# ERYTHROPHAGOCYTOSIS AND HEMOSIDEROSIS IN THE LIVER AND SPLEEN IN SICKLE CELL DISEASE *

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Abnormal destruction of erythrocytes by the cells of the reticulo-endothelial system is characteristic of many hemolytic anemias. However, these cells seldom exhibit morphologic evidence of erythrophagocytosis. It has been seen occasionally in pernicious anemia, hemolytic jaundice, sickle cell anemia, icterus gravis neonatorum and Weil's disease. The degree to which this erythrophagocytosis is apparent differs in various parts of the reticulo-endothelial system. The spleen, as a rule, shows very little evidence of this process.[1]

The pathologic changes of the different organs in sickle cell anemia have been repeatedly summarized.[2-6] It has been stated that the Kupffer cells in the liver frequently exhibit phagocytized red blood cells.[2,5,7] The changes in the spleen also have been thoroughly investigated.[8] The details, however, as to the frequency and extent of these processes are not yet established.

The case of sickle cell disease herein reported is illustrative of a disproportional degree of erythrophagocytosis in the liver and of hemosiderosis in the spleen. A survey of the occurrence of these processes in a number of cases examined at necropsy, with an attempt to correlate the splenic and hepatic changes, is also included. A similar analysis of the reported cases in the literature with complete necropsy data has also been attempted.

## MATERIAL

From 4,094 postmortem examinations at Charity Hospital of Louisiana from January 1, 1939, to January 1, 1942, 12 cases of sickle cell disease were available for study. Cases with incomplete data were excluded. Data pertaining to the age, sex, size of the spleen and liver, and the degree of erythrophagocytosis and hemosiderosis were tabulated. In addition to the routine hematoxylin and eosin staining, the presence of hemosiderin was tested by the Turnbull-blue reaction.[9]

Since the anemia was not established in all cases, the term "sickle cell disease" will be used, rather than "sickle cell anemia."

## REPORT OF ILLUSTRATIVE CASE

A colored male, 14 years of age (see case no. 3, Table II), was admitted with cough, and edema of the face. He gave a history of having had heart trouble for 1 year. Three years prior to admission he had had rheumatic fever.

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Examination revealed a poorly nourished, but fairly well developed patient with a temperature of 99.6° F.; pulse, 110; respiration, 26; blood pressure, 116/40. The heart was markedly enlarged. A blowing systolic apical murmur was heard. The liver was markedly enlarged. On the anterior aspect of the legs there were numerous small scars present.

*Laboratory Findings.* The blood examination was as follows: Hemoglobin, 5.8 gm. per 100 cc., 40% (Hellige); red blood cells, 1,780,000; white blood cells, 19,000; nucleated red cells, 60,000, and platelets, 500,000 per cmm.; hematocrit, 21.5 (Wintrobe); polymorphonuclears, 48 per cent; eosinophils, 4 per cent; monocytes, 9 per cent; lymphocytes, 25 per cent; promyelocytes, 2 per cent; myelocytes, 9 per cent, and metamyelocytes, 3 per cent. The wet preparation showed 90 per cent of the red blood cells to be sickle-shaped. The fragility of the red cells was normal. A sternal puncture revealed a hyperplastic, pronormoblastic marrow. Clotting time and coagulation time (Lee-White) were within normal limits. The urine showed no deviation from the normal. The icterus index was 66; the van den Bergh test showed 7.6 mg. of bilirubin per 100 cc. (direct method). Four days later the van den Bergh test gave 25 mg. of bilirubin per 100 cc. (direct method). The Kline flocculation test was negative. On several occasions the sputum was negative for acid-fast bacilli. Roentgenograms of the chest showed far advanced bilateral pulmonary tuberculosis and cardiac enlargement. The electrocardiograph showed evidence of myocardial disease typical of rheumatic mitral stenosis.

