassemia appears to be relatively frequent. On the contrary, we have not seen a patient with sickle cell anemia originating from the above areas during the last 6 years (patients with microdrepanocytic disease are also included, as we have been looking for them only during the past 3 years). It is probable, then, that the sickle cell trait is either rare or absent in these islands.

It is true, however, that there is at least one focus of sickle cell anemia, i.e., Petromagula and the neighboring areas, where thalassemia is relatively rare. Chalchidiki, in the north, is probably another one. Nevertheless, compared to the whole of Greece, these areas are small, and we do not think that they can greatly influence the conclusions of our observations just mentioned.

Unfortunately our studies on fetal hemoglobin are limited to a small number of patients and only one conclusion can be drawn from them, namely, that in the individual case, the percentage of fetal hemoglobin is of little value in the differential diagnosis between sickle cell anemia and microdrepanocytic disease. Further studies may prove that high values, above 24 per cent, are more frequently encountered in microdrepanocytic disease. However, very high percentages can be found in sickle cell anemia—up to 40 per cent according to Sturgeon.[13] It could be said, therefore, that great amounts of fetal hemoglobin may prove suggestive but never pathognomonic, of microdrepanocytic disease.

Paper electrophoresis, in our hands at least, has also proven of no value in the differential diagnosis between the two diseases. In a few cases of microdrepanocytic disease the hemoglobin band appeared a little wider, which suggested the presence of normal hemoglobin (S + F + A). But the difference was by no means significant enough to be taken into consideration.

It is unfortunate that more extensive family studies, which would throw some light on the problem of allelism or non-allelism of the sickle and thalassemia genes, were not made possible in this investigation. In any case, among the parents whose hemoglobin was examined electrophoretically, no one was found to have microdrepanocytic disease or thalassemia.

* Because of the long distance, there are, however, relatively few patients from the north of the country who come to Athens.

Summary

Among the 57 patients with the hematologic picture of sickle cell anemia, examination of the parents revealed that 44 were suffering from the microdrepanocytic disease and 13 from sickle cell anemia. The much more frequent occurrence of microdrepanocytic disease was expected since the microcytemia trait appears to be much more frequent in Greece than the sickle cell trait.

Most patients with sickle cell anemia originated from areas where the trait has been found in a very high frequency. In contrast, the geographical distribution of patients with microdrepanocytic disease is much wider.

Summario in Interlingua

Inter 57 patientes con le tableau hematologic de anemia a cellulas falciforme, le examine del parentes revelava que 44 suffreva de morbo microdrepanocytic e 13 de anemia a cellulas falciforme. Le multo plus alte incidentia de morbo microdrepanocytic esseva expectate proque il pare que le tracto de microcytemia es multo plus frequente in Grecia que le tracto de cellulas falciforme.

Le majoritate del patientes con anemia a cellulas falciforme esseva originari de areas ubi le tracto habeva essite trovate con alte grados de frequentia. Del altere latere, le distribution geographic de patientes con morbo microdrepanocytic es multo plus extense.

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