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Splenic Functions in Thalassemia Major*

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The spleen is a large mass of lymphoid and phagocytic reticulendothelial cells (RES) with capillaries and non-fenestrated sinusoids which enhance the filtering ability of the organ.[1] Among the number of functions, filtering, phagocytic and storage abilities are closely related to its anatomical structure.

The decrease or loss of phagocytic function of the spleen has been well demonstrated in sickle-cell anemia, in spite of the enlargement of the organ.[2-4] Although the above function of the organ has also been checked in a few cases of β-thalssemia[6] it has not been studied comparatively with filtering and reservoir abilities, to the best of our knowledge.

In this study, splenic phagocytic, filtering and reservoir functions for platelets and antihemophilic globulins have been studied in 15 cases with β-thalassemia major.

Materials and Methods

A total of 41 children were studied by dividing them into 4 groups. Group I consisted of 11 healthy children (2 girls and 9 boys), 4 to 12 years old with AA hemoglobin pattern. Group 2 comprised 8 children, 3 to 12 years of age (2 girls and 6 boys) who were splenectomized 1 to 3 years prior to the study, because of β-thalassemia major (4 cases), idiopathic thrombocytopenic purpura (2 cases) shpherocytosis (1 case) and severe α-thalassemia (1 case). Group 3 was formed by 15 cases of β-thalassemia major patients, with the age range of 3 to 17 years (9 girls and 8 boys). Seven children with the age of 2,5 to 15 years with sickle-cell-β-thaltheir assemia (4 girls and 3 boys) comprised the fourth group.[7]

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The hemoglobinopathy diagnosis was verified by starch (pH 8.6) and agar gel (pH 6.4) electrophoresis[7] in all patients and in both of parents. All children were free of infection and did not have any acute problem at the time that the tests were performed.

The phagocytic function of the spleen was studied in the last two groups one hour after intravenous injection of 99m Tc sulfur colloid (1-3 m Ci) by using Anger scintillation camera pho-Gamma III with low energy collimeter and the images were recored on polaroid film and/or 35 mm Kodak P X 402 film in pre-set time. In addition, anterior and posterior projections, left posterior images were also obtained when previous two projections did not give a splenic image. Scans were graded as normal decreased and no uptake.

Reservoir function for platelets and F-VIII procoagulant activities were evaluated before and 10 minutes after intravenous injection of adrenalin (5 γ/kg epinephrine was diluted in 10 ml 5 % glucose which was infused in 10 minutes). Platelets were enumerated by phase contrast microscopy,[8] from the finger prick and F-VIII procoagulant activity according to McMillan et al.[9] on fresh frozen samples of plasma which were stored at –30° C for no more than 3 weeks.

For the evaluation of filtering function of the spleen, the presence of Howell-Jolly bodies per 500 red blood cells were determined.

Results

In the healthy control cases platelet and AHG levels significantly elevated following epinephrine perfusion ($P<0.001$ and $P < 0.0001$, respectively) without showing relation to the age and sex of the cases. Mean baseline platelet counts and AHG levels were 320.363//µl (S.D. ±71.631, range 220.000-464.000) and 132.8 % (S.D. ±71,19, range 62-286 %) and the elevation of platelets were 8.82 % (the range 3.5 to 22.05 %) and mean AHG elevation was 93.1 % (the range being 23.18 to 151.9 %) (Figure 1 and 2). Platelet count changes could be evaluated in 5 cases in group 2 following epinephrine infusion and the minimal change were most likely due to technique (Figure 2). This group was not homogeneous from the standpoint of their platelet counts and therefore statistical evaluation was not carried out. Baseline AHG levels were normal (mean 110.8 %; range 56 to 194 %) and its elevation after epinephrine perfusion could be carried out in all 8 cases which was significant ($P < 0.01$). The mean elevation of AHG was 70.66 % with the range of 24.2 to 120.6 %.

