The Turkish Journal of Pediatrics 34: 63 - 69, 1992

EVALUATION OF GROWTH HORMONE SECRETION AND INSULIN – LIKE GROWTH FACTOR – 1 IN PREPUBERTAL CHILDREN WITH THALASSEMIA*

*Lebriz Sağlamer MD**, Lamia Ulukutlu MD***, Oya Ercan MD****
*Sezer Hatemi MD***, Hilmi Apak MD*****

Summary: Sağlamer L, Ulukutlu L, Ercan O, Hatemi S, Apak H. (Department of Pediatrics, İstanbul University Cerrahpaşa Faculty of Medicine, İstanbul, Turkey). Evaluation of growth hormone secretion and insulin-like growth factor-1 in prepubertal children with thalassemia. Turk J Pediatr 34: 63-69, 1992.

Growth retardation is a clinical feature of patients with thalassemia major, and endocrine studies have frequently revealed the presence of normal growth hormone (GH) secretion. The present study was undertaken in 14 prepubertal thalassemic children (9 males and 5 females), aged $2^{2/12}$ to $10^{3/12}$ years, with the aim of evaluating GH response to i.v. arginine, oral L-dopa stimulation and insulin-like growth factor-1 (IGF-1) levels. Eleven patients had peak serum GH levels <7 ng/ml and two patients had peak serum GH levels of 7-10 ng/ml with arginine. Similarly, 10 patients had peak levels <7 ng/ml and one patient had a peak level of 7-10 ng/ml with L-dopa. Thus, nine of the patients had GH deficiency and two had partial GH deficiency. Three patients had elevated basal GH values. The serum IGF-1 levels in the patients were not statistically different from the levels in the controls, but three patients had low IGF-1 values. These findings suggest a defect in the regulatory mechanisms of GH secretion. *Key words: thalassemia major, growth hormone, insulin-like growth factor-1.*

Growth retardation, once a constant feature of symptomatically transfused patients with thalassemia major, has been shown to be closely associated with the degree of chronic anemia[1-3]. Following hypertransfusion regimens keeping the hemoglobin (Hb) levels above 10 g/dl, near-normal growth has been reported up until the 8-10 year age range, with the pubertal growth spurt often lacking[4]. Transfusion therapy, although increasing the length and quality of life, causes lesions due to chronic iron overloading with subsequent organ damage[5]. Attention has thus been focused on endocrine abnormalities and chelation therapy[6-13].

Endocrine studies in thalassemic patients have frequently revealed the presence of normal growth hormone (GH) response to provocative stimuli[4,6-11]. Growth

* From the Department of Pediatrics, İstanbul University Cerrahpaşa Faculty of Medicine, İstanbul.

** Assistant Professor of Pediatrics, İstanbul University Cerrahpaşa Faculty of Medicine.

*** Professor of Pediatrics, İstanbul University Cerrahpaşa Faculty of Medicine.

**** Pediatrician, İstanbul University Cerrahpaşa Faculty of Medicine.

***** Resident in Pediatrics, İstanbul University Cerrahpaşa Faculty of Medicine.

retardation has been attributed to a hepatic defect of insulin-like growth factor-1 (IGF-1) biosynthesis[14,15]. Series reported in the literature show differences in terms of age, transfusion and chelation characteristics of patients. The present study was undertaken to assess GH response to provocative stimuli and IGF-1 levels in our group of prepubertal thalassemic patients.

Material and Methods

Fourteen children with homozygous beta-thalassemia (9 males and 5 females), ranging in age from $2^{2/12}$ to $10^{3/12}$ years were studied. The clinical and laboratory findings of the patients are shown in Table I. All patients were given frequent transfusions beginning in the first two years of life except for patient 4 with thalassemia intermedia. As a general rule, blood was given at Hb levels of 9-10 g/dl. As a result of delays in transfusion, due to incompliance, the mean pretransfusion Hb concentration was 7.8 ± 1.0 g/dl. The total units of blood given ranged from 9 to 180 (mean 56 ± 53 units). The group was heterogenous in respect to chelation treatment with only seven having received s.c. desferrioxamine during the last 1-4 years. The mean age at the onset of chelation therapy was seven, due to problems of compliance and finance. Serum ferritin levels were high in all, the average value being 5035 ± 2465 ng/ml. Severe liver disease was excluded on the basis of normal protein levels, normal protein electrophoresis patterns, normal prothrombin activity in all and only modest elevations of the transaminases in some of the patients (mean SGOT 23 ± 13 mU/ml, mean SGPT 36 ± 28 mU/ml).

