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CONTENTS

The Fanconi syndrome of cystinosis: insights into the pathophysiology

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SUMMARY: Sakarcan A. The Fanconi syndrome of cystinosis: insights into the pathophysiology. Turk J Pediatr 2002; 44: 279-282.

Cystinosis is a lysosomal storage disease, and is one of the most common causes of the Fanconi syndrome. In vitro studies of the cystine-loaded tubule provided insights into the pathophysiology of the proximal tubular defect. Proximal tubules loaded with cystine have a generalized proximal tubule transport defect characteristic of the Fanconi syndrome. The decrease in proximal tubular transport with cystine loading is due to a decrease in active transport. In cystine-loaded tubules the ATP production is severely compromised. The cystine-loaded tubule has a lower intracellular phosphate concentration than that of control tubules. This low intracellular phosphate concentration in cystine-loaded tubules likely plays an important role in maintaining intracellular ATP level. Preservation of intracellular phosphate at control levels prevents the decrease in intracellular ATP and the proximal tubule respiratory dysfunction with cystine loading.

Key words: cystinosis, proximal tubule, Fanconi syndrome.

Introduction

Cystinosis in an autosomal recessive disease characterized by a high intracellular concentration of the amino acid cystine in various organs[1-3]. Cystine accumulates predominantly in lysosomes where the exodus of cystine from this compartment to the cystoplasm is defective[2-10].

The proximal tubule reabsorbs 70% of the filtered water and sodium chloride and 80% of the filtered bicarbonate. All of the filtered glucose and amino acids as well as most of the phosphate are reabsorbed by the proximal tubule.

Cystinosis is the most common inherited cause of the Fanconi syndrome[1]. Infants with cystinosis are typically asymptomatic for the first six months of life. They usually present with failure to thrive. Evaluation of serum electrolytes reveals a hyperchloremic metabolic acidosis, hypokalemia, and hypophosphatemia. The urine will have a hyperphosphaturia, positive anion gap, and glucosuria. The electrolyte disturbances seen in cystinosis are caused by cystine accumulation in proximal tubules. The metabolic acidosis and other electrolyte disturbances seen in pediatric patients with the Fanconi syndrome of cystinosis contribute to their failure to thrive and short stature.

Fanconi Syndrome: In Vitro Studies

There is no animal model for the Fanconi syndrome of cystinosis. Foreman et al.[11] were able to load proximal tubules with cystine using cystine dimethyl ester. Cystine dimethyl ester permeates across cell membranes and leads to the intracellular accumulation of cystine. Proximal tubule cells can be loaded to cystine concentrations comparable to those measured in patients with cystinosis[11,12]. The intracellular esterases cleave cystine dimethyl ester to liberate cystine. The in vitro studies have shown that much of the cystine in proximal tubules incubated with cystine dimethyl ester is found in lysosomes[13].

There are several potential mechanisms whereby cystine could produce a generalized dysfunction in proximal tubular transport. The driving force for apical membrane sodium transport is the low intracellular sodium concentration generated by the Na-K-ATPase on the basolateral membrane that is driven by ATP. Therefore, a defect in the generalized proximal tubule transport could be due to any of the processes mediating active transport. Bergeron et al.[14] suggested that the Fanconi syndrome could be explained by an alteration in passive transport. The glomerulus produces an

ultrafiltrate of plasma that is delivered to the proximal tubule. Within the first milimeter, the composition of the luminal fluid changes dramatically[15,16]. There is an active absorption of organic solutes and bicarbonate. This leaves the luminal fluid without glucose and amino acids, and with a lower concentration of phosphate and bicarbonate than the peritubular plasma. The Fanconi syndrome could be due to an increase in permeability of the proximal tubule allowing transported solutes to leak back in to the proximal tubule lumen[15].

A defect of transporters on the apical membrane could lead to an inhibition in active transport. Foreman et al.[17] prepared brush border membrane vesicles from proximal tubules that were incubated with cystine dimethyl ester. The uptake of the amino acid proline was no different in brush border membranes prepared from control or cystine-loaded tubules. Thus, cellular cystine loading did not affect the transporters on the apical membrane to result in the inhibition in transport. Brush border membrane vesicles from rats that received intraperitoneal injections of cystine dimethyl ester also had fewer glucose transporters on renal brush border membrane than control rats[17].

A generalized inhibiton in proximal tubule transport due to decrease in Na-K-ATPase activity was investigated by Coor et al.[18] and Foreman et al.[19] Neither was able to demonstrate that cystine loading inhibited the Na-K-ATPase directly.

Another possible hypothesis is that the reduction in proximal tubular transport in cystine-loaded tubules could result from a reduction in fuel (ATP) to the pump. Coor et al.[18] when a major reduction in intracellular ATP individually dissected proximal convoluted tubules were incubated with cystine dimethyl ester. Incubation of control tubules with exogenous 1 mM ATP did not significantly affect intracellular ATP levels and had no effect on transport in control tubules[18]. However, incubation of proximal tubules with cystine dimethyl ester resulted in a 89% reduction in proximal tubular transport, while in the presence of exogenous ATP there was only a 45% reduction in volume absorption[18]. Therefore, proximal tubule cystine loading resulted in a decrease in intracellular ATP, and repletion of ATP ameliorated the transport defect. A depletion in intracellular ATP content has also been demonstrated by Foreman et al.[19] utilizing suspensions of proximal tubules loaded with cystine.

Some researchers focused on the effect of proximal tubular cystine loading on cellular metabolism, especially the respiration component[20]. In control proximal tubules, addition of ouabain, an inhibitor of the Na-K-ATPase, resulted in a 50% decrease in oxygen consumption. Thus, under basal conditions, half of proximal tubule oxygen consumption is consumed to mediate proximal tubular transport. Oxygen consumption in cystine-loaded tubules was only 50% of control tubules, which is consistent with the previous studies that suggested a reduction in intracellular ATP. This effect has been shown by others[21]. Of interest was the fact that cystine-loaded tubules had an almost total inhibition in the oxygen consumption utilized for active transport[20]. Non-transport directed oxygen consumption remained intact. These studies suggest that when cystine loading injures a proximal tubular cell, maintenance of vital functions necessary for cellular survival are not affected.

Some studies have suggested that depletion in intra-cellular phosphate may play a role in the pathogenesis of the Fanconi syndrome[22]. Infusion of maleic acid results in a generalized proximal tubule transport dysfunction[22]. Al-Bander et al.[22] found that infusion of sodium phosphate attenuated the fall in glomerular filtration rate, the aminoaciduria, and bicarbonaturia in maleic acid treated dogs. These investigators suggested that the reduction in intracellular phosphate produced the defect in proximal tubule transport[22].

The proximal tubule is dependent on phosphate to maintain active transport[23,24]. Perfusion of proximal convoluted tubules with an ultrafiltrate-like solution without phosphate resulted in a total inhibition of active transport[23]. When proximal tubules were perfused, with an ultrafiltrate-like solution without glucose or with glucose and phlorizin, inhibitor of glucose transport, active transport was the same as that measured in the presence of phosphate. Similarly, the rate of proximal tubule oxygen consumption was impaired in the absence of phosphate in the incubation solution, only when glucose was present[23].

Glucose-induced inhibition in proximal tubule transport and oxygen consumption in the absence of phosphate are analogous to Crabtree effect[25], where there is a glucose-dependent decrease in intracellular phosphate due to the accumulation of phosphorylated glycolytic intermediates. In the presence of glucose, there is thus a critical depletion of free intracellular phosphate that compromises oxidative phosphorylation and produces depletion in ATP.

Bajaj et al.[26] incubated isolated proximal tubules with cystine dimethyl ester. Cystine-loaded tubules had a 40% reduction in intracellular phosphate. Following this in vitro study, as an in vivo experiment, rabbits were given an infusion of sodium phosphate, sodium sulfate, or sodium chloride prior to isolation of proximal tubules. Cellular cystine loading produced a reduction in both groups; however, the intracellular phosphate concentration in cystine-loaded proximal tubules that received a phosphate infusion was comparable to that of the proximal tubules from animals that received a sulfate infusion and were cystine-loaded. Thus, cystine-loaded tubules after phosphate infusion had an intracellular phosphate concentration comparable to control tubules. After cystine loading, there was a reduction in oxygen consumption in the sulfate and chloride group, but the tubules from animals that received a phosphate infusion had no reduction in cellular respiration after cystine loading.

Cellular cystine loading with cystine dimethyl ester produced a significant reduction in intracellular ATP in the chloride and sulfate group. Intracellular ATP was not reduced in cystine-loaded proximal tubules prepared from animals that received an infusion of phosphate. The same group of researchers also demonstrated that when cystine-loaded proximal tubules were perfused with solution containing 1 mM phosphate, there was a 75% reduction in tubule volume absorption, whereas the tubules perfused with 4 mM phosphate did not show any change in the transport. These data demonstrate that cystine loading results in a reduction in intracellular phosphate. Maintenance of intracellular phosphate preserves intracellular ATP and prevents the reduction in proximal tubular respiration and proximal tubular transport with cystine loading.

Most of the studies examining the pathogenesis of the Fanconi syndrome of cystinosis have utilized cystine to duplicate its pathogenesis in vivo. In addition, the Fanconi syndrome seen in other diseases may be generated by other mechanisms than those described above in the cystine-loaded tubules.

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Frequency of Mycoplasma pneumoniae among atypical pneumonia of childhood

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We aimed to investigate the frequency of Mycoplasma pneumoniae among atypical pneumonia of childhood that is acquired from the community and to determine a practical approach to the diagnosis of these patients.

In this prospective study, 55 patients (31 male and 24 female) with atypical pneumonia were investigated with conventional laboratory and radiological methods as well as culture and polymerase chain reaction (PCR) on throat swab. In addition, serum of the patients was tested for M. pneumoniae specific IgM. The patients were reevaluated clinically at 3-5 days and 3-4 weeks and serologically at 3-4 weeks. The data on patients with M. pneumoniae pneumonia were compared with the other patients with atypical pneumonia and controls. All patients were treated with macrolide antibiotics. The mean age of the patients was 7.8±2.9 years. The frequency of M. pneumoniae by this method was 34.5%. Neither clinical, laboratory, or epidemiological data nor response to macrolide antibiotics was useful in detecting the etiology of atypical pneumonia. Diagnostic sensitivity and specificity of IgM+IgG antibodies plus PCR on throat swab were estimated as 100%. M. pneumoniae was an important microorganism in the etiology of atypical pneumonia of childhood in our community. In order to prevent loss of time with beta-lactamase antibiotics, which are usually started in severe pneumonia, serologic tests and PCR must be done during the initial evaluation of the patient for the reliable diagnosis of M. pneumoniae, which will increase the chance of early and appropriate therapy.

Key words: childhood, infection, Mycoplasma pneumoniae, pneumonia.

Mycoplasma pneumoniae infections have been commonly reported over the past 25 years[1]. Laryngotracheitis is the most common clinical presentation of M. pneumoniae infections. The infection is asymptomatic in 15 to 55% and especially during infancy. Pneumonia develops in 3 to 10% of the patients[1,2]. M. pneumoniae, which was the etiologic factor in 5 to 10% of community-based pneumonia cases in the 1980s, has accounted for 20 to 30% of cases in the past few years[3-6]. M. pneumoniae infections may not be diagnosed in the majority of the patients in our country, and there are only a few studies in the pediatric age group. In this investigation, we tried to determine the incidence of M. pneumoniae infections among patients with community-based pneumonia that presents like atypical pneumonia with its clinical and radiological features. Atypical pneumonia was defined as clinical diagnosis of pneumonia with poor correlation between the clinical findings and chest x-ray[7]. The chest x-ray of these patients showed typical patchy infiltration for atypical pneumonia. In addition, we compared the clinical, radiological, and epidemiological features as well as the response to macrolide antibiotics of M. pneumniae-positive and –negative patients with atypical pneumonia in order to propose a practical approach and management strategy for patients with community-based pneumonia in an outpatient setting.