*Clinical Course.* Patient became progressively more anemic and developed jaundice, dyspnea, and pitting edema of both legs. His temperature became of septic type, and he expired 77 days after admission to the hospital.

## *Necropsy Findings*

There was marked edema of the face and lower extremities. Both legs presented numerous scars on the anterior aspects. The heart weighed 500 gm. and showed marked right ventricular hypertrophy and mitral stenosis. The lungs presented a far advanced exudative tuberculosis with numerous cavities. Enlarged caseous hilar nodes were present. The spleen was small, firm, purplish in color and weighed 24 gm. Its cut surfaces were fibrous. The liver weighed 3600 gm., and the cut surfaces revealed diffusely scattered grayish yellow areas measuring 1 mm. in diameter. The kidneys were markedly enlarged, weighing together 720 gm. The sternum, thoracic vertebrae and the middle portion of the femur presented red, active marrow.

## *Histologic Examination*

A large proportion of the red blood cells were sickle-shaped in all organs. The sinusoids of the liver were markedly distended; the Kupffer cells were enlarged and their cytoplasm contained numerous sickle-shaped red blood cells (Fig. 4). The liver cells were compressed and contained finely granular cytoplasm, with well stained nuclei. Occasional small areas of extramedullary myelopoiesis were seen. Some bile capillaries contained bile plugs. The spleen showed a marked increase in connective tissue. Around the blood vessels large amounts of dark brownish green pigment were deposited, which varied in intensity. Also aggregations of dark blue incrustations were evident. The

lymph follicles were small and without germinal centers. Some sinusoids were distended with sickle-shaped red blood cells. The walls of the sinusoids were thickened and the sinus endothelial cells were conspicuous, occasionally containing phagocytized red blood cells. In addition to the tuberculous lesions, the lungs showed large alveolar phagocytes containing sickle-shaped red blood cells. The kidneys showed hyperemia, interstitial edema and small areas of erythropoiesis. Active pronormoblastic erythropoiesis was seen in the bone marrow. The mesenteric lymph nodes presented small follicles and markedly distended sinusoids, with active erythrophagocytosis by sinusoidal cells. The Turnbull-blue stain was strongly positive in the spleen and slightly positive in the liver, kidneys, bone marrow and mesenteric lymph nodes.

### *Summary of Case*

In the case presented the spleen was small and fibrous with calcium and iron incrustations. The liver was greatly enlarged (Fig. 6) and exhibited a marked degree of erythrophagocytosis. The various portions of the reticulo-endothelial system appeared to be in quite different functional stages. The splenic reticulo-endothelium appeared to be inactive while the Kupffer cells exhibited marked hyperactivity.

## Observations, Including Additional Cases

The 12 cases were divided into two groups according to the size of the spleen. The first group (Table I) included 7 cases in which the spleen was of normal weight or enlarged (average 286.4 gm.). The

Table I

*Cases of Sickle Cell Disease with Spleens of Normal Weight, or Larger*

| Case no. | Age | Sex | Spleen gm. | Liver gm. | Liver-spleen ratio | Erythrophagocytosis | | Hemosiderosis | | Complicating disease |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | Spleen | Liver | Spleen | Liver | |
| 1. W. S. | 28 | M | 140 | 1535 | 10.96 | — | + | — | — | Brain abscess |
| 2. E. H. | 47 | M | 160 | 1720 | 10.75 | — | — | + | + | Acute thyrotoxicosis |
| 3. L. M. | 40 | F | 225 | 2400 | 9.60 | + | — | + | — | Typhoid fever |
| 4. M. S. | 6 | M | 70 | 640 | 9.14 | — | + | + | + | Wilms' tumor |
| 5. A. L. | 49 | M | 150 | 2200 | 14.66 | — | ++ | ++ | — | Type 23 pneumococcus meningitis |
| 6. J. W. | 54 | M | 190 | 1790 | 9.42 | + | ++ | — | — | Hypertension |
| 7. Z. K. | 17 | M | 1070 | 1650 | 1.54 | + | ++ | + | — | Abdominal crisis |
| Average | 34.4 | | 286.4 | 1705 | | | | | | |

livers of this group were of normal weight or moderately enlarged (average 1705 gm.). The average age of the patients in this group was 34.4 years, the ages ranging from 6 to 54 years.