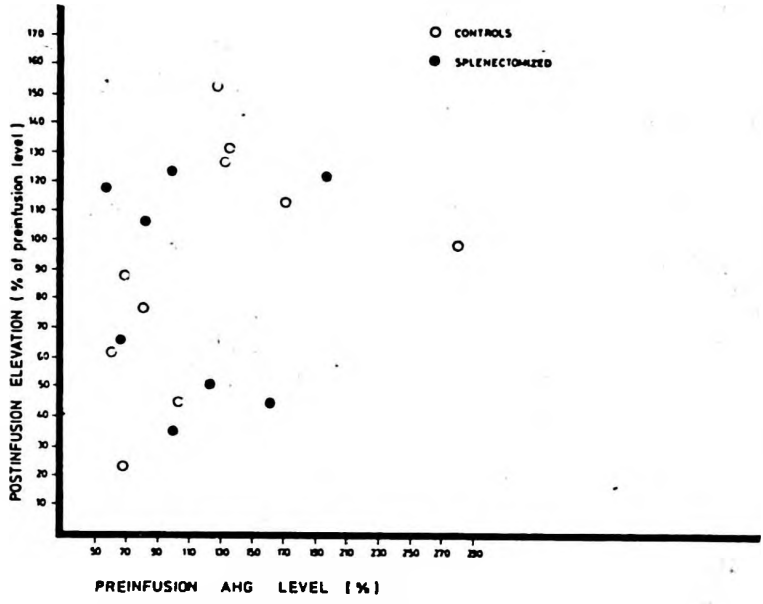

Figure 1
Preinfusion F-VIII level and its post-adrenalin elevation in controls and splenectomized children.

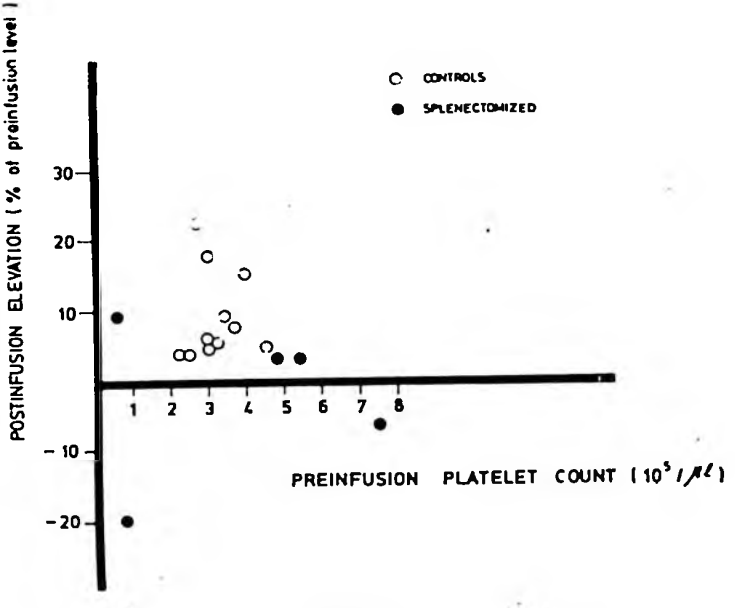

Figure 2
Preinfusion platelet counts and its post-epinephrine changes in controls and splenectomized children.

TABLE I

INITIAL PLATELET AND F-VIII LEVELS WITH THEIR ELEVATION FOLLOWING ADRENALIN PERFUSION, SPLENIC SIZE, AND SCANNING RESULTS IN PATIENTS WITH β-THALASSEMIA MAJOR.

Patient's Initials	Age/ Sex	Spleen (cm)	Platelets		Factor VIII		Splanic* scanning
			Initial ($x10^3/\mu l$)	elevation (%)	Initials (%)	elevation (%)	
Z.T.	3,F	4	100	16.0	210	61.2	N
H.T.	3,F	6	224	8.92	145	80.68	N
E.E.	4,M	2	284	5.63	121	50.41	N
A.K.	4,F	3	172	7.0	101	72.27	N
E.T.	5,F	3	272	4,41	165	63.0	N
A.Y.	5,M	3	288	19.44	152	31.57	N
A.P.	5,M	7	304	3.94	106	62.26	N
H.Y.	5,F	7	276	8.69	142	79.57	N
G.Y.	6,F	8	236	11.86	208	73.	N
M.K.	6,F	6	296	14.86	172	100.58	N
F.K.	7,F	4	148	21.62	106	37.73	N
A.K.	9,F	7	156	5.12	164	98.17	N
A.K.	10,M	4	152	5.26	116	50	N
M.P.	12,M	10	160	2.5	214	61.2	H
M.S.	17,M	2	296	9	143	87.4	N
		Mean:	224	9.28	151	67.25	

* N: Normal; H: Hypoactive. areas.