Material and Methods

This prospective study was carried out in the outpatient clinic of the Department of Pediatrics at İstanbul University Faculty of Medicine

between April 1995 and May 1996. It was conducted with 55 patients with atypical pneumonia and 20 healthy children as the control group. M. pneumoniae-positive patients were named Group 1, and negative patients were named Group 2. A third group served as the control group and consisted of 20 healthy children who had no respiratory complaints during the past month. The history and physical findings as well as the family history and epidemiological features, such as seasonal variations of respiratory symptoms in the family, were recorded. A complete blood count, differential leukocyte count, AST, ALT, and a chest X-ray were obtained from all patients. M. pneumoniae was investigated with culture and polymerase chain reaction (PCR) of the throat swab. We used Hayflick biphasic medium in culture technique[8]. Dot-blot hybridisation method was applied to the PCR products. In addition, M. pneumoniae specific IgM was measured by immunocapture ELISA (Platelia, Sanofi Diagnostica Pasteur, France), and M. pneumoniae specific IgG was measured by ELISA (Platelia, Sanofi Diagnostica Pasteur, France) in the sera of all patients during the acute phase and convalescent phase) (3^{rd}-4^{th} week of follow-up period)[9]. Inclusion criteria of the patients in Group 1 were defined as follows. 1) All patients had atypical pneumonia. Atypical pneumonia was defined as clinical diagnosis of pneumonia with poor correlation between the clinical findings and chest X-ray[7]. The chest X-ray of these patients showed typical patchy infiltration for atypical pneumonia. 2) Those who had used antibiotic therapy over the past two weeks were excluded. 3) Patients were considered as having M. pneumoniae when a significant increase in IgM and IgG+IgM levels occurred between the two serum samples, and when PCR was positive with the presence of accompanying clinical evidence. Patients who had only IgM antibodies against M. pneumoniae were not accepted as M. pneumoniae pneumonia because this positive result of IgM might have been due to a silent M. pneumoniae infection in the past 3-4 weeks[10]. The patients in Group 2 were also diagnosed as atypical pneumonia, but their infections were caused by agents other than M. pneumoniae. Sera in Group 2 did not show any rise of M. pneumoniae specific antibody. In the control group (Group 3), we were able to analyze IgM and IgG antibodies against M. pneumoniae at the beginning of the study. The frequency of M. pneumoniae positivity was compared between Groups 1 and 2 and the control group. In addition, Groups 1 and 2 were compared with respect to clinical and radiological findings as well as other laboratory findings and the response to therapy. All patients in Groups 1 and 2 were treated with macrolide antibiotics: erythromycin 40 mg/kg for 10 days, clarithromycin 15 mg/kg/day for 10 days, or azithromycin 10 mg/kg/day for 3 days. The patients were reevaluated clinically at 3-5 days and 3-4 weeks and serologically at 3-4 weeks. Recovery was accepted as total absence of the clinical symptoms. Student's t test, chi-square or Fisher's exact test was utilized for statistical evaluation. A p value of <0.05 was considered statistically significant.

Results

The mean age of the patients was 7.8 ± 2.9 (1.3-14) years. Seven patients (12.7%) were <5 years of age, 32 (58.2%) were 5-9 years, and 16 (29.1%) were >9 years old. The age distribution among M. pneumoniae-negative patients was 6 (16.6%), 17 (47.3%), and 13 (36.1%), and among M. pneumoniae-positive patients was 1 (5.2%), 15 (78.9%), and 3 (15.7%), respectively. Although the frequency of M. pneumoniae among 5.9-year-old patients was higher, this difference did not reach statistical significance.

All 55 patients in the study groups (31 male, 24 female) had serologic tests. Forty-three patients had a throact swab for culture and PCR. Forty-nine patients were available for a clinical and serologic evaluation at 3-4 weeks. Nineteen patients (19/55, 34.5%) received diagnosis of M. pneumoniae pneumonia. The results of the serologic tests and PCR of these patients are summarized in Table I. None of the patients had M. pneumoniae positive culture in their throat swab. We could not isolated any other agents in culture medium. Two patients with positive IgG in non-Mycoplasma group (Cases 19 and 25) were considered to have past infection, because there were no increases in IgG level, and PCR was negative (Table II). Cases 29, 31 and 36 in the non-Mycoplasma pneumonia group were included in this group due to negative PCR test results according to inclusion criteria. We suggested that all cases

with positive PCR tests had increased IgG antibody levels in the convalescent period. The mean age of the control group was similar at 8.8±3.3 (1-13) years. Three children in this group had positive IgM (3/20,15%). However, they did not have any clinical findings of pneumonia and their PCR tests in throat swab were negative for M. pneumoniae.

Among patients who were diagnosed by acute and convalescent period serology according to increasing level of IgM and IgG positivity, 18 (90%) had positive PCR. This ratio increased to 100% after applying dot-blot hybridisation method to the PCR products. The diagnostic sensitivity of single sera IgM antibodies against M. pneumoniae pneumonia was 76%, and specificity was 97.1%. The positive predictive value of this test was 95%, and the negative predictive value was 84.6%. If positive sera against M. pneumoniae and PCR on throat swab were used together in diagnosis of M. pneumoniae pneumonia in the acute period, sensitivity, specificity, and positive and negative predictive values increased to 100%.

Table I. Properties of M. Pneumoniae-Positive Patients (n:19)

		1st Day		3rd-4th Week		1st Day
Case#	Age (year)	IgM	IgG	IgM	IgG	PCR
1	5.3	+	+	+	+	+
2	5.6	–	–	+	–	+
3	5.2	+	+	+	+	+
4	9	+	+	+	+	+
5	7.8	+	+	+	+	+
6	9	+	+	+	+	+
7	10	++	+	++	+	+
8	8	++	+	++	+	+
9	2	–	–	+	+	+
10	5.8	–	+	–	+	+
11	8.5	+	–	+	–	+
12	7	+	–	0	0	+
13	7	+	–	0	0	+
14	8	+	+	+	+	+
15	5	+	–	0	0	+
16	9	–	–	+	+	+
17	9	+	+	+	+	+
18	13	–	–	+	+	+
19	10	–	–	+	+	+

0 : Not analyzed.
PCR: polymerase chain reaction.

Table II. Properties of M. Pneumoniae-Negative Cases (n:36)

		1st Day		3rd-4th Week		1st Day
Case#	Age (year)	IgM	IgG	IgM	IgG	PCR
1	10	–	–	–	–	–
2	10	–	–	–	–	0
3	5.5	–	–	–	–	–
4	6.6	–	–	–	–	–
5	10	–	–	–	–	0
6	14	–	–	–	–	–
7	11	–	–	–	–	0
8	9	–	–	–	–	–
9	4.1	–	–	–	–	0
10	13	–	–	–	–	0
11	11	–	–	–	–	0
12	8	–	–	–	–	0
13	8	–	–	–	–	0
14	5.8	–	–	–	–	0
15	12	–	–	–	–	0
16	8.5	–	–	–	–	–
17	5.3	–	–	–	–	–
18	10.5	–	–	–	–	–
19	6	–	+	–	+	–
20	8	–	–	–	–	0
21	12	–	–	–	–	–
22	1.8	–	–	–	–	–
23	5.5	–	–	–	–	–
24	6	–	–	–	–	0
25	1.2	–	+	–	+	–
26	12	–	–	–	–	–
27	10.1	–	–	–	–	–
28	7	–	–	–	–	–
29	3.5	–	–	0	0	–
30	8	–	–	–	–	–
31	2.5	–	–	0	0	–
32	4	–	–	–	–	–
33	8	–	–	–	–	–
34	10	–	–	–	–	–
35	8	–	–	–	–	–
36	8	–	–	0	0	–

0: Not analyzed; PCR: polymerase chain reaction.

There were no differences between M. pneumoniae-positive and –negative patients with respect to mean age, the nature and duration of complaints, clinical findings, or epidemiological features (Table III). The most frequent radiological finding was unilateral or bilateral interstitial infiltration in both M. pneumoniae-positive (55.5%) and –negative (54%) patients.

Although the response rate to macrolide antibiotics was higher in Group 1 than Group 2 (94.4% vs. 80.6%), the difference was not statistically significant. A total of six patients in both groups did not respond to therapy (2 erythromycin, 3 azithromycin, and 1 clarithromycin).

Table III. Characteristics of Patients in Group 1 and 2 (n: 55)

	M. pneumoniae positive cases (n:19)	M. pneumoniae negative cases (n:36)	Significance
Age (mean±SD; year)	7.6±2.3	7.9±3.2	NS
Duration of complaints (mean±SD; day)	14.8±13.4	16.8±14.1	NS
Complaints n (%)			
Cough (dry)	14 (74)	21 (58)	NS
Cough (productive)	6 (32)	14 (39)	NS
Rhinorrhea	6 (32)	15 (42)	NS
Headache	9 (47)	10 (28)	NS
Chest pain	2 (11)	6 (17)	NS
Dyspnea	4 (21)	6 (17)	NS
Nausea/vomiting	7 (37)	12 (33)	NS
Anorexia	14 (74)	17 (47)	NS
Weight loss	3 (16)	2 (6)	NS
Fever	4 (21)	7 (19)	NS
Myalgias/arthralgias	4 (21)	10 (28)	NS
Dermatologic lesions	2 (11)	2 (6)	NS
Malaise	6 (32)	14 (39)	NS
Wheezing	2 (11)	2 (6)	NS
Clinical finding n (%)			
Pharyngeal erythema	13 (68)	26 (72)	NS
Cervical adenopathy	1 (5)	8 (22)	NS
Generalized adenopathy	2 (11)	3 (8)	NS
Rhonchi	14 (74)	28 (78)	NS
Wheezing	6 (32)	14 (39)	NS
Sinusitis	7 (37)	13 (36)	NS
Positive family history n (%)	11 (58)	19 (53)	NS
Admission in June to January n (%)	10 (53)	17 (47)	NS

NS: Not significant.

Discussion

The diagnosis of M. pneumoniae pneumonia is done by the isolation of the microorganism by culture from the respiratory secretions, the detection of antigen and nucleic acids, or the serologic tests mentioned above. Although culture used to be the 'gold standard' for diagnosis, it may take 1-3 weeks to grow the organism, it may yield 30% false negative results[11], and it may continue to be positive during convalescence[12]. Culture was not helpful in our investigation either. It was known that cold agglutinin could be positive in cases with atypical pneumonia[13]. But its diagnostic value is limited. We did not use cold agglutinin assay because ELISA has been the most powerful method to search for specific IgM in recent years[11,14,15]. The disadvantage of this method is that IgM cannot be detected for at least one week after the symptoms have started and some patients may be missed in the acute phase[14,16]. If single serum analysis of IgM level is used for diagnosis, healthy children who have had recent M. pneumoniae infection may get misdiagnosed. Although a second test during convalescence covers the gap, it may only give a retrospective diagnosis and does not contribute to the planning of therapy. In our study, we used immunocapture ELISA, which was reported to be sensitive[17-20], as well as PCR, which was proposed to be a rapid and reliable method[21,22]. On the other hand, the results of dot-blot hybridisation along with PCR are completely parallel to the serologic tests on double sera, and the diagnosis can be established rapidly and reliably. These data are similar to those of Waris et al.[9] who found that ELISA when used with PCR in the acute phase is the most rapid and reliable method of diagnosis for M. pneumoniae. Dorigo-Zetsma et al.[22] also proposed that a combination of PCR and complement-fixation test may cover diagnosis of all patients. Similar to our results, Kessler et al.[21] reported that the sensitivity of

PCR increases when used in combination with the hybridisation technique. The frequency of 34.5% among atypical pneumonia of childhood shows that M. pneumoniae is a common etiologic factor in community-based pneumonia[6,23,24]. Dereli et al.[25] researched IgM as a diagnostic tool for Chlamydial infections in infants. They demonstrated that IgM antibody was positive in 12 cases among 20 culture-positive cases. However, its frequency may be underestimated due to utilization of a single ELISA method on a single serum sample. It has been emphasized in the literature that M. pneumoniae pneumonia is being under-reported because of inadequate use of the diagnostic tools[5,26,27]. On the other hand, a cost-benefit analysis between ELISA and PCR test was done, and the ELISA test is cheaper than PCR. With two repeated analyses, cost of ELISA was only 30% of PCR cost.

Although M. pneumoniae infection is most frequently encountered between 5 to 9 years of age, occurrence in lower age groups has been reported[28]. The age range of our patients coincided with this trend, and only 5% were younger than 5 years of age. The lack of a significant difference between the M. pneumoniae-positive and –negative patients within a certain age range among those with atypical pneumonia implicates that M. pneumoniae may be a causative agent at younger ages as well. These data are also compatible with the literature[9,29]. Until recently, it has been thought that epidemiological features may be helpful in presumptive diagnosis[30]. However, recent studies show that not only epidemiological, but also clinical and radiological findings, are unhelpful in distinguishing M. pneumoniae pneumonia from other atypical pneumoniae or community-based pneumonia[7,26,31]. Our results support this view.

Although patients with M. pneumoniae pneumonia have responded better to macrolide antibiotics, the difference from other atypical pneumoniae is not significant[5,22,32-34]. Our results also reveal that macrolides may be good alternatives for patients over 2 years of age with pneumonia who can be treated as outpatients. We suggest that cases unresponsive to macrolides might be attributed to other agent, such as viruses.

In conclusion, M. pneumoniae was the etiologic agent in 34.5% of atypical pneumonia in childhood in our community. The epidemiological, clinical, and radiological findings failed to distinguish the etiology of the pneumonia. Consequently, patients with community-based pneumonia seem to be effectively treated with macrolides on an outpatient basis. However, severely ill patients who require admission need specific M. pneumoniae tests with ELISA and PCR in order to minimize the loss of time with beta lactam antibiotics, which are usually the first-line treatment in this group of patients.