The second group (Table II) consisted of 5 cases in which the spleen was small (average 19.7 gm.). The livers varied from a normal size to markedly enlarged (average 2106 gm.). The average age was 19 years, the ages ranging from 14 to 39 years.

Because of the wide age range in both groups the hepatic-splenic ratio was computed according to the method given by Ahronheim, [10]

TABLE II

*Cases of Sickle Cell Disease with Small Spleens*

| Case no. | Age | Sex | Spleen gm. | Liver gm. | Liver-spleen ratio | Erythrophagocytosis | | Hemosiderosis | | Complicating disease |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | Spleen | Liver | Spleen | Liver | |
| 1. E. L. | 22 | M | 5.4 | 1950 | 361.1 | — | +++ | — | + | Abdominal crisis |
| 2. E. R. | 39 | F | 7.0 | 1250 | 178.6 | — | — | +++ | — | |
| 3. C. O. H. | 14 | M | 24.0 | 3600 | 150.0 | ++ | ++ | +++ | + | Tuberculosis |
| 4. W. M. L. | 18 | F | 50.0 | 3080 | 61.6 | — | + | +++ | — | Postpartum thrombophlebitis |
| 5. F. W. | 2 | F | 12.0 | 650 | 54.2 | + | — | ++ | — | Rheumatic fever |
| Average | 19.0 | | 19.68 | 2106 | | | | | | |

who found that the normal ratio between hepatic and splenic weight is 9.3:1. The ratios for the cases tabulated in Table I were close to or slightly above this value, with two exceptions—cases nos. 5 and 7. Case no. 4, a boy of 6 years, showed absolute values above the normal for his age, [11] but the hepatic-splenic ratio was within normal limits. The cases tabulated in the second group showed very high values for the ratio, indicating a marked diminution of the spleen and enlargement of the liver in all instances.

### *Histopathology of the Liver*

The livers were enlarged, reddish brown and firm. Histologic study revealed marked congestion, the sinusoids being distended and filled with sickle-shaped red blood cells. The Kupffer cells were swollen and exhibited varying degrees of erythrophagocytosis. This was most marked in case no. 1, Table II (Fig. 4). The liver cords were regularly arranged and the parenchymal cells were compressed or slightly swollen and granular. Infrequently lipoidosis in varying degrees was seen.

### *Histopathology of the Enlarged and Normal-Sized Spleens*

The spleens of the first group revealed a marked congestion of the sinusoids and venous sinuses, which were filled with sickle-shaped red blood cells. The lining cells of the sinusoids formed thin, irregular, netlike structures. The central arteries were also markedly dilated and the follicles were without germinal centers. Often there were pools of extravasated blood around the follicles. This was most marked in case no. 7, Table I, in which the spleen weighed 1070 gm. (Fig 5). Varying amounts of iron-containing pigment were found. The reticulum showed in places a somewhat thicker fibrillar network, upon which hemosiderin was deposited in the form of myceliumlike threads (Fig. 1). These changes were often confined to the vicinity of the small blood vessels. Around the periadventitial tissue the reticulum coalesced to form a thick hyaline network, and here the Turnbull-blue stain revealed thick rods of deposited iron (Fig. 2). In irregular areas calcium deposits were also present and small hemorrhages and old infarcts of small size were often seen.