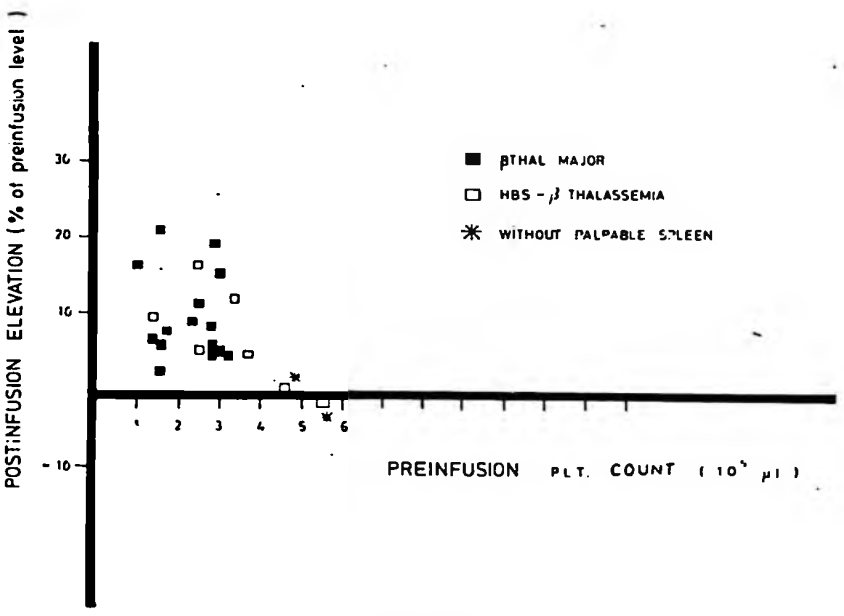

Figure 3

Preinfusion platelet count and its post-adrenalin changes in patients with β-thalassemia major and Hb-S-β-thalassemia.

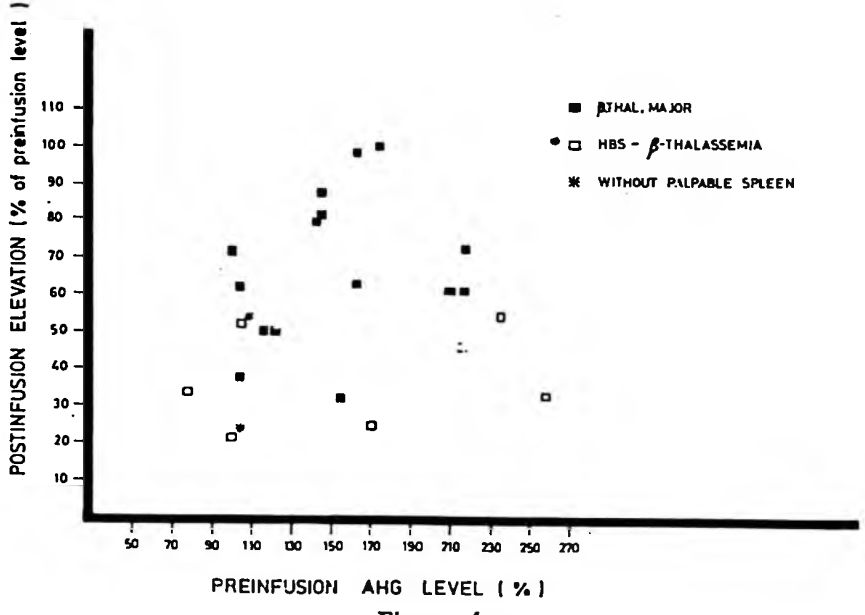

Figure 4

Baseline F-VIII level and its post-adrenalin elevations in β-thalassemia and Hb-S-β-thalassemia.

TABLE II

INITIAL PLATELET AND F VIII LEVELS WITH THEIR ELEVATIONS FOLLOWING ADRENALIN PERFUSION, SPLENIC SIZE, SCANNING AND HbF LEVELS IN PATIENTS WITH HbS-β-THALASSEMIA

Patient's Initials	Age Sex	Spleen (cm)	HbF (%)	Platelets		Factor VIII		Splenic* scanning
				Initial (μl)	Elevated (%)	Initial (%)	Elevation (%)	
1. M.G.	2.5;M	3	14	352.000	3.4	235	53.61	N
2. H.K.	8; F	3	19	328.000	12.2	215	46.04	N
3. A.A.	9; M	4	6.3	244.000	4.9	77	33.76	N.U.
4. E.H.	15; F	15	22	124.000	9.67	258	32.17	N
5. G.S.	15; F	4	18	240.000	16.66	170	25.29	N
6. Y.C.	9; M	N.P.**	11.3	456.000	0.87	104	52.88	D
7. F.S.	10; F	N.P.	20	560.000	0.7	100	21	N.U.
		Mean:	15.8	329.142	6.71	165.5	37.82	

*N: Normal; D: Decreased; N.U.: No uptake.
**N.P.: Not palpable.

Spleen scanning and platelet counts evaluation were performed in all 15 cases with β-thalassemia major. Spleen was found enlarged in all cases as was clinically evident; only in one case hypoactive areas were seen.

The baseline platelet count (mean 224,266/μl with the range of 100.000 to 304.000/μl) in this group did not differ from the controls ($P > 0.05$). The post infusion platelet elevations of all 15 cases were statistically significant ($P < 0.01$ /Table I, Figure 3). The mean preinfusion F-VIII procoagulant activity (151 %) was higher than controls and following adrenalin perfusion were significantly elevated ($P < 0.001$). Table I / Figure 4).