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Evaluation of soluble transferring receptor levels in children with iron deficiency and beta thalassemia trait, and in newborns and their mothers

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In this study we first aimed to investigate the value of soluble transferrin receptor levels (sTfR) in healthy, iron deficient and beta thalassemia trait children and to determine whether sTfR is a useful indicator of iron deficiency. Secondly, we investigated the effects of iron supplementation of sTfR levels in a group of iron deficient children. Third was to describe sTfR in newborn infants and determine whether or not maternal iron deficiency is an important predictor of infant sTfR. Six groups were formed: Children with iron deficiency (n=22), post-iron therapy (n=16), beta thalassemia traits (n=19), healthy children (n=19), full-term newborns (n=20), and their mothers (n=19), Complete blood count (CBC), serum iron, iron-binding capacity, ferritin and sTfR levels were measured. sTfR/log ferritin indexes were calculated. sTfR levels of children with iron deficiency and with beta thalassemia trait were found to be significantly higher than those of healthy children ($p<0.0001$ and $p<0.001$). Children with iron deficiency showed a greater increase in the levels of sTfR than those with beta thalassemia traits ($p=0.008$). Although sTfR levels of subjects having iron therapy decreased, the levels still remained high compared to controls ($p=0.002$). Newborns had significantly higher levels of sTfR than controls ($p<0.0001$). Although sTfR levels of mothers with iron deficiency were higher than those of mothers having no iron deficiency ($p=0.009$), there was no difference in the levels of sTfR between newborns of both groups of mothers ($p=0.790$). sTfR is a useful parameter which shows body iron status as well as erythropoietic activity in children. It is independent of mother's iron status, and is due to erythropoietic activity in newborns.

Key words: soluble transferring receptor, iron deficiency, beta thalassemia trait, newborn, children.

Iron plays an essential role in the body. Cellular iron uptake is achieved by transferring receptors on the membrane. Transferrin receptor (TfR) is a transmembrane dimeric glycoprotein composed of two identical 95 kDa subunits, linked by disulfide bonds. In the human body, 80% of the transferring receptors are located in the erythroid marrow[1]. Although TfR can be identified on nearly every cell type, it is predominantly expressed by maturing cells of the erythroid lineage, which have high iron requirements for heme synthesis, and syncytiotrophoblasts. TfR expression increases with cell proliferation, differentiation and iron need.

Th TfR is found in a soluble form in serum. Serum TfR (sTfR) is a truncated form of the intact TfR and has been identified as a monomeric fragment of the extracellular domain with a molecular mass of 85 kDa. sTfR levels are not affected by age, sex and pre- or postmenopausal status of adults or by acute or chronic infections[1-3]. In one report, sTfR concentration in 485 healthy infants between 9 and 15 months of age was not related to age or sex[4]. With these features sTfR seems to be a new and useful parameter in the diagnosis and differentiation of iron deficiency[1-3].

Studies on sTfR levels of children are few. In this study we first aimed to investigate the value of sTfR in healthy, iron deficient and beta thalassemia trait children and to determine whether or not sTfR is a useful indicator of iron deficiency. Second, we investigated the effects of iron supplementation of sTfR levels in a

group of iron deficient children. Third was to describe sTfR in newborn infants and determine whether or not maternal iron deficiency is an important predictor of infant sTfR.

Material and Methods

Six study groups were formed. None of the subjects in these groups had an acute or chronic disease, or had used iron preparations within the last one year. Group 1 consisted of 22 children (12 male, 10 female) aged 2.2±1.5 years old (10 months to 6.5 years old) and diagnosed as iron deficiency according to the criteria which included low serum ferritin (<10 ng/ml) with or without low transferring saturation (<14%) and low hemoglobin (<11 g/dl)[5]. Group 2 included 16 subjects from Group 1 who were appropriately treated with iron (6 mg/kg/d, peroral) for two months. All of the children in Group 2 had serum ferritin levels >10 ng/ml and hemoglobin >11 g/dl. Group 3 consisted of 19 beta thalassemia trait children (11 male, 8 female) aged 8.4±3.6 years old (6 months to 14 years old) and diagnosed by hemoglobin electrophoresis as having >3.5% HbA_2. Group 4 (control group) included 19 healthy children (6 male, 13 female) aged 4.3±2.4 years old (9 months to 8 years old). All of the healthy subjects had serum ferritin levels >10 ng/ml and hemoglobin >11 g/dl. Group 5 consisted of 20 healthy full-term newborns with mean birth weight of 3,300 g. Within the first day of life blood samples were taken from the newborns. Group 6 were the mothers of newborns in Group 5, aged 26.3±5.4 years old (17 to 35 years old). Group 6 was divided into those with iron deficiency (ferritin <12 ng/ml and/or Hb<10.5 g/dl) (n=9) and those without iron deficiency (n=10).

The subjects were outpatients. The parents were informed and consented to the study.

Cell blood counts, serum iron, total iron-binding capacity, ferritin and sTfR levels of the subjects were analysed. Cell blood counts were determined with Cell-DYN 3500R (Abbott) automatic cell counter. Serum iron, total iron-binding capacity and ferritin were measured by standard methods. sTfR levels were analysed with fluorescence polarization immunoassay (AIA-Pack-sTfR kit, Belgium) by an automated analyser (TOSOH, Japan). For the statistical evaluations, Mann-Whitney U and Pearson correlation tests were used.

Results

Mean hemoglobin, hematocrit, mean corpuscular volume (MCV), serum iron, iron-binding capacity, ferritin, sTfR levels and sTfR/log ferritin indexes of the groups are given in Table I. Results of sTfR levels are shown in Figure 1.

Table I. Hemoglobin, Hematocrit, MCV, Serum Iron, Total Iron-Binding Capacity, Ferritin, sTfR Levels and sTfR/log Ferritin Indexes of the Groups

Groups	Hemoglobin (g/dl)	Hematocrit (%)	MCV (fl)	Serum iron (μ/dl)	Total iron-binding capacity (μ/dl)	Ferritin (ng/ml)	STFR (U/ml)	STFR/log ferritin index
Group 1: Iron deficiency (n=22)	10.3±1.3 (7.7-12.2)	31.8±3.2 (26.7-36.5)	68.8±6.9 (55.0-80.8)	42.5±29.1 (7-118)	401±55 (281-500)	5.0±2.8 (1.5-9.7)	925±268 (607-1500)	1198±2553 (681-8158)
Group 2: After iron therapy (n=16)	11.9±0.8 (11.0-13.3)	36.9±2.4 (34.0-41.5)	74.7±4.3 (67.3-82.7)	56.7±20.2 (32-192)	353±87 (220-503)	25.2±11.6 (10.9-53.8)	686±137 (557-1151)	485±168 (358-1009)
Group 3: β-thalassemia traits (n=19)	10.6±0.8 (9.5-12.4)	33.2±2.6 (27.6-37.8)	58.7±2.8 (53.3-67.4)	71.5±23.0 (47-109)	303±55 (216-399)	39.8±19.1 (15.4-85.2)	725±153 (488-1074)	488±90 (332-667)
Group 4: Healthy children (n=19)	11.8±0.5 (11.0-12.9)	35.5±1.5 (33.0-38.3)	78.7±4.4 (72.8-90.9)	75.4±50.8 (7-229)	309±50 (224-421)	41.8±33.5 (10.3-151)	578±80 (389-725)	391±86 (269-585)
Group 5: Newborns (n=20)	17.6±1.3 (14.6-19.5)	52.4±4.3 (42.6-59.1)	100.3±2.8 (96-105)	46.9±26.5 (18-141)	215±70 (108-403)	260.2±142.0 (8.8-560)	849±186 (575-1315)	359±244 (224-1398)
Group 6: Mothers of newborns (n=19)	11.5±1.0 (9.7-13.9)	35.4±2.9 (30.2-41.5)	83.9±6.1 (69.0-96.7)	57.4±18.8 (30-98)	386±103 (147-568)	20.8±25.1 (6.5-101)	672±278 (233-1500)	661±358 (155-1704)
With ID (n=9)	10.9±0.9 (9.7-12.3)	33.9±2.5 (30.2-36.7)	82.4±5.3 (69-87)	58.4±21.5 (35-98)	387±69 (292-505)	9.1±1.4 (6.5-11)	836±279 (536-1500)	829±335 (525-1704)
Without ID (n=10)	11.9±0.9 (10.7-13.9)	36.8±2.6 (32.8-41.5)	85.3±6.7 (76.4-96.7)	56.6±17.1 (30-78)	385±130 (147-568)	31.4±31.7 (12.1-101)	524±185 (233-101)	392±201 (155-756)

* ID: iron deficiency: MCV: mean corpuscular volume; STFR.serum transferrin receptor.

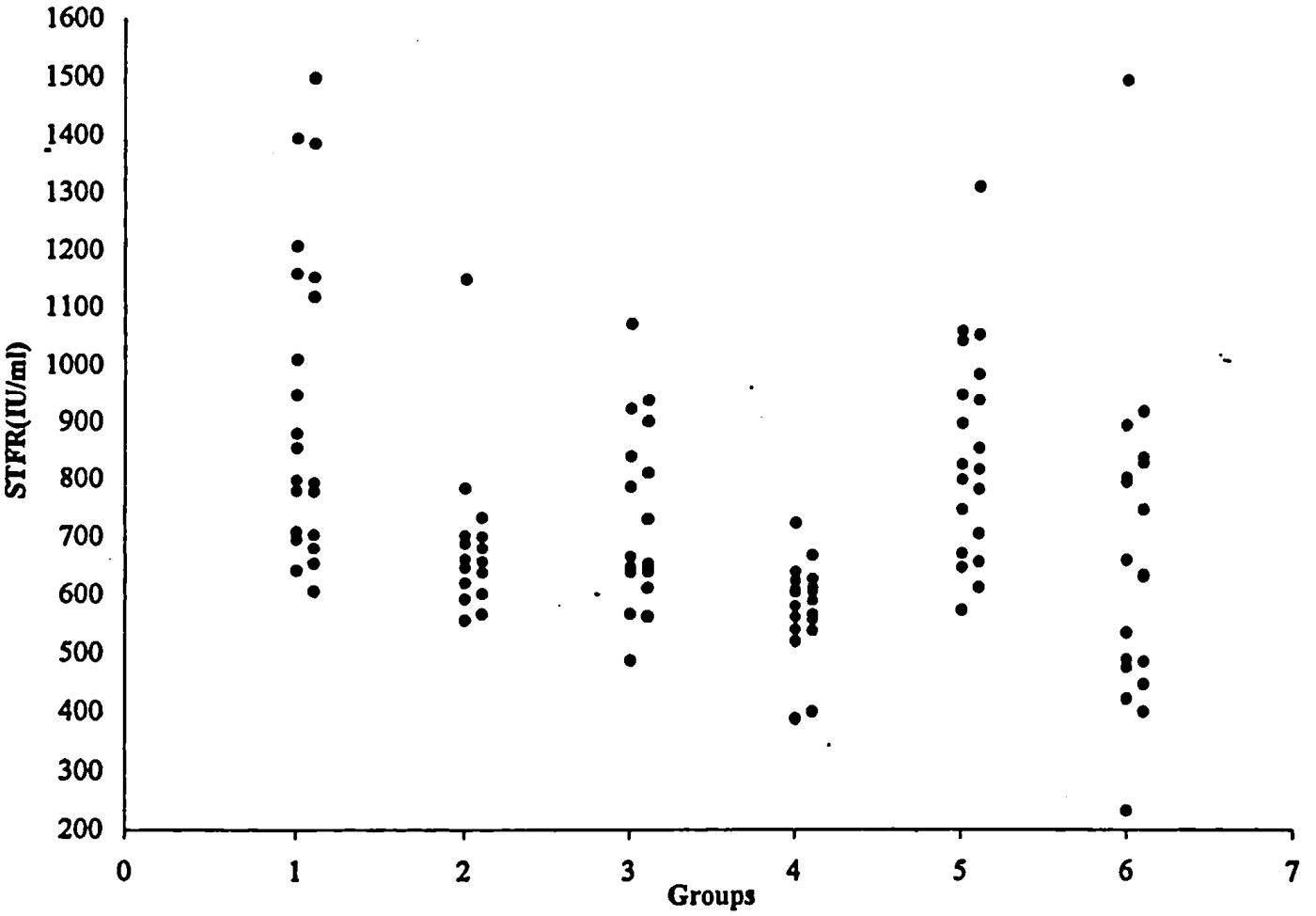

Fig. 1. Results of sTfR levels. Group 1: iron deficiency; Group 2: after iron therapy; Group 3: beta thalassemia trait; Group 4: healthy children; Group 5: newborns; Group 6: mothers of newborns.

Serum transferring receptor levels of children with iron deficiency and with beta thalassemia trait were found to be significantly higher than those of healthy children ($p<0.0001$ and $p<0.001$). Children with iron deficiency showed a greater increase in the levels of sTfR than those with beta thalassemia traits ($p=0.008$). Although sTfR levels of subjects having iron therapy decreased, the levels still remained high compared to controls ($p=0.002$). Newborns had significantly higher levels of sTfR than controls ($p<0.0001$). Although sTfR levels of mothers with iron deficiency were higher than those of mothers having no iron deficiency ($n=10$) ($p=0.009$), there was no difference in the levels of sTfR between newborns of both groups of mothers ($p=0.790$).