TABLE I: Clinical and Laboratory Findings of Patients

Case No.	Age (yrs)	Age at first transfusion (yrs)	Age at initiation of chelation (yrs)	Ferritin (ng/ml)	Pretransfusion Hb (g/dl)	Transfusion load (units)	SGOT (mU/ml)	SGPT (mU/ml)
1.	10 3/12	0 6/12	6	8474	7.5	180	32	64
2.	10 3/12	0 5/12	9	5314	6.0	140	12	19
3.	9 8/12	0 6/12	6	9803	7.0	142	43	68
4.	9 3/12	8 0/12	–	970	7.0	9	8	11
5.	9 0/12	0 6/12	5	4695	8.5	77	23	38
6.	7 11/12	0 7/12	–	5021	6.5	44	19	11
7.	7 10/12	0 10/12	5	5408	8.5	63	17	90
8.	6 4/12	1 10/12	–	3861	7.0	30	27	31
9.	4 8/12	0 6/12	2	5928	9.5	47	51	74
10.	3 8/12	0 5/12	2	1930	9.0	28	19	23
11.	2 6/12	0 4/12	–	3188	8.0	15	21	9
12.	2 5/12	1 4/12	–	1772	9.0	13	17	12
13	2 3/12	0 9/12	–	–	8.5	16	19	16
14	2 2/12	0 4/12	–	–	7.5	16	–	–
Mean	6 4/12	0 8/12*	7	5035*	7.8	56	23	36

* Case 4 excluded.

The patients were studied at least two weeks after the last transfusion. Height and weight were measured and bone age was determined according to the standards of Greulich and Pyle[16]. Arginine and L-dopa stimulation tests were performed on two different days to assess GH response. After collection of one baseline sample at 0 minutes in the fasting state, all patients received 0.5 g/kg arginine as a 10 % solution i.v. over a 30-minute period. Blood was collected at 30, 60, 90 and 120 minutes. Samples were drawn before and 30, 60, 90, 120, 180 and 240 minutes after oral L-dopa administration (patients over 22.5 kg, 500 mg; 11 to 22.5 kg, 250 mg; below 11 kg, 125 mg). The sera were stored at −20 °C, and plasma GH was measured by RIA (double antibody; Diagnostic Products Corporation, Los Angeles). Peak levels below 7 ng/ml on both tests were indicative of GH deficiency, while having at least one peak level of 7-10 ng/ml was accepted as partial GH deficiency. IGF-1 was measured by the immunoradiometric method with reagents from the Diagnostic Systems Laboratories. Control subjects were a group of healthy, prepubertal children (4 males and 5 females) aged 3 to $10^{7/12}$.

Results

The physical characteristics of the patients are shown in Table II. The bone age was significantly lower than the chronological age ($p < 0.01$) and the height age was also significantly lower than the chronological age ($p < 0.01$). Two of the

TABLE II: Physical Characteristics of Patients

Case No.	Sex	Age (yrs)	Bone Age (yrs)	Height age (yrs)	Height SDS
1.	M	10 3/12	8 0/12	9 9/12	−0.3
2.	M	10 3/12	7 0/12	9 9/12	−0.5
3.	M	9 8/12	7 0/12	8 3/12	−1.3
4.	M	9 3/12	6 0/12	6 6/12	−2.6
5.	F	9 0/12	9 0/12	8 6/12	−0.7
6.	F	7 11/12	6 10/12	7 2/12	−0.7
7.	M	7 10/12	7 0/12	6 9/12	−1.2
8.	M	6 4/12	5 0/12	6 6/12	+0.2
9.	M	4 8/12	4 0/12	5 0/12	+0.6
10.	F	3 8/12	3 0/12	3 6/12	+0.1
11.	F	2 6/12	–	1 6/12	−2.7
12.	M	2 5/12	1 9/12	1 9/12	−2.2
13.	F	2 3/12	2 4/12	2 3/12	−0.2
14.	F	2 2/12	2 0/12	1 9/12	−1.4
Mean		6 4/12	5 2/12	5 8/12	−0.92
± SD		3 3/12	2 5/12	3 0/12	1.03

patients were below the third percentile for height. The mean standard deviation score (SDS) for height was 0.92 ± 1.03.