With one exception, all the patients in group 4 had Hb-S-β°-thalassemia. The spleen could not be visulized in one of the patients with splenomegaly and phagocytic dysfunction of the organ was shown in two the cases (no uptake in one and decreased in one) without clinical splecases (no uptake in one and decreased in one) without clinical splenomegaly (Table I). Although platelet elevation following adrenalin infusion was significant ($P < 0.05$) no response was found in 3 patients whose splenic 99mTc uptake were abnormal. In spite of preinfusion thrombocyte counts were lower in patients with palpable spleen, post perfusion rises were more marked in them (Table II/Figure 3). Mean preinfusion AHG activity in this group (165.5) %) was higher than controls and after adrenalin, statistically significant ($P < 0.01$), but lowest elevation of F-VIII activity (37.82 %) among the groups was observed.

Howell-Jolly bodies were present on the peripheral blood of all splenectomized children and of patients with Hb-S-β-thalassemia but not in normal controls and in thalassemics.

Discussion

"Functional hyposplenia" has not been reported in negroes with Hb-S-β-thalassemia[2, 6] but was present at least in two of our 7 patients with this disorder (28.6 %). With one exception all of our Hb-S-β-thalassemia combinations had β° thalassemia and their Hb-S concentrations were comparable to our patients with Hb-SS disease and their Hb F values were not significantly different from our sickle-cell anemia patients.[10] Their Hb, Hct values and ages were also comparable to the patients with sickle-cell anemia. Therefore if only decreased 99m Tc sulfur colloid uptake is correlated to Hb S concentrations, it would be expected in more of our patients with Hb S-β-thalassemia.

Platelet storage of the spleen, shown by adrenalin infusion in patients with Hb-S-β-thalassemia was normal, but F-VIII increment,

although significant, was lowest among the groups. These results indicate relative dissociation of the splenic function of these patients, as was indicated by Schwartz,[6] Falter[4] et.al., and us[11] in sickle-cell anemia cases. Platelet elevation following epinephrine perfusion was more marked in patients with enlarged spleen as indicated by Libre et al.[12] Aster and his colleagues[13] and Branehög and co-workers.[14] Libre[12] and associates[12] mentioned that factor VIII elevations did not seem to be dependent on the concomitant platelet rises following adrenalin infusion which was also observed in our cases.

In patients with β-thalassemia major, with neither of these tests, functional derangement of the organ was shown. Although the presence of Howell-Jolly bodies are reported in β-thalassemia major,[15] were not found in any of our cases.

Howell-Jolly bodies were observed n all our patients with Hb-S-β-thalassemia, Hb SS- disease[11] and in this study in all splenectomized cases, but neither in normal controls nor in thalassemics.

AHG elevation in splenectomized individuals in this study does not fit to the results of Libre and his colleagues.[12] But Falter et.al.[4] also showed AHG elevation after epinephrine infusion in splenectomized individuals; therefore its elevation may very cautiously be interpreted as showing the presence of splenic function in at least in our way of perfusion studies.

Penny and co workers[16] have reported that the spleen contains about one third of the platelet mass. It has been shown by Alter[13] and Branehog and associates[14] that the pooled platelets in the spleen are exchangeable and available to the circulation with epinephrine infusion, which is abolished by splenectomy as in our splenectomized controls. There was also not statistically significant platelet elevation after epinephrine perfusion in 3 patients with Hb-S-β-thalassemia in whom functional hyposplenism was shown by scanning (Table II). More pronounced platelet elevation in thalassemics after adrenalin, would be related to their splenomegaly in accordance to the findings of Aster[13] and Branhög et al.[14] Higher platelet counts in children and adult patients with Hb SS disease have been reported by Schwartz[6] and Freedman-Karpatkin.[17] Marked thrombocytosis was also present in two of our patients with Hb S- β-thalassemia without palpable spleen.

In conclusion splenic function in β-thalassemic patients by 99m Tc sulfur colloid phagocytosis and platelet elevation after adrenalin infusion is normal. But by phagocytosis, it is found decreased in at least 30 % of the patients with Hb-S-β-thalassemia.

Summary

Platelet and F-VIII storage and phagocytic function of the spleen have been studied in 15 patients with β-thalassemia major who were not splenectomized and in 7 patients with Hb-S-β-thalassemia. Eight splenectomized patients, 4 had β-thalassemia major, and 11 healthy children served as controls. F-VIII elevation following adrenalin was not found to be sensitive index in the evaluation of "functional hyposplenia". No functional derangement of the organ was shown in the patients with β-thalassemia major. However, at least 30 % of patients with Hb-S-β-thalassemia had functional hyposplenism with one of the tests. Higher platelet counts were observed in the latter patients, without palpable spleen, and the independence of the splenic functions were shown.

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