A significant negative correlation was found in Group 1 (iron deficiency) between sTfR and hemoglobin, hematocrit, MCV and ferritin levels. This correlation disappeared after iron therapy. sTfR levels of newborns and their mothers showed a correlation with hemoglobin levels and hematocrit but no correlation with iron parameters (serum iron, total iron-binding capacity and ferritin levels). Iron parameters also showed no correlation with hemoglobin levels, hematocrit or MCV in newborns and their mothers.

Discussion

Transferrin receptor is responsible for the transfer of iron bound to the transferrin into the cell. Serum TfR is a truncated from of the intact TfR and can be determined by immunoassay methods[1-3]. sTfR levels increase specifically in tissue iron deficiency and hyperplastic erythropoiesis. Because sTfR is not an acute phase reactant like ferritin, is a more sensitive parameter compared to other iron parameters, and is a more stable analyte against physiologic changes, it has recently become a more popular diagnostic tool[6-9]. Use of sTfR has become wide spread because of its easier determination in a small amount of serum and its close correlation with TfR[1-3].

Generally studies on sTfR have been performed in adults. Little information has been reported on children in the literature. We hope that this study can make some contribution to this issue because the subjects chosen were children. The most comprehensive study so far has been the work of Young et al.[4] on 485 infants investigating normal percentile estimates for sTfR Virtanen et al.[10] reported that sTfR levels were higher in infants and prepubertal children than in adults.

When iron deficiency occurs in tissues, TfR expression increases rapidly. This is achieved directly by the effects or iron regulatory proteins (IRP) and erythropoietin[1,11]. Many studies have shown that in patients with iron deficiency anemia, sTfR levels rise 3 to 5 fold, related to the severity of iron deficiency, whereas in iron overload sTfR levels are normal or slightly decreased[4,6-9]. In the absence of a hyperplastic erythropoiesis, the sTfR is a sensitive marker of early tissue iron deficiency[1]. sTfR reflects the tissue iron requirements. The only parameter reflecting the tissue iron deficiency exactly within the period between the beginning of anemia and consumption of iron stores are sTfR levels. In our study, sTfR levels of children with iron deficiency were also significantly higher than those of healthy children. This marked rise almost recovered after iron therapy.

The amount of TfR on erythroblasts with high iron need and turnover is higher than that of the mature erythrocyte. Because of this, sTfR may be used as a non-invasive method in the evaluation of erythropoiesis[12]. In some studies, decreased levels of sTfR have been reported in cases with decreased erythropoiesis such as in aplastic anemia and renal failure, while an increase in sTfR has been showed in cases with increased erythropoiesis such as in haemolytic anemia and thalassemia[3,13]. In our study, we found higher levels of sTfR in children with beta thalassemia traits than in controls.

Prevalence of beta thalassemia trait is high in the southern and western parts of Turkey like in other Mediterranean countries. Differential diagnosis of iron deficiency from beta thalassemia is essential in these areas. Ferritin levels as well as sTfR levels have to be analysed since sTfR levels are high in both situations. Dimitriou et al.[14] reported then necessity of the use of sTfR-ferritin index for the differentiation of beta thalassemia trait from those with iron deficiency. In that report the sTfR values, but not the sTfR/log ferritin index values, were found increased in children with beta thalassemia trait. However, in our study the sTfR/log ferritin indexes were found increased in children with iron deficiency and with beta thalassemia trait.

Serum transferrin receptor levels of newborns do not change with sex, gestational age or birth weight, and are independent of iron parameters. However, there have been controversial results in these situations. In our study there were no sex differences in sTfR levels of newborns. Rusia et al.[15] found inversely related results between sTfR and serum hemoglobin, MCV, iron, and total iron-binding capacity in newborns. In this study, it was found that sTfR levels of newborns of anemic mothers were increased while iron parameters were normal. Kuiper-Kramer et al.[16] reported that sTfR levels of newborns were independent of iron parameters. Choi et al.[17] showed that iron parameters of newborns were not affected unless mothers had marked iron deficiency. In that study, sTfR levels of newborns whose mothers had iron deficiency without anemia were found to be elevated. In our study, we found increased levels of sTfR in newborns. sTfR level showed correlations with hematocrit and hemoglobin but no correlation with iron parameters in newborns. We found significantly increased levels of sTfR in mothers with iron deficiency compared to others. This finding correlates with the literature showing that the sTfR level is the best parameter reflecting iron deficiency during prenancy[18,19]. However we could not find any difference in the levels of sTfR between newborns of mothers with or without iron deficiency. Although the number of cases in our study was small, these results suggest that iron is consumed for the benefit of newborns.

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Effect of immunotherapy on autoimmune parameters in children with atopic asthma

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There is an increased rate of reported autoantibody production in patients with atopic and nonatopic asthma. The possibility of generating autoantibodies after the induction of immunotherapy can be explained by several mechanisms. One of these is immune deviation from TH2 to TH1 response by the effect of immunotherapy in favor of unregulated response to self-antigens. The other theory is a possible antigenic mimicry enabling autoantibody formation in these patients, Sixty-three atopic asthmatic children were included in the study. The patients were divided into three groups: Group I: patients with atopic bronchial asthma without immunotherapy: Group II: patients receiving immunotherapy for a maximum of 3 years; Group III: patients receiving immunotherapy for 4-5 years. The autoantibodies examined in the study population were anti-nuclear antibody, anti-double stranded DNA, rheumatoid factor, liver-kidney microsomal antibody, anti-mitochondrial antibody, anti-thyroglobulin and anti-microsomal antibody, anti-Smith antibody and lupus anticoagulant. An overall incidence of 17.5% autoantibody positivity was observed in patients, with no statistical significance between the treatment groups. IgG levels were significantly elevated in Group III when compared with Group I. Based on these findings it is suggested, in accordance with other studies, that long-term immunotherapy in the pediatric age group does not cause a significant autoantibody formation other than the overall increased incidence that occurs in asthmatic patients.

Key words: atopic, bronchial asthma, immunotherapy, autoantibodies.

Development of bronchial asthma usually involves a complex cascade of events depending upon the bronchial hyperreactivity and immunological features of the host. T cell derived cytokines are considered to play a key role in the pathogenesis of both humoral and cell-mediated aspects of allergic inflammation[1,2]. There is also a well established association between TH2 type cytokine production and allergic pathology and TH1 type cytokine production with non-atopic responses[3-6]. It is suggested that in intrinsic (non-atopic) asthma, viral airway infection and chronic eosinophilic inflammation of the airways may affect optimal lymphocyte regulation, making the individual more prone to develop autoantibodies[7]. On the other hand, the presence of antigens from microorganisms that share determinants with self-antigens creates the opportunity for antigenic mimicry[1,4,8]. In previous studies, organ and non-organ specific autoantibodies have been observed to be between 41-71% in intrinsic and 21-39% in extrinsic asthma (atopic) in comparison to 11-16% of healthy controls[4,9,10].

The efficacy of specific allergen immunotherapy in selected patients with IgE mediated disease has led to considerable interest in the mechanisms underlying this treatment. One way in which immunotherapy may act is by modifying the T lymphocyte response to subsequent natural allergen exposure. Studies in peripheral blood and target organ exhibit a shift in the balance of T cell subsets away from TH2 (production of IL-4 and IL-5) to TH1 (preferential production of IFN-gamma)[3]. Enhanced TH1 response and autoimmune

activation are more frequently observed in intrinsic asthma rather than in atopic asthma which has TH2 type cytokine profiles. Thus, it can be assumed that immunotherapy which is known to change TH2 T cell profile to TH1 might have profound effects in developing autoantibodies. To our knowledge, in children with atopic asthma receiving immunotherapy studies about developing autoimmunity are insufficient. We therefore planned to examine the effect of immunotherapy on autoantibody generation in atopic asthmatic children.

Material and Methods

Sixty-three children with the diagnosis of atopic bronchial asthma (ABA) were included in the study. The diagnosis of ABA was based upon the history, clinical findings and routine laboratory tests in addition to high total and specific IgE, total eosinophil count and skin test positivity. All the patients had moderate asthma (exacerbation of wheezing more frequent than one a week, low grade degree of wheezing between acute episodes, diminished exercise tolerance, clinically and/or radiographically evident hyperinflation, and evident signs of airway obstruction on pulmonary function tests) on admission.

The study population consisted of 21 female (33.3%) and 42 male (66.7) patients ranging in age from 5 to 20 (12.8±3.4) years. History for rheumatologic diseases, local or systemic infection or treatment with immunosuppressants were reasons for exclusion.

The patients were classified into the following groups: I. Patients with ABA who had not received immunotherapy (n=21) II. Patients with ABA who had received conventional immunotherapy for a period equal to or less than 3 years (n=22) III. Patients with ABA who had received conventional immunotherapy for a period of 4-5 years (n=20). The immunotherapy given to the patients included the allergens to which they were sensitive. The more common ones were house dust mites, and tree and weed pollens for the majority of the patients.

Routine and specific laboratory analyses including autoantibodies performed for all study subjects were as follows: erythrocyte sedimentation rate (ESR) (Westerngreen), >13 mm/h was accepted as high; C-reactive protein (CRP); rheumatoid factor (RF); total immunoglobulins (IgG, IgM, IgA); C3 and C4 (by Beckman, Nephelometer 100 Analyzer); liver-kidney microsomal antibodies (LKM) and anti-mitochondrial antibodies (AMA): indirect immunofluorescence, values above 1/100 were considered as positive (Euroimmune, Germany); anti-thyroglobulin antibody (anti-T) and anti-microsomal antibody (anti-M): values above 1/400 were considered as positive (Thymune M, Murex, U.K.); antinuclear antibody (ANA): indirect immunofluorescence, Hep-2 cells as substrate, values above 1/40 were considered as positive (Meridian Diagnostics Milano, Ital); anti-Smith (anti Sm): ELISA, values above 25 U/ml were considered as positive (The Binding Site, Birmingham, U.K.); anti double stranded DNA (anti ds DNA): ELISA, values above 60 IU/ml were considered as positive (The Binding Site, Birmingham, U.K.); and lupus anticoagulant (LA): qualitative assay, (Staclot LA, France). Mann-Whitney U and chi-square tests were used for statistical analysis.

Results

The mean ages of the patients with respect to their distribution within study groups were 10.6±3.9 years (5-7 years), 118±3.9 years (6-19 years), 13.3±3.5 years (8-20 years) in Group I (ABA patients without immunotherapy), Group II (ABA patients with immunotherapy for maximum of 3 years) and Group III (ABA patients with immunotherapy for 4-5 years), respectively. The ratio of female patients to male patients was 6/15 in Group I, 8/14 in Group II and 7/13 in Group III.

Erythrocyte sedimentation rate (ESR) was found to be mildly elevated (>13 mm/h) in 82.5% (52/63) of the patients, the mean value being 16.3±6.2 mm/h. There were only three children (4.7%) having ESR values exceeding 20 mm/h. However, no significant difference was found in ESR values between the three groups ($p>0.05$).

Serum levels of CRP, C3, C4, IgA and IM were normal in all of the subjects. The difference of age, CRP, C3, C4, IgA and IgM between the three study groups was not statistically significant (Group I vs Group II, Group I vs Group III, Group II vs Group III) ($p>0.05$).

The IgG levels were higher than age-related normal values in nine patients (40%) in Group I, in 14 (63.6%) of Group II and in 18 (81%)

of Group III. IgG values were 1255±410 mg/dl, 1422±411 mg/dl, and 1740±674 mg/dl in these three groups, respectively. These levels were found to be elevated in 41 patients (65%) in the whole study population. Only the difference in IgG levels between Group I and Group III was found to be statistically significant (p=0.044).

The immunotherapy given to the patients with respect to their sensitivity to specific allergens include house dust mites, of which Dermatophagoides farinae constituted 46% (n=29) and Dermatophagoides pteronyssinus 41.2 (n=26) and both were applied to 26.9% (n=17) of the patients. The hypersensitivity to house dust mites was most prevalent when compared with tree pollens given to 28.5% (n=18) of patients and weed pollens given to 28.5% (n=18) of patients.

The percentage of all patients with one or more autoantibody positivity was 17.5% (n=11). These were outlined as: positive reactions for ANA in 3 (4.7%), anti ds DNA in 2 (3.1%), ANA+anti ds DNA in 2 (3.1%), LKM in 1 (1.6%), AMA in 2 (3.1%) and LA in 4 (9.5%) patients (Table I). RF was positive in 3 patients (4.7%). Anti-T/anti-M and anti-Smith were negative in all of the subjects. None of the patients with antibody positivity nor their relatives presented evidence of autoimmune pathology.

Discussion

Significant IgG elevation between the asthmatic children who did not receive immunotherapy (Group I) and those who received immunotherapy for 4-5 years (Group III) was observed. Serum IgG level in Group II (patients receiving immunotherapy for maximum of 3 years) was also higher than in Group I, but the difference was not statistically significant. One of the distinctive features of immunotherapy is its ability to inhibit late-phase responses. During immunotherapy, allergen specific IgG (blocking antibody) increases, but this increase has not been found to be correlated with clinical response[3,11]. In Egeskjold et al's study[12], 87% of atopic patients showed positive reactions for IgG anti-IgG antibodies, which increased to 100% after more than 13 months of hyposensitization program; 7% of the controls exhibited IgG anti-IgG antibodies. Djurup and Maling[13] reported large increases in G1 and G4 antibodies specific for the allergen used during imunotherapy. Although we did not have the chance to measure anti-IgG antibodies and IgG subgroups specific for the allergen, our findings support the data of other studies that have observed elevated IgG values in direct relation with the period of immunotherapy.

