Table 2. A second trial involving analysis of a further 10 blood samples, some of which had been deliberately contaminated with red cells from a second sample matched for ABO and Rh(D) antigens. The percentage of red cells positive by flow cytometry for 12 blood group antigens are tabled. Certain samples (*) showed distinct differences in expression of the antigen depending on whether homozygous or heterozygous, since the intensity of expression of the antigen (mean log fluorescence 1, MFL1) was dependent on the number of antigenic sites on the surface of the red cell.

Antibody to blood group antigen	*Sample number*									
	1	*2*	*3*	*4*	*5*	*6*	*7*	*8*	*9*	*10*
Rh(C)	100	88.3	0	100	100	100	87.5	100	100	100
Rh(c)	100	100	100	100	91.2	0	100	100	100	90.3
Rh(E)	100	90.4	0	0	91.3	92.8	87.4	0	0	87.9* 8.9*
Rh(e)	100	100	100	100	89.6	7.1	100	100	100	90.6* 9.9*
K1 (Kell)	0	89.4	0	0	0	0	0	0	0	0
K2 (Cellano)	100	100	100	100	100	100	100	100	100	100
Fy^a ($Duffy^a$)	0	100	100	100	100	100	100	100	100	100
Fy^b ($Duffy^b$)	100	100	100	88.7	90.1	0	100	100	9.0	100
Jk^a ($Kidd^a$)	100	100	100	100	88.7	100	100	0	100	100
Jk^b ($Kidd^b$)	0	0	0	0	100	0	0	0	100	100
S	0	0	0	9.0	100	0	0	0	0	100
s	100	100	100	90.6	100	100	11.7	100	8.7	90.5

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Enlargement of hepatoduodenal ligament lymph nodes in β thalassemia children receiving multiple transfusions: a common observation

Of 37 thalassemic major patients receiving regular blood transfusion and chelation therapy, 12 (32.4%) had enlarged hepatoduodenal ligament nodes. Of these 12 patients, 9 (83%) had a ferritin level > 2,500 ng/mL. At the 24-month follow-up 8 patients showed persistent lymphadenopathy. It is important to recognize hemosiderin lymphadenopathy and avoid unnecessary investigation in asymptomatic thalassemic patients.

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Despite modern iron chelation therapy, iron overload and secondary hemochromatosis are still major clinical problems for thalassemia patients. Thalassemic patients are frequently referred for abdominal ultrasonography for the detection of abnormalities such as liver cirrhosis and gallstones. Incidental findings of hemosiderin-laden lymph nodes have been reported in thalassemic patients. Visualization of these nodes by plain radiography,[1] abdominal lymphography[2] and computed tomography[3,4] is feasible.

We conducted a study using ultrasonography to determine how often hepatoduodenal ligament (HDL) nodes were detected in these patients and to evaluate whether their presence had any clinical significance or relationship with ferritin levels.

Thirty-seven thalassemic patients were recruited (20 boys and 17 girls, age 2.75 to 21 years, mean 11.3 years). These patients were receiving regular blood transfusion and chelation therapy. Abdominal ultrasonography was performed by the same pediatric radiologist (WCWC) and double-checked by the second investigator (CM).

The lengths of the long and short axes of the largest HDL node identified in each patient were measured. Three measurements were made for each axis and the mean value was calculated. Enlargement of the node was diagnosed when either the mean

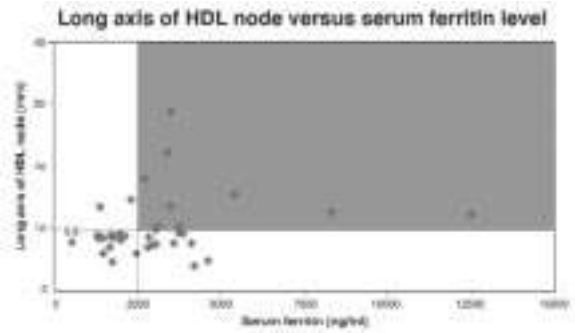

Figure 1. Long axis of the HDL nodes versus serum ferritin level in all patients. A cut-off point was taken at 9.8 mm which corresponds to one standard deviation above the mean long axis of HDL nodes in normal subjects. Note that 9 out of 11 patients (82%) with a nodal length exceeding 9.8 mm have a serum ferritin level greater than 2,500 ng/mL.