### *Histopathology of the Fibrotic Spleens*

There was an increase of hyaline connective tissue and widening of the trabeculae in the fibrotic spleens. In this connective tissue, irregular areas of dark bluish green or brownish clumps, often arranged in parallel rods resembling bamboo sticks, were formed. In other instances, the deposits formed structureless bluish brown masses (Fig. 3). The walls of the sinusoids were thickened, and their endothelial lining cells were flattened. Distorted red blood cells filled the sinusoids. Erythrophagocytosis by the sinus endothelial cells was minimal. The follicles were inconspicuous and without germinal centers. The iron stain was strongly positive in all but one instance (case no. 1, Table II), in which the spleen was very small and in which the parenchyma was replaced by fibrous connective tissue.

## Review of Cases from the Literature

A large number of cases of sickle cell disease have been reported but complete necropsy data are available in only a limited number. In reviewing the literature, only 13 cases were found in which there were included data pertaining to the age, sex, size of the spleen and liver, and the degree of erythrophagocytosis and hemosiderosis. These cases have been grouped according to the size of the spleens.

The first group consisted of 5 cases with normal or enlarged spleens (Table III). No average can be given since only one adult was included in this group, but the hepatic-splenic weight ratio clearly

indicates splenomegaly. Case no. 1, a colored girl, 4 years of age, was included because the weights of her spleen and liver were within normal limits.[11] The remainder of the cases showed a definite splenomegaly.

The second group consisted of 8 cases with siderofibrotic spleens and correspondingly high values for the hepatic-splenic ratios (Table IV). The age range in this group was from 6 to 38 years, the average age being 20.5 years. The spleens again were markedly atrophic

TABLE III

*Collected Cases of Sickle Cell Disease with Spleens of Normal Weight, or Larger*

| Author and year | Age | Sex | Spleen gm. | Liver gm. | Liver-spleen ratio | Erythrophagocytosis | | Hemosiderosis | | Complicating disease |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | Spleen | Liver | Spleen | Liver | |
| Wollstein and Kreidel,[5] 1928 | 4 | F | 30.0 | 460 | 15.3 | — | ++ | ++ | + | |
| Wollstein and Kreidel,[5] 1928 | 3 | M | 210.0 | 460 | 2.2 | — | ++ | + | + | |
| Wollstein and Kreidel,[5] 1928 | 3 | M | 182.0 | 430 | 2.4 | — | ++ | + | + | |
| Lash,[14] 1934 | 21 | F | 960.0 | 2420 | 2.5 | — | — | — | — | Cervical cesarean section |
| Ryerson and Terplan,[15] 1935 | 4 | F | 425.0 | 500 | 1.2 | ++ | ++ | ++ | — | |

(average 15.4 gm.). The livers were of normal size or enlarged (average 1834 gm.), as indicated by the uniformly high values for the liver-spleen ratios.

## COMMENT

This survey indicates that varying degrees of erythrophagocytosis were present in sickle cell disease. The Kupffer cells of the liver most frequently exhibited engulfed red blood cells, while the splenic reticulum cells only occasionally showed signs of erythrophagocytosis. The changes in the spleen, however, were more varied in nature and more severe than those of the liver. An abnormal distention of the perifollicular sinusoids, hemorrhages and hemosiderosis constituted the different stages of the splenic lesion, producing the siderofibrotic spleen.

In the literature markedly enlarged spleens were reported chiefly in young children (Table III). There was only one adult in this group, a female, 21 years old. Cases with normal spleens, or with

spleens but slightly smaller than normal, were included in Table I. These probably are the less severe forms of sickle cell disease, as death in all instances was due to complicating conditions.

The cases tabulated in Tables II and IV are the severe forms of sickle cell disease with siderofibrotic spleens. Assembled respectively from the records of Charity Hospital of Louisiana and from the reported cases in the literature, these two groups show remarkable similarities.