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Table I. Patients with Autoantibody Positivity and the Allergens They Received During Immunotherapy

	Autoantibodies						Allergens received by immunotherapy			
Patient group	ANA	Anti dsDNA	AMA	LKM	LA	RF	DP	DF	Weed	Tree
I	+	–	–	–	–	–	–	–	+	–
I	–	–	–	–	+	–	+	+	–	–
I	–	–	–	–	+	–	–	–	–	+
II	–	–	–	–	+	–	–	–	+	–
II	–	–	–	–	–	+	–	–	+	+
II	+	+	–	–	–	–	+	+	–	+
III	–	–	+	–	+	–	+	+	+	–
III	–	–	–	–	–	+	+	+	+	+
III	–	–	+	+	–	–	+	+	–	–
III	–	–	–	–	–	+	+	+	–	–
III	+	+	–	–	–	–	–	–	–	+

ANA : antinuclear antibody.
anti ds DNA : anti double stranded DNA.
AMA : antimitochondrial antibody.
LKM : liver-kidney microsomal antigen.
LA : lupus anticoagulant.
RF : rheumatoid factor.
DP : Dermatophagoides pteronyssinus.
DF : Dermatophagoides farinae.

Group I : Atopic bronchial asthma (ABA) patients without immunotherapy.
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patients. However, no significant difference was found between the three groups, suggesting that the elevation is related with the inflammatory process in atopic bronchial asthma and not with immunotherapy or its duration. As in our study, some authors have reported mild ESR elevations in atopic asthmatic patients[1,14].

The results of our study yielded a 17.5% autoantibody positivity for all patients but we did not encounter a statistically significant difference between the study groups. In a study designed with three treatment groups similar to ours, elevated incidence of ANA (23.3%) in the whole study group was found, but no statistical difference was found between the groups[15]. In Fujimori's study[7], the presence of ANA was evaluated in the sera of patients with atopic asthma, non-atopic asthma, lung cancer and control subjects, and the incidence of positive ANA was found to be 20%, 53%, 30% and 11%, respectively. In Menon's study[4], an incidence rate of 20% of anticytoplasmic antibody, which was statistically significant in relation to controls, was observed in atopic asthmatic adults. LA positivity observed in four of our subjects did not lead to any clinical relevance, although these patients are still being closely followed for possible consequences of LA positivity. Our ANA incidence rate for the study group is in accordance with the ratios attained in the literature, supporting no enhanced risk in respect to immunotherapy, its content or its duration.

Proliferation assays in grass pollen and bee-venom sensitive patients have shown induction of peripheral T cell unresponsiveness to allergen after immunotherapy[3]. Atopic individuals receiving five or more years of hyposensitization with allergenic extracts showed no increased autoimmune, collagen, vascular, or lymphoproliferative disease[16]. Besides these findings, it seems that neither dose nor duration of anti-asthma medication and immunotherapy influences the presence of antinuclear or anticytoplasmic antibodies[4].

In conclusion, immunotherapy still has its promising place in the treatment of atopic asthma of childhood, with almost no specific effect on autoantibody generation other than the well known increased incidence of autoantibody occurrence due to the disease process itself.

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II	–	–	–	–	+	–	–	–	+	–
II	–	–	–	–	–	+	–	–	+	+
II	+	+	–	–	–	–	+	+	–	+
III	–	–	+	–	+	–	+	+	+	–
III	–	–	–	–	–	+	+	+	+	+
III	–	–	+	+	–	–	+	+	–	–
III	–	–	–	–	–	+	+	+	–	–
III	+	+	–	–	–	–	–	–	–	+

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A generally neglected threat in infant nutrition: incorrect preparation of infant formulae

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SUMMARY: Egemen A, Kusin N, Akşit S, Emek M, Kurugöl Z. A generally neglected threat in infant nutrition: incorrect preparation of infant formulae. Turk J Pediatr 2002; 44: 298-303.

Breast milk is the most appropriate food for infants. At least 4-6 months of breast feeding is sufficient for all babies if appropriate growth is monitored monthly. However, for those infants unable to breast-feed sufficiently or at all, formula can be given as an alternative. However, serious health problems such as hypernatremic dehydration, malnutrition, and obesity may develop if powdered formula is not appropriately prepared. In the present study, our aim was to investigate whether or not mothers of formula-fed babies in Özkanlar (İzmir, Turkey) district prepared powdered infant formula appropriately. For this purpose, we visited all (328) families with infants younger than 12 months of age. Forty-two (13%) of these 328 infants were still fed formula. The mothers of the infants were asked to prepare formula for two different meals, and duplicate samples from each prepared formula were taken for the measurement of dry matter. Fifty percent of the mothers diluted formula with 10% more or 10% less water for the second meal as compared with the first meal. Four (10%) mothers diluted formula with 10% or less of the required water, while 27 (64%) prepared formula with 10% or more of the required water. It is concluded that mothers must be informed about the preparation of formula when formula is prescribed for their babies.

Key words: formula, infant nutrition, preparation.

Breast milk is the most ideal food and can solely supply all the biological and physiological requirements of babies during the first four to six months. Although the composition of infant formula is similar to breast milk, it is not a perfect match, because the exact chemical content of breast milk is still unknown and contains unique structures and compounds like living cells, hormones, active enzymes, and immunoglobulins, which cannot be replicated in infant formula. However, developments in technology, industrialization and rapid urbanization have changed the role of women in society, requiring them to be more active in the working environment. As a result, mothers and babies are sometimes forced to stay apart for a major part of the day and thus breast milk is not given to the baby in the required amounts or at optimal frequency to compensate in such situations and in those cases when mothers cannot breastfeed due to severe health problems, infant formulae can be used as the best alternative to breast milk[1-9].

Correct use of formula is as important as the correct choice of formula. It is extremely important to prepare formula according to instructions. For instance, if formula is not prepared in hygienic conditions, diarrhea may develop; if the quality of the water used for the dilution of formula is not good (i.e. if water contains toxic substances or inappropriate mineral concentrations), it may cause serious detrimental effects on the rapidly growing infant[10-14].

One of the most critical aspects of formula preparation is to dilute powdered formula according to instructions on the label of the package. It is especially important not to add more or less water than recommended. Families who are short of money may be tempted to add extra water to make the formula last longer. Formulae are designed to provide energy (about 20 calories per ounce) and nutrients that a baby needs for proper growth. If the formula is too weak, the infant will be underfed and may have

protein energy malnutrition. On the other hand, not adding enough water to formula can result in hypernatremic dehydration, obesity and other potentially serious disorders[15]. The aim of the present study, therefore, was to determine whether or not mothers of formula-fed infants in Özkanlar (İzmir, Turkey) district prepare infant formulae correctly.

Material and Methods

In the Özkanlar district, all families (328) with children aged 0 to 12 months were visited. Among these families, all the formula-fed infants were identified. During a face-to-face interview, demographic characteristics of the families were obtained through a questionnaire, including a detailed enguiry regarding nutrition status of the infants. The mothers of the formula-fed infants were also asked to prepare formula for their babies for two different meals, and samples of the prepared formula were obtained. Duplicated samples were collected from each meal, resulting in a total of four samples per baby. These samples were contained in clean and dry glass bottles, with lids closed, and they were refrigerated until measurement.

In order to measure the amount of dry matter, 5 ml of diluted formula sample was taken into a petri dish that was dried and tared previously. The sample was then cooked in an oven at 100 °C until completely dry and at constant weight. The mass of dry matter in the samples was found by subtracting the weight of the petri dish from the final measurement. This number was then multiplied by 20 in order to find the total dry matter in the 100 ml diluted formula. The dilution difference (technical error) in the duplicate samples taken from the same meal was expressed as percent and was calculated as follows:

$$\frac{A-B}{A} \times 100 \quad \text{where}$$

A=weight of dry matter in the first sample
B=weight of dry matter in the second sample

The mean (±SD) difference in the dry matter content between the two samples collected at the same meal was calculated as 3.9±3.1 % (using absolute values). As this number was an acceptable value, it was considered that the technique used to measure dry matter content of formula was appropriate and further analyses were carried out.

The average of duplicated dry matter measurements for each of two different meals was then calculated, and the dilution difference between the averages of dry weight measurements of two different meals were determined as follows:

$$\frac{C-D}{C} \times 100 \quad \text{where}$$

C= average of the amount of dry matter in the duplicate samples (100 ml) for the first meal

D= average of the amount of dry matter in the duplicate samples (100 ml) for the second meal

In order to determine whether or not mothers diluted the formula according to the instructions, the following equation was used.

$$\frac{E-C}{E} \times 100 \quad \text{where}$$

E=original dry matter content in 100 ml diluted formula written on the label of the package

Results

In the Özkanlar district, of a total of 328 families, 42 babies (13%) were still formula-fed (Table I). Of these infants, 16 were boys (38%) and 26 were girls (62%). The mean age of this group was 5.6±3.3 months (median, 4.0 months).

Table I. Demogrophic Characteristics of the Study Subjects

Characteristics	Value*
Characteristics of the mothers	n=42
• Age (year)	
Mean±SD	26.0±3.8
Min-max	19-36
• Education	
Primary school or less	10 (23.8)
Junior high school	3 (7.1)
High school	14 (33.3)
University	15 (35.7)
• Working/not working	16 (39)/26 (61)
• Economic status of the family	
Low	6 (14.4)
Middle	14 (33.4)
High	22 (52.2)
Characteristics of the infants	
• Age (months)	
Mean±SD	5.6±3.3
Min-max	4.0
• Sex	
Male	16 (38)
Female	26 (62)

* The numbers in parentheses are percent values.

The age of the mothers ranged between 19 to 36 years (mean, 26.0±3.8 years). Mothers were mostly housewives (61%); 33% were high school graduates and 35% university graduates.

While only two (4.8%) babies were solely formula-fed, the others were either fed supplementary foods (35%), supplementary foods and breast milk (40.5%), or breast milk (19%), in addition to formula. Twelve babies (29%) were fed less than 30 ml of formula per kilogram body weight, whereas 10 (23%) were fed more than 90 ml of formula (Table II).

Nineteen babies (45.3%) started to take formula within the first month after birth and nine of them (21.4%) took their first formula on their first day. Thirty-four mothers (81%) indicated that they started formula as a result of doctor's recommendation, whereas six of them (14.3%) started on their own, without seeking outside advice.

In this study, all mothers were questioned regarding where they learned to prepare the formula and it was learned that doctors and midwives provided such information. Twelve (28.6%) mothers used this information, 25 (59.9%) reported that they benefited from instruction on formula packages, 1 (24%) learned from books and television and 2 (4.8%) from their own experiences. All mothers prepared the formulae with boiled and then cooled tap water for every meal.

Fourteen mothers (33.3%) indicated that they kept formula in the fridge once the package was opened, while 28 (66.7%) kept the package at room temperature.

When they were asked how to prepare formulae, 39 (92.9%) indicated that they added the powder formula with a special measurement instrument and 3 (7.1%) said prepared based on visual estimates (Table III).

Table II. Nutritional Pattern of the Infants

Characteristics	n	%
Current nutrition		
• Excusively formula	2	4.8
• Breast milk+formula	8	19.0
• Formula+supplementary foods	14	35.7
• Breast milk+formula+supplementary foods	17	40.5
Daily formula intake of the infants per kg body weight		
• <30 ml	12	29
• 30-59 ml	11	27
• 60-89 ml	9	21
• ≥90 ml	10	23

Table III. Mothers' Knowledge About Preparation and Preservation of Infant Formulae

	n	%
Knowledge from		
• Instructions on the formulae packages	25	59.5
• Doctor	12	28.6
• Doctor+Instructions on the formulae packages	2	4.8
• Own experiences	2	4.8
• Books and television	1	2.4
How to prepare		
• By measuring	39	92.9
• Visual estimate	3	7.1
Evaluation of mothers' explanation		
• Correct preparation	26	61.9
• Incorrect preparation	16	38.1
Preservation of opened packages		
• Refrigerator	14	33.3
• At room temperature	28	66.7

The mean dilution difference between the duplicate samples of the same meal was found as 3.9±3.1. When mean difference ± two standard deviations (10%) was accepted as the limit value, 27 (64.2%) mothers diluted formula more than recommended in the instructions and 4 (9.5%) mothers used less water than required (Fig. 1). Those who diluted formula more than required used an average of 22.2±14.5% more water, while those who diluted less than required used 12.4±13.0% less water. Among those who diluted formula excessively, one mother used 60% more water than required. In contrast, one mother used 43% less water than recommended for the preparation of formula. When all the cases were considered, mothers used inappropriate amounts of water by 20.3±15.1% during the first meal and by 18.9±14.3% during the second meal (mean ± SD).