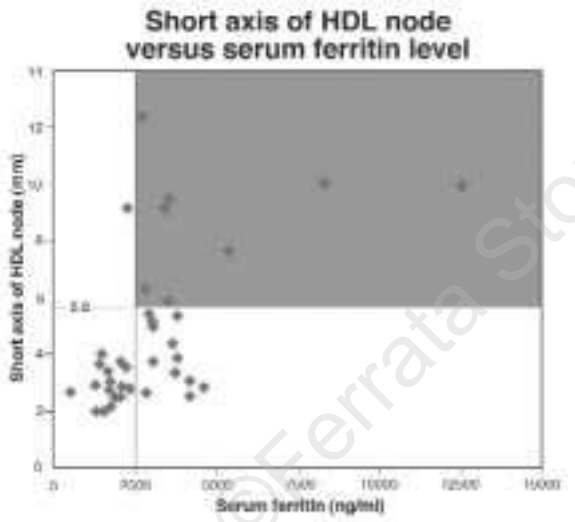

Figure 2. Short axis of the nodes versus serum ferritin level in all patients. A cut-off point was taken at 5.8 mm which corresponds to one standard deviation above the mean short axis of HDL nodes in normal subjects. Note that 8/9 patients (89%) with a nodal width exceeding 5.8 mm have a serum ferritin level greater than 2,500 ng/mL.

long axis or the mean short axis was greater than one standard deviation of the mean value quoted for a group of 128 normal subjects (of similar age range as the patients in this study) by Yang *et al.*[5]

Blood tests for serum ferritin and hepatitis serology were carried out within one month of the ultrasonography examination. Positive serology for viral hepatitis was defined as either positivity for HbsAg and/or the presence of antibodies to hepatitis C virus.

Twelve out of the 37 thalassemic children (32%) had enlarged HDL nodes. Eight patients in this group had both long and short axes measurements of HDL nodes greater than one standard deviation above the mean of the control group.[6] Three patients had only the long axis measurement more than one standard deviation, i.e. greater than 9.8 mm. One patient had only the short axis measurement more than one standard deviation. i.e. greater than 5.8mm. There was no significant direct linear correlation between the ferritin level and the sizes of the nodes ($p > 0.05$). However, 82% (9 out of 11 patients) with enlarged nodes as diagnosed by an increased long axis and 89% of the patients (8 out of 9 patients) with enlarged nodes as diagnosed by an increased short axis measurement had a high ferritin level of >2,500 ng/mL. (Figures 1 and 2). A ferritin level >2,500 ng/mL was defined as high because this level has been shown to be associated with cardiomyopathy.[6] Correlations between long and short axis measurements and a high ferritin level are equally good.

Eight out of 12 patients with enlarged HDL nodes showed no significant change in nodal size ($p=0.907$) at follow-up ultrasonography performed 24 months later. No patient had a clinically or radiologically detectable abdominal neoplasm during the two-year follow-up period. These patients had no significant change in their ferritin levels ($p=0.617$). The other four patients were lost from follow-up.

The presence of enlarged HDL nodes in patients with hepatic pathology is well documented.[7,8] Previous studies have reported that enlarged HDL lymph nodes are commonly found on ultrasonography examination in patients with benign hepatobiliary disease.[5,6] In our study, four patients were found to have positive hepatitis serology and they all had enlarged HDL nodes. However, these patients only accounted for less than half (33%) of the thalassemic patients with enlarged lymph nodes. Therefore hepatitis virus status may account in part but not completely for the enlargement of HDL nodes in thalassemia. Other causes such as neoplastic disease, inflammatory disease were also excluded by an unremarkable clinical course and unchanged imaging findings at a 24-month follow-up. We therefore suggest that these enlarged nodes are likely to represent haemosiderin lymphadenopathy. The enlargement of HDL nodes in these patients could be due to hemosiderin deposition to the liver causing reactive changes in the HDL nodes, which provide the lymphatic drainage for the liver. Another possible explanation is that the nodal enlargement is caused by direct hemosiderin deposition in the nodes themselves. Whatever the mechanism, to a certain extent the result reflects the amount of iron stores in the patient's body, though a linear relationship is not expected since ferritin values may be affected by a variety of conditions.[9]

Our data suggest that persistently enlarged HDL nodes are common findings in ultrasonography examinations in children with thalassemia major. The nodal enlargement is usually associated with a high serum ferritin level. It is important to recognize this observation. In the absence of other clinical symptoms, additional investigation is unnecessary.