TABLE IV

*Collected Cases of Sickle Cell Disease with Small Spleens*

| Author and year | Age | Sex | Spleen gm. | Liver gm. | Liver-spleen ratio | Erythrophagocytosis | | Hemosiderosis | | Complicating disease |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | Spleen | Liver | Spleen | Liver | |
| Graham,[16] 1924 | 30 | F | 28.0 | 2567 | 92.0 | — | + | +++ | + | Streptococcus septicemia |
| Sydenstricker,[12] 1924 | 6 | M | 7.9 | 690 | 87.0 | — | — | + | — | |
| Jaffé,[17] 1927 | 8 | M | 10.0 | 900 | 90.0 | — | ++ | ++ | — | Tuberculosis |
| Jaffé,[17] 1927 | 6 | F | 10.0 | 1300 | 130.0 | — | ++ | ++ | + | |
| Ching and Diggs,[18] 1933 | 18 | F | 10.5 | 2210 | 210.0 | — | +++ | — | — | |
| Yater and Hansmann,[19] 1936 | 38 | F | 7.0 | 1715 | 245.0 | — | — | +++ | + | Abdominal crisis |
| Yater and Hansmann,[19] 1936 | 25 | F | 35.0 | 2090 | 59.7 | — | — | +++ | ++ | Abdominal crisis |
| Bauer,[20] 1940 | 33 | F | 15.0 | 3200 | 213.0 | + | + | ++ | — | |
| Average | 20.5 | | 15.4 | 1834 | | | | | | |

From the available clinical data it is difficult to establish the duration of active sickle cell disease. Diggs,[8] who studied thoroughly the changes in the spleen in sickle cell anemia, concluded that there is no direct correlation between the size of the spleen and the duration of the disease as indicated by the age of the patient. A large spleen was usually found in the early phases of the disease, while a small atrophic spleen was characteristic of the later phases. This is, however, not without exception. Therefore, the duration of the disease could be determined more accurately by the histologic changes present.

Cases included in Table I were characterized by normal or enlarged spleens, with histopathologic changes suggestive of shorter duration, and cases in Table II revealed siderofibrotic spleens, indicating longer duration. It is difficult to correlate these changes with the duration of the disease when one considers that the average age of the first group was 34.4 years and of the second, 19.0 years. There exist both active and latent varieties of sickle cell disease,[12] differing in degree rather than in kind. The first group probably represented the latent cases, and the second group the active cases. However, the pathologic changes differed only in degree. The enlarged spleens and the normal-sized spleens presented similar pathologic changes, differing again only in degree. In the active cases the liver-spleen ratios were much lower (Table III) or much higher (Tables II and IV) than normal, while in the latent form (Table I), they were nearly normal.

No direct correlation existed between erythrophagocytosis and complicating infectious disease [13] (Tables I to IV).

## Summary

An example of sickle cell disease is reported in which there was a very marked degree of erythrophagocytosis.

Twelve cases of sickle cell disease were studied from 4,094 autopsies. These were divided into two groups: those with large and normal-sized spleens and normal or slightly enlarged livers; and those with siderofibrotic spleens and enlarged livers. These correlations were definitely demonstrated by the liver-spleen ratios.

Thirteen cases with complete necropsy data were collected from the literature and also tabulated according to the weight of their spleens. Definite splenomegaly was present in 4 cases. There was a normal-sized spleen in one case, and eight cases showed marked siderofibrotic changes with high values for the liver-spleen ratio.

The Kupffer cells were the most actively participating parts of the reticulo-endothelium in erythrophagocytosis. This process is not dependent upon coexisting infection.

The splenic changes may serve as criteria for the degree of activity but not for the duration of the sickle cell disease. The latent cases also present definite changes in the spleen and liver similar to those seen in active cases. The spleen may be enlarged, normal, or markedly fibrosed in sickle cell disease.