In order to determine whether or not mothers always diluted formula in a similar manner, the dilution of the formula prepared for two different meals was compared (Fig. 2). The formula prepared for the second meal was more or less diluted than the first meal by 16.2±20.7% (mean ± SD). For one of the mothers, the dilution difference between the first and second meals was 100%.

Discussion

In this study, it was found that 74% of mothers diluted formula incorrectly due to a lack of education regarding infant formula preparation. During a previous research[15], one group of babies had been fed with ready-to-use liquid formula since birth, while another group was fed with powdered formula requiring dilution at every meal. At the end of the sixth month while only 5% of those babies fed with ready-to-use formula weighed more than normal, the weight of 30% of the babies in the other group was higher than normal. The reason for this difference was believed to be the incorrect dilution of formula.

Another interesting result determined in this study was the inconsistent practices of the mothers for the dilution of formula. In other words, one meal might be overdiluted, whereas the next one might be underdiluted, resulting in significant differences, as high as 100% between two different meals.

If an exclusively formula-fed baby is given overdiluted formula, protein energy malnutrition may develop. On the other hand, and particularly if a baby is fed with concentrated formula for an extended time, if extra water is not given to the baby, especially during the hot summer days,

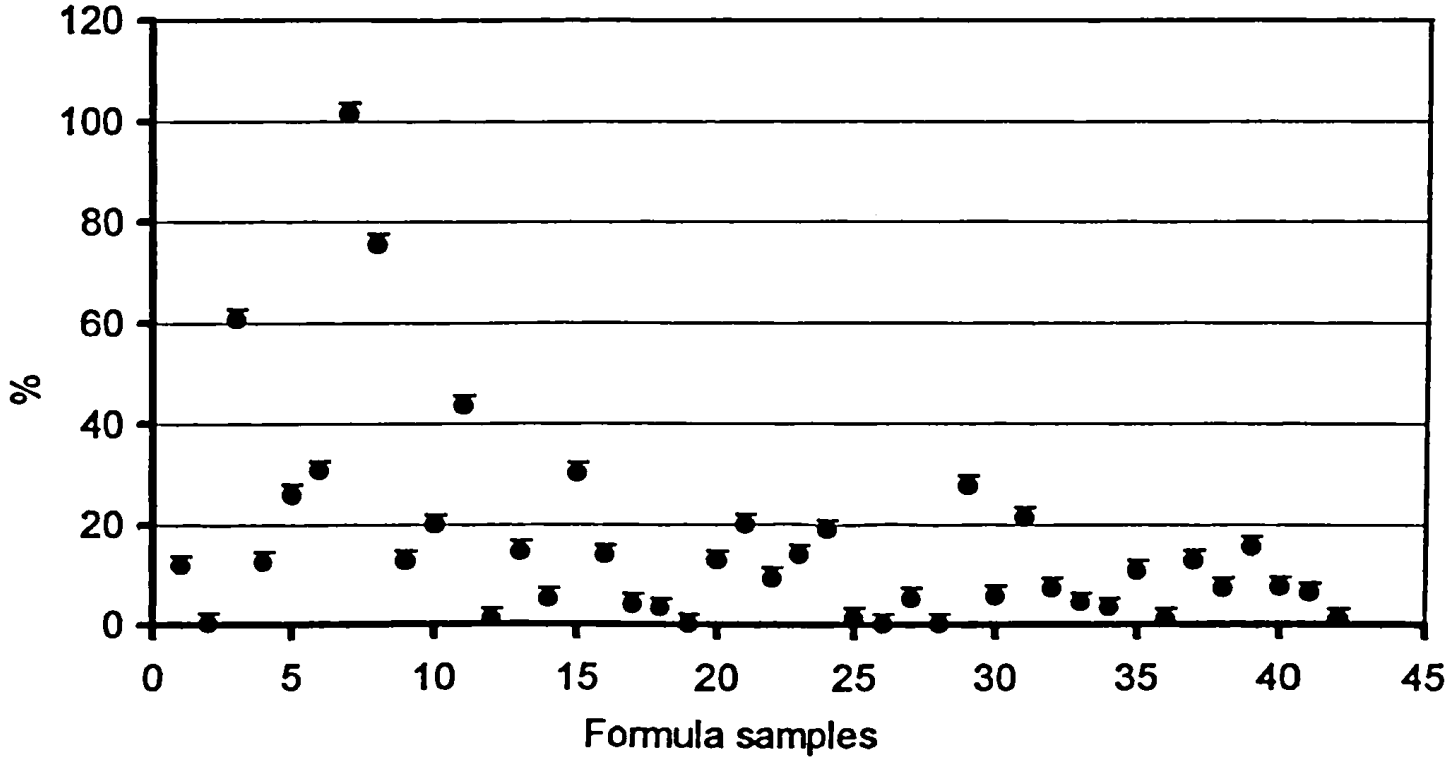

Fig. 1. Difference in the amount of water used for preparation of formula compared with recommended amount of water written on the label of formula package.

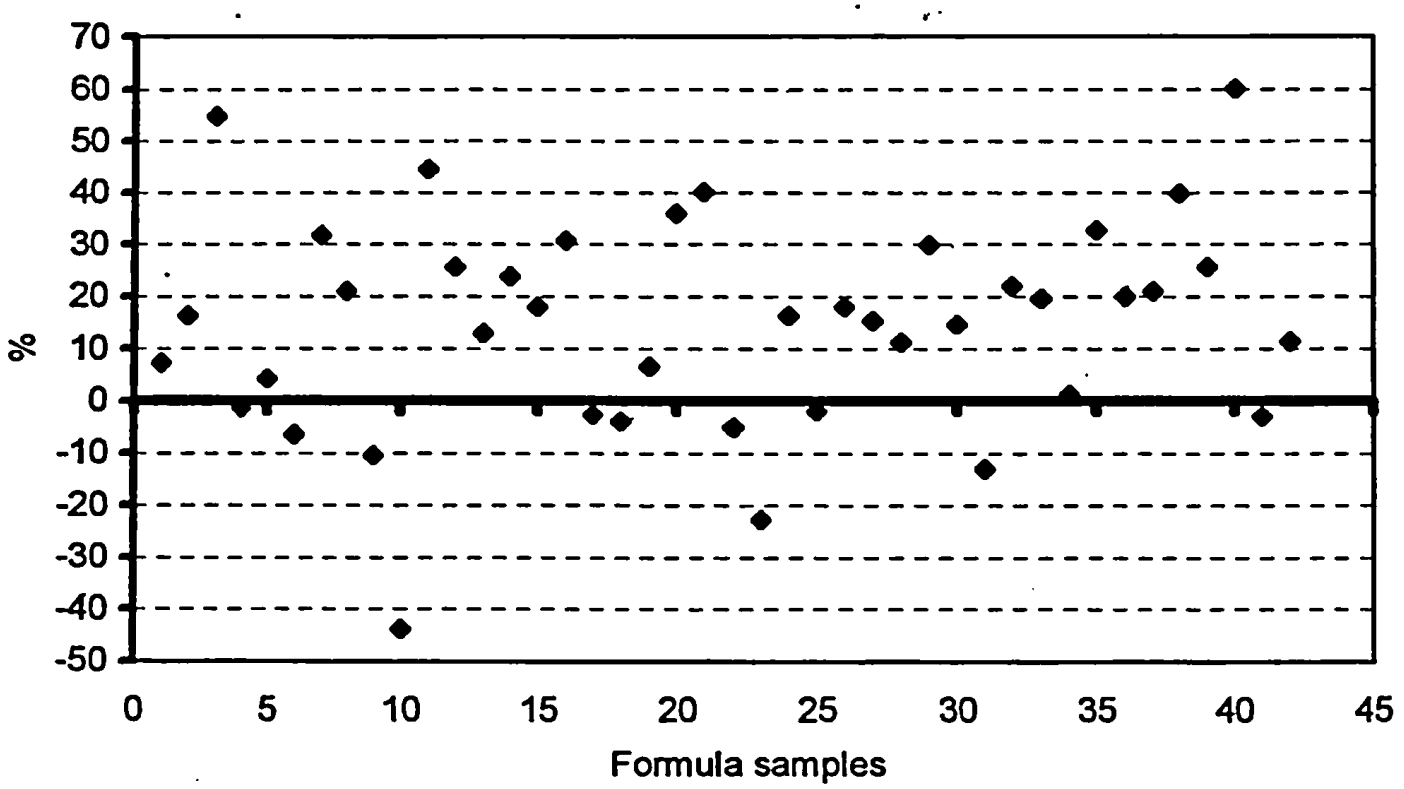

Fig. 2. Dilution differences of the formulae between the first and second meals.

severe health problems such as hypernatremic dehydration or obesity may develop. It may seem contradictroy that although most of the babies included in our study were fed with overdiluted formula none of the babies was suffering from malnutrition. This situation can be explained in two ways. First, only two of the 42 babies were exclusively formula-fed; the others were fed breast milk and/or supplementary foods in addition to formula. Therefore, the amount of formula that the babies were given was relatively low, and only 23% of the babies were fed a daily dose of 90 ml of formula or more per kilogram body weight. Secondly, mothers were inconsistent in their food preparation techniques. Thus, any lack of protein or energy caused by overdilution of formula might be compensated by the overconcentration of formula during the next meal.

In literature from the last 20 years, we could find only one study on the dilution of infant formulae[17]. According to that study, carried out in Australia, 30% of the mothers diluted formula incorrectly. Half of those mothers made serious mistakes, and formula was usually prepared overconcentrated. A majority of the mothers in our study had average socio-economic status, and 69% were high school or university graduates.

During recent years, intensive research has been conducted to adapt several good qualities of breast milk, including immunological characteristics, to formula through biotechnology[18]. However, it should be remembered that no matter how similar formula is to breast milk, the formula will not benefit babies if not prepared correctly. For this reason, pediatricians are responsible for explaining the preparation of formula to mothers at the time they recommend its usage. In this study, 14.3% of mothers indicated that they started to use formula on their own and without a doctor's recommendation, whereas 2.4% followed a pharmacist's advice and 2.4% followed a midwife-nurse recommendation. However, 71.4% of the mothers started formula because they concluded that their breast milk was not enough. For this reason, during routine growth monitoring after birth, evaluations regarding the sufficiency of breast milk of mothers should be made based on the growth curve of their children. Mothers should be fully informed in this regard, and should be prevented from starting formula unnecessarily. Many falsely assume their infant's crying is an indication that their breast milk is not sufficient.

In summary, it should be remembered that breast milk alone can provide all the biological and even physiological needs of babies for four

to six moths. In order not to deprive babies of this unique and useful food, mothers should be fully informed both before and after they give birth. If formula should be used because a mother cannot breastfeed due to health problems or for other reasons, then; mothers should be educated and trained about the preparation and preservation of formula.

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Surgical outcome of congenital valvar aortic stenosis

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Valvar aortic stenosis is a common congenital heart defect for which surgical procedures can be done with low risk except in infants whose conditions are seriously compromised. The purpose of this report was to present our experience with the results of surgical aortic valvotomy for congenital valvar aortic stenosis performed at our hospital.

The study group consisted of 24 patients, 3 females and 21 males, with ages ranging from 1 to 15.5 years (mean age 8.5 years), who underwent aortic valvotomy for valvar aortic stenosis. The case records of all the patients were retrospectively reviewed. They were followed for 1-10.5 years (mean 5.02±2.38 years) after relief of aortic stenosis, and were scheduled for reevaluation.

Sixteen of the 24 patients were recatheterized and 15 (93.7%) were found to have aortic regurgitation on angiography. Peak systolic pressure gradients (mean±SD) were 65.9±19.5 mmHg before and 36.7±14.8 mmHg ($p<0.05$) after the operatilon. Of the 24 patients, 45.8% had a new postoperative diastolic murmur. Twenty patients (83.3%) had residual stenosis and three (12.5%) had recurrent stenosis. Two patients (8.3%) had undergone reoperation six to seven years after the initial aortic valvotomy, and most of the others will require reoperation in the future. No sudden deaths occurred in this series.

Timely relief of obstruction prevents sudden death and produces symptomatic improvement in valvar aortic stenosis, but aortic valvotomy is only a palliative measure.

Key words: congenital heart defect, valvar aortic stenosis, surgical aortic valvotomy.

Congenital aortic stenosis occurs in 3-6% of patients with congenital heart disease[1,2]. Left ventricular outflow tract obstruction is most frequently at the valvular level, being observed in over 50% of the children who initially have findings of aortic stenosis[3,4]. It is a progressive disorder, and the worsening may be due to the fact that the stenosis is becoming more severe. Thickening and increased rigidity of the valve tissue and varying degrees of diminished commissural separation comprise the basic malformation[1]. The occurrence of sudden death in young patients with isolated congenital aortic stenosis is well recognized. Campell[5] reported an annual sudden death rate of 0.9% in children with this lesion.