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Thalidomide abolishes transfusion-dependence in selected patients with myelodysplastic syndromes

Among 25 transfusion-dependent patients with myelodysplastic syndromes (MDS) receiving up to 300 mg/d thalidomide p.o., 5 became transfusion-free within 4-9 weeks and for 6 to +24 months. Responders had a recent diagnosis, normal karyotype, no excess of marrow blasts and were younger than non-responders. Thalidomide may be effective for treating anemia in selected MDS patients.

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The potential efficacy of thalidomide in myelodysplastic syndromes (MDS), although recently reported,[1-5] has not been extensively investigated so far. We conducted a pilot study by administering thalidomide to 25 patients with MDS (14 males, 11 females, mean age 65 years, range 48-85), previously unresponsive to treatments including recombinant erythropoietin, alone or in association with other growth factors or amifostine.[6-8] All patients were heavily transfusion-dependent (Hb < 8 g/dL), requiring 4-8 units of packed red-cell transfusions every month. According to the WHO classification, there were 12 cases of refractory anemia (9 with trilineage myelodysplasia), 8 of refractory anemia with blast excess (4 < 10%, 4 > 10%), and 5 cases of refractory anemia with ring sideroblasts (1 with trilineage myelodysplasia). The International Prognostic System Score was low in 9 patients, intermediate 1-2 in 13 and high in 3 patients. Thalidomide was given at the dose of 100 mg/d *per os*, at bedtime, for 1 week (to test tolerance) and then the dose was progressively increased every 4 weeks. No patient tolerated more than 300 mg/d.

Table 1. Characteristics of transfusion-dependent MDS patients who responded to thalidomide.

	1	*2*	*3*	*4*	*5*
Age	62	69	48	64	51
Sex	M	M	F	M	F
IPSS	int-low	low	int-low	low	low
WHO	RA	RARS	RA	RA with fibrosis	RA
Time from diagnosis (months)	9	11	3	8	11
Karyotype	46XY	46XY	46XX	46XY	46XX
Marrow blasts (%)	< 5	< 5	< 5	< 5	< 5
Transfusions/Month	3	4	6	4	4
EPO (miu/L) pre/post	673/105	286/424	257/3900	155/667	303/612
WBC ($\times 10^9$/L) pre/post	1.9/2.3	9.3/4.2	2.9/2.1	6.7/8.3	3.5/3.2
PLT ($\times 10^9$/L) pre/post	39/26	236/108	42/32	319/632	107/122
Hb (g/dL) pre/post	7.8/11.3	7.3/9.3	7.5/10	6.6/9.3	7.1/11.4
Hb F (%) pre/post	1.1/n.d.	1/ 4.1	*13/48.3	0/0.5	1/5.1
Dose of thalidomide (mg)	200	200	250	200	300
Duration of response (months)	+24	6	12	+5	+19

**Concomitant thalassemic syndrome caused by a β°39 point mutation.*

Ten patients (eight more than 75 years old), stopped thalidomide early because of relevant side effects (fatigue, somnolence, constipation, numbess and tingling in fingers and/or toes, fluid retention, renal failure, skin rash). Ten additional patients stopped the treatment after 2 months because of inefficacy. The remaining 5 patients became completely transfusion-free within 4-9 weeks (Table 1). Due to a slight worsening of peripheral white blood cell and platelet counts, thalidomide was stopped in two of the responders. Since the hemoglobin value rapidly dropped to less than 8 g/dL, the drug was re-started and both patients returned to being transfusion-free (Figure 1). No further significant cytopenias were recorded in non-responders. Erythroid responses are currently maintained in 3 patients (Table 1). Two of them are still receiving thalidomide therapy, with adjusted doses of 50 to 100 mg/d. The patient who maintains his erythroid response after 2 years received the drug for only 12 months because of the subsequent occurrence of gastric carcinoma.

Few studies have investigated the therapeutic role of thalidomide in MDS so far, and all of them agree that thalidomide may significantly increase Hb levels in about one third of treated patients.[1-5] In one study thalidomide also improved neutropenia and thrombocytopenia in some patients.[5] Our findings confirm that thalidomide, at a relatively low dose, may be a very effective therapy for treating anemia in a selected group of