The basal growth hormone level was above 7 ng/ml on three occasions. Peak levels were attained at 30 to 90 minutes with arginine and at 60 to 120 minutes with L-dopa. Although the mean peak levels were below 7 ng/ml, there was a statistically significant difference between the basal and peak values in both tests ($p < 0.05$). The basal and peak serum GH values with arginine and L-dopa stimulation and the IGF-I levels are shown in Table III. Arginine induced a normal GH response in one (> 10 ng/ml), subnormal responses in two, and responses characteristic of GH deficiency (< 7 ng/ml) in 11 subjects. L-dopa induced normal GH responses in three, a subnormal response in one and responses characteristic of GH deficiency in 10 subjects. When the results of the two provocative tests were evaluated together, nine patients had GH deficiency and two had partial GH deficiency.

TABLE III: Serum GH Response (ng/ml) to Arginine and L-dopa and IGF-I Levels (mU/ml) in Thalassemic Children

Case No.	Arginine		L-dopa		
	Basal	Peak	Basal	Peak	IGF-I
1.	0.28	8.80	0.21	13.50	1680
2.	1.10	4.80	1.40	2.20	348
3.	0.80	4.00	0.68	4.20	3000
4.	0.05	0.14	0.13	0.09	1800
5.	0.68	1.40	1.60	1.70	4320
6.	8.00	5.80	3.60	11.50	1680
7.	0.01	6.40	0.27	5.20	1440
8.	0.30	2.30	0.35	1.50	1440
9.	7.20	4.60	2.90	3.40	2280
10.	3.80	5.80	15.00	6.80	3000
11.	4.20	11.00	8.00	11.00	–
12.	2.10	0.18	0.01	1.30	360
13.	2.30	6.00	0.25	9.80	3000
14.	0.35	10.00	1.70	5.80	348
Mean	2.23	5.09	2.58	5.57	
± SD	2.64	3.37	4.16	4.40	

The mean serum IGF-1 level in nine boys was 2185.3 ± 1558.0 mU/ml and in four girls 2007.0 ± 1269 mU/ml. There was no statistically significant difference between the mean IGF-I levels of the patients and the controls ($p > 0.90$ and

$p > 0.30$ for boys and girls, respectively), but the IGF-1 levels of three patients were clearly low.

Discussion

Although the patients studied were given transfusions regularly, the pretransfusion Hb levels were suboptimal and some were receiving no or only short-term iron chelation therapy. In spite of this, the present data on growth agree with earlier observations, in that growth retardation in beta-thalassemia patients given frequent transfusions is not severe until the age of ten[4,6]. Impaired growth has been attributed to chronic anemia and iron overload. The height SDS values of our patients showed no statistically significant correlation with age, mean pretransfusion Hb, duration of chelation therapy, transfusion load or serum ferritin levels. A tendency to follow the height of parents, though slight, could be observed.

Chronic anemia, tissue damage by hypoxia and severe iron overloading could cause endocrine abnormalities in thalassemia. In previous studies, mostly normal GH responses to provocative stimuli have been reported[6-9,11]. Stimulation by insulin-induced hypoglycemia has usually been the method used and the lower normal limit has been variable, down to 5 ng/ml. In a few patients, subnormal GH responses to different stimuli have also been reported[3,10]. By contrast, growth hormone release after provocative stimuli was found to be deficient in most of our patients, which was similar to the findings of Pintor et al[17] who also showed a reduction of the pituitary response to growth hormone releasing hormone (GH-RH)[17]. No correlation was found between peak GH levels and bone or chronological age, serum ferritin levels or total blood received. A normal GH response to GH-RH has also been reported[18]. Endocrine abnormalities have usually been attributed to iron toxicity. On the other hand, growth retardation with defective GH responses to stimulation have been reported in thalassemic patients receiving little or no blood support therapy[19].

As the results of provocative tests have usually suggested normal pituitary secretion of GH, attention has been focused on IGF-I as the cause of growth retardation. IGF-1 levels have invariably been reported to be low in thalassemic patients[14,15,17]. We observed a decrease of IGF-1 in only three of our 14 patients, which can partly be explained by the young age of our group, since IGF-1 levels have been found to be low in only a small percentage of thalassemic children below 10 years of age[14]. In older children, the IGF-1 levels considered low for age could be normal for the pubertal status, since puberty is frequently delayed. Studies on the mechanism for deficiency of IGF-1 have implicated a specific defect at the hepatic GH receptor or post-receptor levels[20]. It has been considered unlikely that generalized hepatic damage is a major cause of this condition since no major hepatic disease was present and hepatic protein

synthesis was normal in these patients[14,20]. Serum IGF-1 values have been found to correlate with the degree of iron overload[14].