Incision of congenitally fused aortic valve commissures can be dramatically beneficial in relieving aortic stenosis. Aortic valvotomy under direct vision was first described in 1956[6,7], and accepted as a palliative operation that provides initial benefit by decreasing risk of sudden death, but it is usually followed by an increasing incidence of reoperation. Most reported series conclude that all patients treated with valvotomy will ultimately require aortic valve replacement[8-11]. In the First Natural History Study of Congenital Heart Defects, the incidence of bacterial endocarditis was high for patients with aortic stenosis and it was greater in patients who underwent surgical aortic valvotomy[12].

This study was presented in the Third World Congress of Pediatric Cardiology and Cardiac Surgery held in Toronto-Canada 27-31 May 2001.

To evaluate the effectiveness of the operation, we reviewed our experience with 24 consecutive children who underwent aortic valvotomy. In this study the clinical records of all the patients were retrospectively reviewed, and they were scheduled for reevaluation.

Material and Methods

Twenty-eight patients underwent surgical intervention for congenital aortic stenosis at the İhsan Doğramacı Children's Hospital. One 19-day-old infant died during the operation and three patients aged 21 days, 75 days and 10.5 months died after the operation, yielding an operative mortality of 14%. The remaining 24 patients who had neither septal defect nor additional valvular lesions were included in this series. There were 21 boys and 3 girls whose mean age was 8.5 years (range 1 to 15.5 years) at the time of the operation.

Symptoms were present in three cases of the study group at the last evaluation after surgery. Determination of functional classification in accordance with the New York Heart Association's (NYHA) criteria revealed 21 patients to be in Class 1, two patients in Class III and one patient in class IV (Table I).

Table I. Clinical Features of the Study Patients (Survivors) at the Last Evaluation

Pt no.	Age (yrs)	Sex (M/F)	Follow-up period (yrs)	NYHA class
1	26	M	10.5	I
2	22	M	7.5	I
3	21	M	8.5	I
4	21	M	7	IV
5	19	M	6	I
6	16	M	2	I
7	15	M	7	I
8	14	M	4	III
9	14	M	1	I
10	14	M	1	I
11	13	M	5.5	I
12	13	M	4.5	I
13	12	F	6.5	III
14	12	F	4	I
15	11	M	4.5	I
16	11	M	4	I
17	11	M	5.5	I
18	10	M	3.5	I
19	10	M	4	I
20	9	M	7.5	I
21	9	M	2.5	I
22	8	M	7	I
23	8	F	4	I
24	6	M	3	I

NYHA: New York Heart Association.

Preoperative peak systolic pressure gradients ranged from 45 to 128 mmHg, with a mean value of 65.86±19.49 mmHg. All but eight patients underwent preoperative cardiac catheterisation. Follow-up data by clinical evaluation in our Cardiology Department were obtained for all of the patients from 1-10.5 years (mean 5.02±2.38 years) postoperatively. The follow-up was based essentially on physical examination, electrocardiogram (ECG), chest X-ray and echo Doppler examination. Left ventricular hypertrophy was assessed by ECG using the criteria established by Romhilt and Estes[13]. Cardiomegaly was reported if the cardiothoracic ratio was greater than 0.5.

Sixteen patients were recatheterized. Outflow tract gradients were calculated from left ventricular aortic pullback tracings, and the presence of aortic regurgitation was confirmed or ruled out by aortic root angiography. Associated lesion was present in two patients: one patient (No.9) had aortic coarctation and the other (No.14) had subaortic ridge. Bicuspid aortic valve was present in 14 patients (58.3%) and none had unicuspid valve. Aortic balloon valvotomy was not performed in this series.

An M-mode echocardiogram with two-dimensional echocardiographic guidance was obtained at the mid-left ventricular cavity level. Measurements of left ventricular cavity size, ventricular septum, and posterior wall were made at end diastole and end systole. Offline system was used for calculation of ejection and shortening fraction[14]. Continuous-wave Doppler examination of the aortic valve gradient was performed. The aortic stenosis jet was recorded from suprasternal transducer position. The peak instantaneous echocardiographic aortic valve gradient (echograd) was calculated using the simplified Bernoulli equation[14].

In this study, we used color Doppler echocardiography for non-invasive evaluation of aortic regurgitation. The Doppler examinations were performed with a Toshiba sonolayer SSH-60A Doppler echocardiograph equipped with 2.5, 3.75 and 5 MHz phased-array transducers.

Color Doppler Examination

The regurgitant jet was studied in the parasternal long-axis view. Aortic regurgitation was considered to be present when an abnormal diastolic flow (a mosaic pattern indicating turbulent flow) originating from the aortic valve was visualized in the left ventricle[15-17].

Cardiac Catheterization

The peak to peak gradient across the left ventricular outflow tract (cathgrad) was measured by catheter pullback. The presence and severity of aortic regurgitation was assessed from biplane angiography. Aortic root angiography was performed with injection of 1 ml/kg contrast material in the left and right anterior oblique view. The degree of aortic regurgitation was graded independently from the echocardiographic findings by the method of Grossman[18] as follows:

Grade I, faint opacification of part of the left ventricle which clears with each systole; Grade II, opacification of the left ventricle to a degree less than that in the aorta; Grade III, opacification of the left ventricle equal to that of the aorta; Grade IV, complete dense opacification of the left ventricle in one beat with a constant density greater than that in the aorta.

Mortality was defined in terms of perioperative and late death. Perioperative death occurred within 30 days of valvotomy.

Statistical Methods

Normally distributed data are presented as mean±standard deviation. Non-normally distributed data are described as median and range. Comparison of transaortic valve gradient before and after aortic valvotomy was performed using t-tests for paired samples. A p value of <0.05 was considered significant.

Results

Reintervention included reoperation in two patients (8.3%). One patient had severe aortic regurgitation, and an aortoplasty was performed seven years after surgical aortic valvotomy, while the other patient required aortic valve replacement six years after initial surgical intervention for asymptomatic restenosis.

The ECG was normal in 15 patients, revealed a strain pattern in two patients, and left ventricular hypertrophy without strain in nine patients. Of these, one had ventricular extrasystoles and Holter monitoring had demonstrated the presence of multiple couplets and triplets, therefore antiarrhythmic therapy was prescribed. Radiographic measurement revealed 17% with cardiomegaly, and 29.1% had dilatation of the ascending aorta.

Hemodynamic Data

Heart catheterisation was done preoperatively in 18 and postoperatively in 16 patients of the study group. The left ventricle could not be entered preoperatively in nine and postoperatively in one patient. The mean preoperative aortic cathgrad of 78.2±33.13 mmHg (range 45-128 mmHg, n=9) was decreased to 35±8.98 mmHg (range 20-55 mmHg, n=16) after surgery.

Aortic regurgitation assessed angiographically before aortic valvotomy (n=18) was of grade I in 4, II in 1, IV in 1 patient, and absent in the remainder (67%). Postoperative assessment (n=16) revealed aortic regurgitation of grade I in 2, II in 3, III in 7, IV in 3 and absence in 1 patient (Table II).

Table II. Assessment of Aortic Regurgitation (AR) and Transaortic Gradient Before Aortic Valvotomy and at the Last Follow-up

	Preoperative		Postoperative	
No.	AR	Aortic gradient	AR	Aortic gradient
1	No	70*	No	10*
2	No	67**	No	22*
3	No	115*	No	70**
4	No	60*	IV	38
5	No	51**	III	35
6	No	80**	No	40*
7	I	50	III	30
8	No	70**	I	30*
9	No	46**	II	30
10	II	65	II	50*
11	No	68*	III	35
12	No	68**	No	45*
13	No	128	II	24*
14	I	45	II	38
15	No	90	III	35
16	No	82	III	30
17	No	100	I	55
18	I	89**	III	30
19	I	114	II	40*
20	No	80**	II	32
21	No	80**	I	35
22	No	60*	III	50
23	No	76*	IV	20
24	IV	65	IV	25

* Gradient as detected by echocardiography because cardiac catheterization was not performed.

** Gradient as detected by echocardiography because left ventricle could not be entered during catheter.

No: aortic regurgitation is not evident.

Echocardiographic Findings

All of the survivors were assessed echocardiographically during follow-up, and the mean echograd was 36.65±14.78 mmHg (range 20-70 mmHg), whereas preoperative mean

echograd was 65.86±19.49 mmHg (t=6.66, P=0.000). Figure 1 demonstrates the transaortic gradient assessed either by echocardiography or cardiac catheterisation before aortic valvotomy and at last evaluation after the surgical intervention.

The recurrence of significant stenosis was evident in three patients (No: 3, 16, 17) after a follow-up period of 8.5, 4, and 5.5 years, respectively. In this series, restenosis rate was found to be 12.5%. However, 20 patients had residual aortic stenosis of mild-to-moderate degree (systolic gradient between 20 and 50 mmHg), and residual gradient was absent in only one patient (4%).

Postoperative aortic regurgitation graded by color Doppler echocardiography was mild in 6, moderate in 10, severe in 1 and absent in the rest. There was agreement between angiographic and color Doppler assessment of aortic regurgitation in 14 of 16 cases (Table III). Echocardiography performed before the initial intervention showed aortic regurgitation in four of these patients. In this study, approximately 25% of patients had aortic regurgitation before surgical valvotomy, whereas postoperative aortic regurgitation was evident in 71% of these patients (Fig. 2). Although aortic regurgitation causes left ventricular volume overload that may result in left ventricular dysfunction, this was not evident in this review.

Figure 3 illustrates the number of cases that had progression or new development of aortic regurgitation postoperatively.

Table III. Echocardiographic and Angiographic Evaluation of AR in the Study Group

Case no.	Color doppler	Aortography
1	NO	–
2	NO	–
3	NO	NO
4*	III	IV
5	III	III
6	NO	–
7	III	III
8	I	–
9	II	II
10	II	–
11	III	III
12	NO	–
13	II	–
14	III	II
15	III	III
16	III	III
17	NO	I
18	III	III
19	II	–
20	I	II
21	NO	I
22	III	III
23	IV	IV
24	III	IV

*Aortoplasty was performed due to severe aortic regeurgitation which developed after valvotomy.

NO: aortic regurgitation (AR) is not evident.

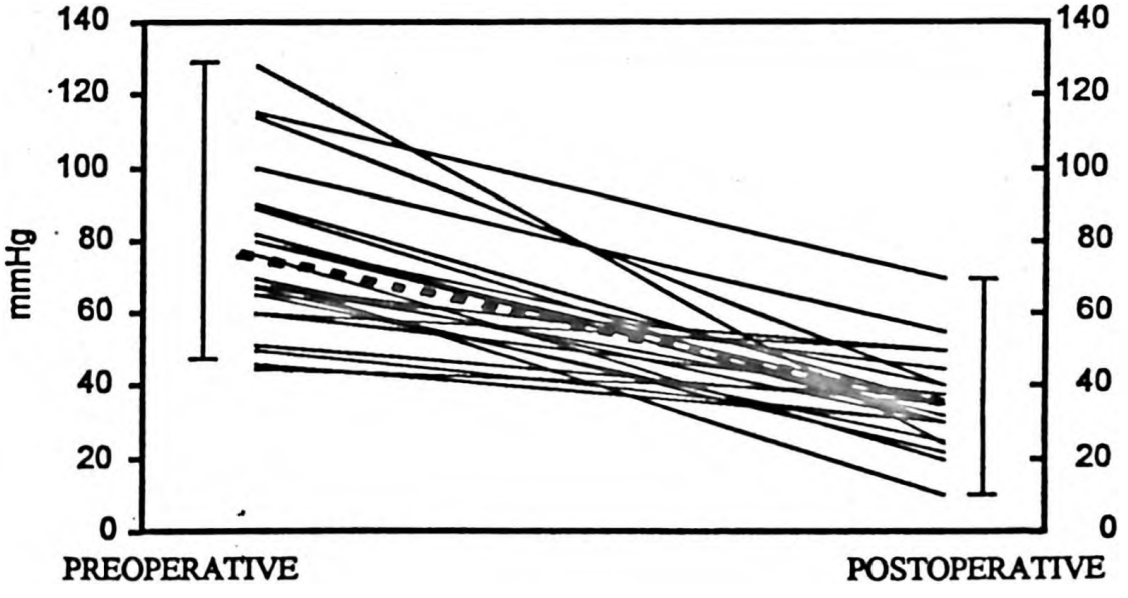

Fig. 1. Assessment of transaortic gradient before and after surgery at the last follow-up.