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[ *Illustrations follow* ]

## DESCRIPTION OF PLATES

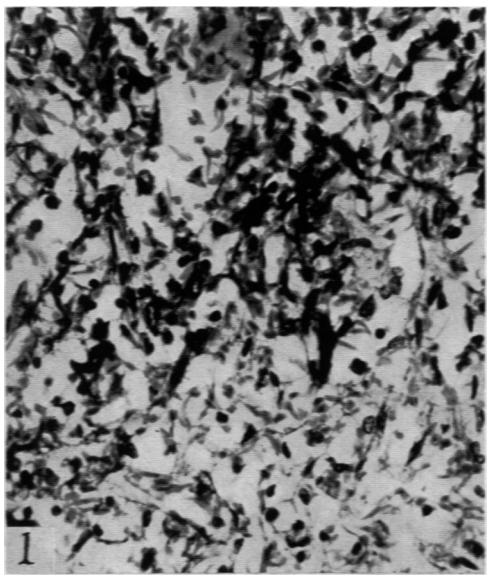
### PLATE 27

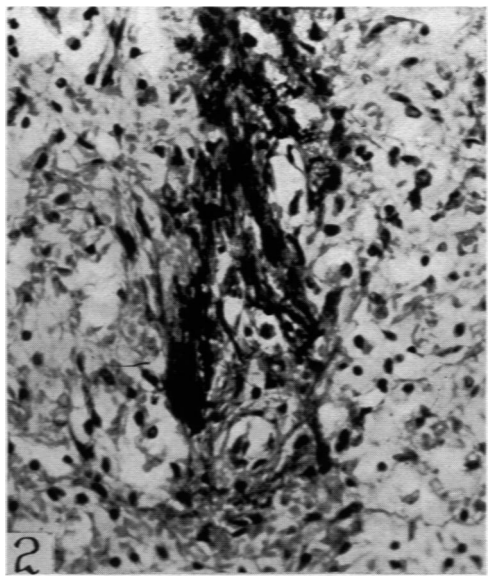
FIG. 1. (Case no. 4, Table II.) Spleen: marked distention of the sinusoids. Hemosiderin has been deposited in myceliumlike fashion. Turnbull-blue stain. × 300.

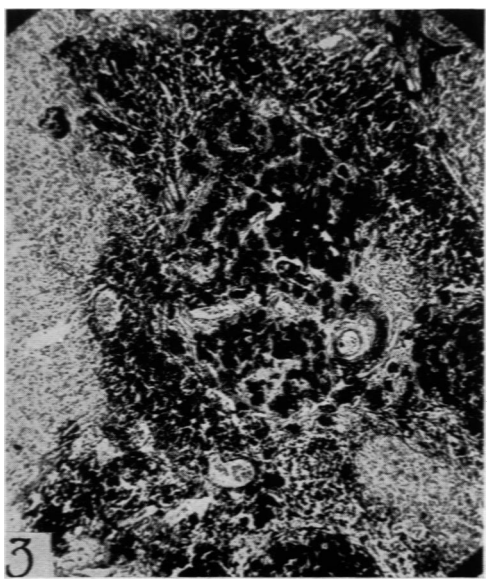
FIG. 2. (Case no 4, Table II.) Spleen: rodlike deposits of hemosiderin. Turnbull-blue stain. × 335.

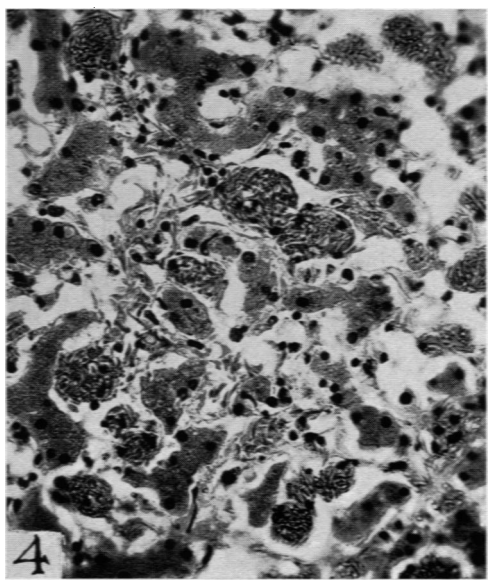
FIG. 3. Case no. 1, Table II.) Spleen: a typical siderofibrotic area with marked hemosiderin and calcium incrustation. Turnbull-blue stain. × 45.