Contrary to a previous belief, that endocrine function in terms of GH secretion is normal in thalassemic patients, recent observations reveal various defects, especially in spontaneous GH secretion[21]. The discordance between low GH responses to provocative stimuli and normal IGF-1 values in our patients might be explained by the positive correlation shown between IGF-I levels and mean 24-h GH concentrations[22]. High basal GH levels in some of our patients also merit attention. Patients with elevated basal GH levels, indicating decreased somatostatin secretion, have been reported[22]. Alterations in the regulatory mechanisms of GH secretion are suggested, but further investigations such as endogenous GH secretion are necessary.

REFERENCES

1. Wolman IJ. Transfusion therapy in Cooley's anemia: growth and health as related to long-range hemoglobin levels. A progress report. *Ann NY Acad Sci* 119:736, 1964.
2. Beard ME, Necheles TF, Allen DM. Clinical experience with intensive transfusion therapy in Cooley's anemia. *Ann NY Acad Sci* 165:415, 1969.
3. Kattamis C, Touliatos N, Haidas S, Matsaniotis N. Growth of children with thalassemia: effect of different transfusion regimens. *Arch Dis Child* 45:502, 1970.
4. Kattamis C, Liakopoulou T, Kattamis A. Growth and development in children with thalassemia major. *Acta Pediatr Scand Suppl* 366:111, 1990.
5. Modell B, Letsky EA, Flynn DM, et al. Survival and desferrioxamine in thalassemia major. *Br Med J* [Clin Res] 284:1081, 1982.
6. Canale VC, Steinherz P, New M, Erlandson M. Endocrine function in thalassemia major. *Ann NY Acad Sci* 232:333, 1974.
7. Lassman MN, O'Brien RT, Pearson HA, et al. Endocrine evaluation in thalassemia major. *Ann NY, Acad Sci* 232:226, 1974.
8. Flynn DM, Fairney A, Jackson D, Clayton BE. Hormonal changes in thalassemia major. *Arch Dis Child* 51:828, 1976.
9. McIntosh N. Endocrinopathy in thalassemia major. *Arch Dis Child* 51:195, 1976.
10. Costin G, Kogut MD, Hyman CB, Ortega JA. Endocrine abnormalities in thalassemia major. *Am J Dis Child* 133:497, 1979.
11. Masala A, Meloni T,Gallisai D, et al Endocrine functioning in multitransfused prepubertal patients with homozygous beta-thalassemia. *J Clin Endocrinol Metab* 58:667, 1984.
12. Krüger N, Stubbe P, Tillmann W, Schröter W. Längenwachstum bei Patienten mit homozygoter beta-Thalassämie während kontinuierlicher subkutaner Deferoxamin-Infusionen. *Dtsch Med Wochenschr* 111:176, 1986.
13. Maurer HS, Lloyd-Still JD, Ingrisano C, et al. A prospective evaluation of iron chelation therapy in children with severe beta-thalassemia. A six-year study. *Am J Dis Child* 142:287, 1988.
14. Saenger P, Schwartz E, Markenson AL, et al. Depressed serum somatomedin activity in beta-thalassemia *J Pediatr* 96:214, 1980.

15. Werther GA, Matthews RN, Burger HG, Herington AC. Lack of response of nonsuppressible insulin-like activity to short term administration of human growth hormone in thalassemia major. *J Clin Endocrinol Metab* 53:806, 1981.

16. Greulich WW, Pyle SI. *Radiographic Atlas of Skeletal Development of the Hand and Wrist* (6th ed). New York: Stanford University Press, 1970.

17. Pintor C, Cella SG, Manso P, et al. Impaired growth hormone (GH) response to GH-releasing hormone in thalassemia major. *J Clin Endocrinol Metab* 62:263, 1986.

18. Leger J, Girot R, Crosnier H, et al. Normal growth hormone (GH) response to GH-releasing hormone in children with thalassemia major before puberty: a possible age-related effect. *J Clin Endocrinol Metab* 69:453, 1989.

19. Vannasaeng S, Ploybutr S, Visutkul P, et al. Endocrine function in thalassemia. *Clin Endocrinol* (Oxf), 14:165, 1981.

20. Herington AC, Werther GA, Matthews RN, Burger HG. Studies on the possible mechanism for deficiency of nonsuppressible insulin-like activity in thalassemia major. *J Clin Endocrinol Metab* 52:393, 1981.

21. Shehadeh N, Hazani A, Rudolf MC, et al. Neurosecretory dysfunction of growth hormone secretion in thalassemia major. *Acta Paediatr Scand* 79:790, 1990.

22. Bercu BB, Shulman D, Root AW, Spiliotis BE. Growth hormone (GH) provocative testing frequently does not reflect endogenous GH secretion. *J Clin Endocrinol Metab* 63:709, 1986.