FIG. 4. (Case no. 1, Table II.) Liver: the Kupffer cells show a marked degree of erythrophagocytosis. Hematoxylin and eosin stain. × 335.



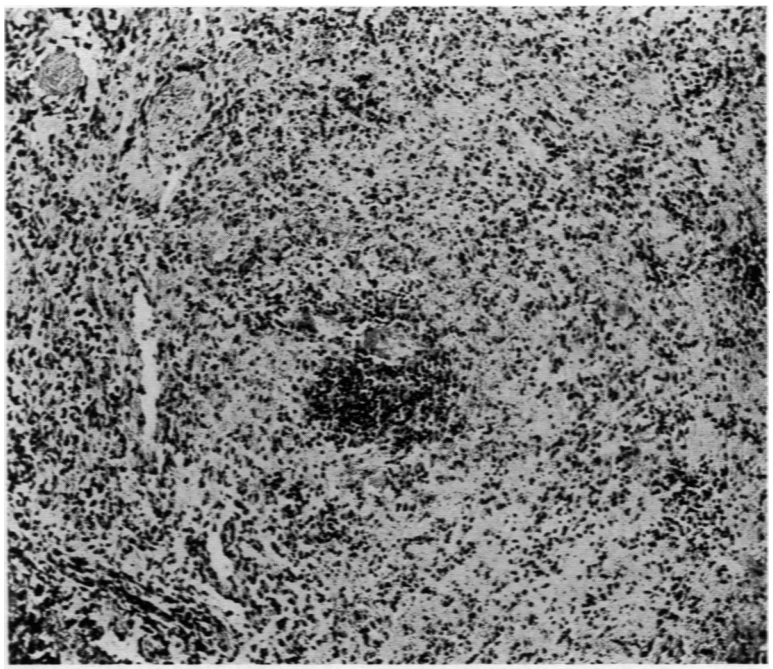

Stasney

Erythrophagocytosis in Sickle Cell Disease

PLATE 28

FIG. 5. (Case no. 7, Table I.) Spleen: markedly distended sinusoids. Sickle-shaped red blood cells are found in the pool around the lymph follicle. Hematoxylin and eosin stain. × 100.

FIG. 6. (Case no. 1, Table II.) Spleen and liver: marked disproportion in size.



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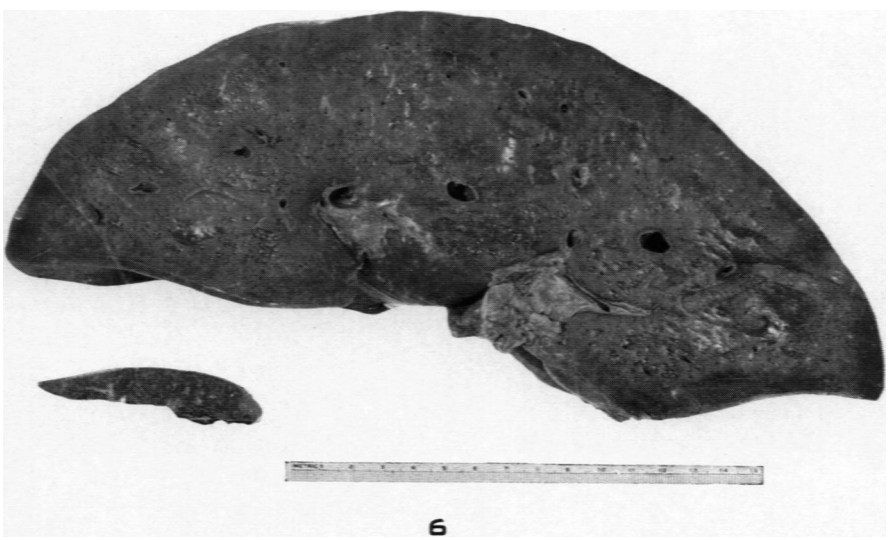

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