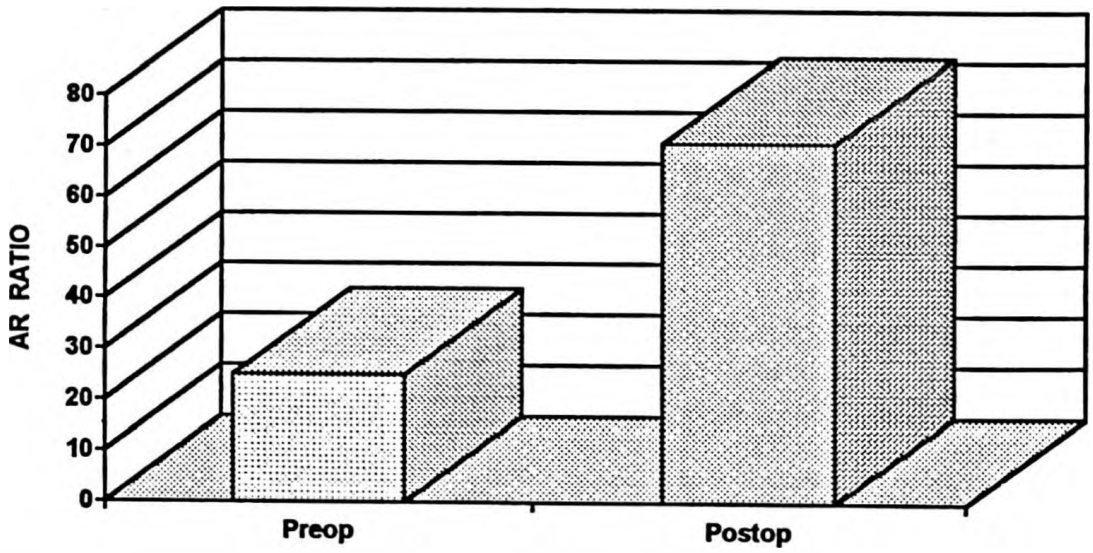

Fig. 2. The percent of aortic regurgitation (AR) detected by echocardiography or aortography after valvotomy.

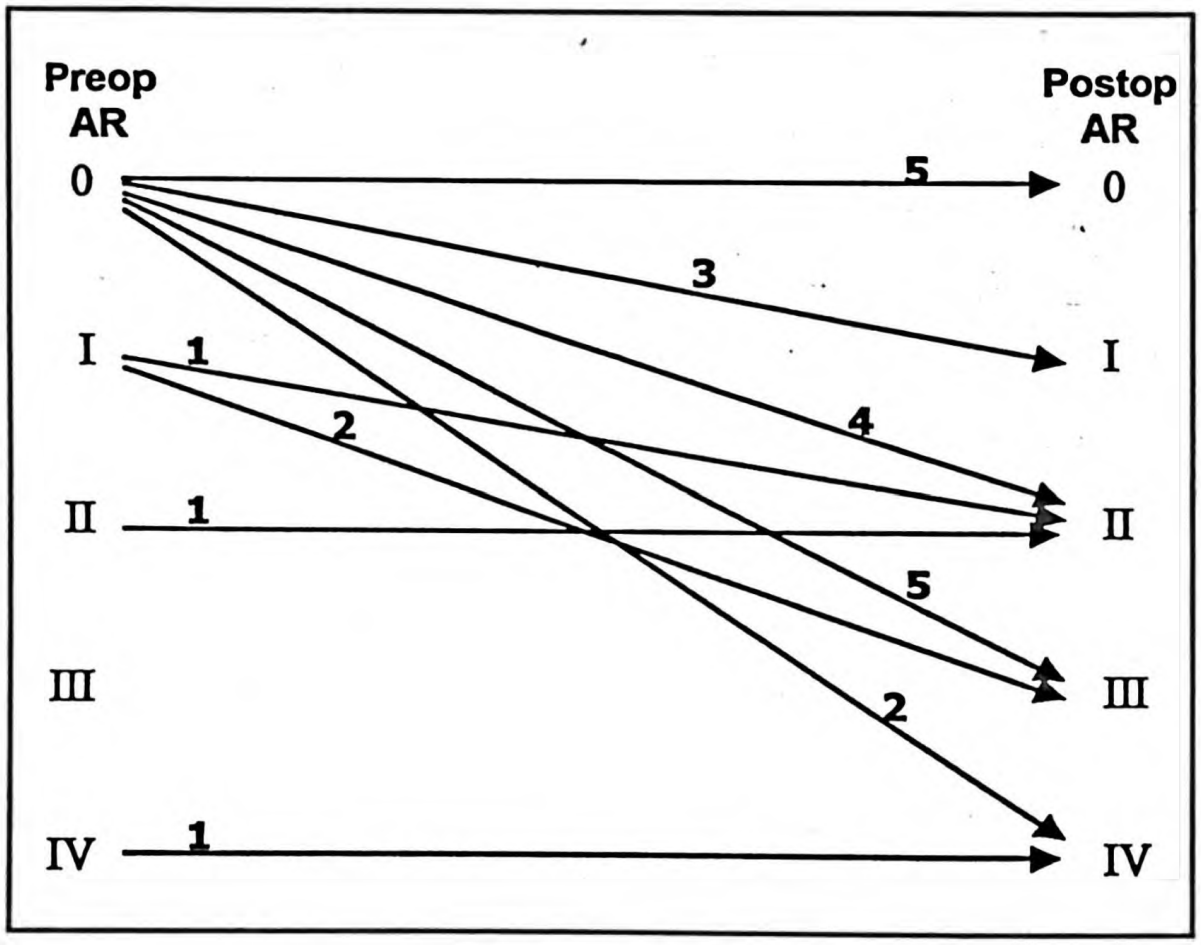

Fig. 3. Evaluation of aortic regurgitation (AR) among the study group during preoperative and postoperative period at the last evaluation (the number of cases is shown).

Discussion

Sudden death in children with congenital aortic stenosis constitutes the major indication for hemodynamic study and surgical intervention. Sudden death has been reported to occur 1-19% of these patients[19-20]. The absence of sudden death in this study confirms the safety and timely relief of aortic stenosis. Although open aortic valvotomy is well established with a low surgical mortality, it is still regarded as palliative therapy because most patients require reoperation, since aortic regurgitation may

occur and restenosis is common. Lababidi and associates[21] first reported that percutaneous balloon aortic valvuloplasty might be an alternative method of treating aortic stenosis by minor tearing of the aortic commissures and stretching of the valve leaflets. Others have also reported that valvuloplasty can successfully treat valvular aortic stenosis in children[22-23].

Reports of surgical results have indicated that reoperation will be necessary in a significant number of patients[8-11]. Two of our 24 patients (8.3%) have, thus far, needed reoperation six to seven years after the initial operation, and most of the other patients are likely to need further surgery in the future. Most of these reoperations will be for cardiac decompensation secondary to iatrogenic aortic regurgitation, residual stenosis or recurrent stenosis. Postoperative reductions in aortic valve gradient were significant on paired testing, and mild-to-moderate residual aortic systolic pressure gradient (20-70 mmHg) was recorded in 95.8% of this series. The stiffness and abnormal configuration found in most of these valves may account in part for the frequency of residual gradients[24].

Postoperative gradients of 50 mmHg or greater predispose patients to sudden death. Hence, annual non-invasive testing is recommended for patients with either residual stenosis or aortic regurgitation[11]. Aortic stenosis is a progressive disease[25], and progression of the obstruction is a well known late complication in postsurgical patients[4]. This was also observed in this study. The reason for the commonly observed progression of disease may lie, in part, in the configuration of the valve and the degree of valve dysplasia[10]. In our series, postoperative regular echocardiographic examinations revealed that the transaortic gradient increased significantly in three patients (No. 3, 16, 17); one of them required aortic valve replacement, but his parents would not give their consent for the operation.

Twenty-one (87.5%) of the study patients were clinically asymptomatic at postoperative follow-up despite well known iatrogenic incompetence, and residual and recurrent stenosis. Similar findings have been reported by others[20]. One symptomatic patient (No, 4) with effort intolerance was restudied, and angiography showed severe aortic regurgitation. In this case, a second operation (aortoplasty) was performed. The other two patients (No. 8, 13) with angina had residual gradients of 30 and 24 mmHg, respectively, with evidence of mild-to-moderate aortic regurgitation.

The most important technical complication of aortic valvotomy is the production of aortic regurgitation. Approximately 8% of preoperative patients had an early diastolic murmur of regurgitation, whereas 46% of our patients developed a new diastolic murmur postoperatively. Similar figures have been noted by others[26,27]. Postoperative catheterisation was performed in 16 (66.6%) patients and 93.7% of them had angiographic evidence of aortic regurgitation. In 63%, the degree of aortic regurgitation was found to be moderate or severe. Of these, two patients showed progression of preoperative mild aortic regurgitation to moderate degree postoperatively at the last follow-up. Evaluation of all patients after valvotomy by Doppler echocardiography or angiography revealed an aortic regurgitation ratio of more than 70%. The high incidence of aortic regurgitation reflects a surgical aim to relieve stenosis maximally and prevent sudden death with acceptance of some regurgitation. In contrast, Chartrand and colleagues[28] reported much lower figures of postoperative aortic insufficiency (12%) and they postulated that this finding was related to the careful limitation of the commissurotomy. On the other hand, 24% of their patients required a second intervention due to recurrence and progression of valvular stenosis, and the rate of sudden death in their series was 3% at the postoperative follow-up.

Bacterial endocarditis has been considered an important risk in the setting of aortic stenosis. Gersony and associates[12] reported that the incidence of bacterial endocarditis in patients with aortic stenosis managed surgically was greater than seen in those managed medically, and they suggested that development of aortic regurgitation and persistence of some postoperative gradient might have contributed to the development of bacterial endocarditis. They concluded that the increased incidence in patients with aortic stenosis after surgical valvotomy was a function of severity and not a function of surgery[12]. However, in our study group bacterial endocarditis was not observed in any patient. Open aortic valvotomy for relief of congenital aortic valve obstruction in children and adolescents is associated with a low

mortality[4,24]. The results obtained in this group of patients indicate that surgical aortic valvotomy can be performed with an acceptable operative risk. However, surgical repair of aortic stenosis is associated with a high risk in infants, because they have a more severe form of the disease[29,30]. The low operative mortality and long-term relief of stenosis and symptoms provided by open aortic valvotomy in patients aged more than one year establishes it as the standard procedure. However, aortic insufficiency in these patients is progressive, and valve replacement may eventually be required. Aortic insufficiency was the indication for valve replacement noted by Sandor and his associates[20]. They concluded that reoperation had a higher risk and that results were not as satisfactory as those for the first operation.

This natural history of surgical aortic valvotomy provides a framework for comparison with long-term results of balloon aortic valvotomy. Gatzoulis et al.[31] compared the contemporary results of balloon dilatation and surgery for valvular aortic stenosis in infants and children. They considered both balloon and surgical valvotomy palliative forms of treatment, as the majority of these patients are likely to require aortic valve replacement later in life. Balloon dilatation offers the additional attraction of deferring the definitive surgery and preserving the surgical field, and the hospital stay is significantly shorter in the balloon dilatation group, with obvious financial benefits. Justo et al.[32] compared the degree and rate of progression of aortic regurgitation, relief of obstruction, complications and mortality after balloon aortic valvotomy versus surgical valvotomy. Their results suggested that progression of aortic regurgitation, and acute and late reduction of gradients were similar for both approaches.

We conclude that although timely relief or left ventricular outflow tract obstruction in children prevents sudden death and provides excellent symptomatic relief, the operation is only palliative. The common occurrence of postoperative aortic incompetence, residual aortic stenosis, persistence of cardiomegaly and ECG abnormalities, and the frequent need for reoperation demonstrate that some congenitally deformed valves cannot be satisfactorily reconstructed for the long-term with standard operative techniques. The goal of aortic valvotomy is to facilitate the child's growth to the point where a prosthetic valve may be inserted.

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Sinus node dysfunction in children and adolescents: treatment by implantation of a permanent pacemaker in 26 patients

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Sinus node dysfunction has been reported rarely in pediatric patients with structurally normal hearts. It has been diagnosed with increasing frequency in children and young adult patients with congenital heart defect, especially in patients who have undergone corrective cardiac surgery related with atrial tissue.

Between 1984-1999, 26 patients who were under 22 years of age underwent implantation of a permanent pacemaker for treatment of sinus node dysfunction at our medical center. This subset of patients represents 18.5% of all patients who required permanent pacemakers during this time. The mean age of the 17 male and 9 female patients at initial implantation was 9.2±6 years (range, 0.5 to 22 years). Of the 26 patients, 18 (69%) had associated cardiovascular disease and in 11 (34.6%) patients, sinus node dysfunction developed after a cardiac operation.

The patients were followed up for a total 1,227 (47±45, range 2-176, median 34) pacing months. All symptomatic patients noted a resolution of symptoms after pacing had been performed, and they remained free of symptoms at the latest follow-up examination. Mean acute pacing thresholds and mean latest pacing thresholds for the endocardial atrial and ventricular leads, mean acute impedance and mean latest impedance for the endocardial atrial and ventricular leads and mean acute p wave voltage and the latest p wave voltage did not differ significantly.

In this report, we review our experience in children who required implantation of a permanent pacemaker for treatment of sinus node dysfunction during a 15-year period.

Key words: sinus node dysfunction, children, adolescents, permanent pacemaker implantation.

Sinus node dysfunction (SND) that necessitates permanent pacemaker therapy is much less common in children. The diverse clinical and electrocardiographic manifestations of this disorder were first described in adults[1]. But, it has been diagnosed with increasing frequency in children and young adult patients with congenital heart defect, especially in patients who have undergone corrective cardiac surgery related with atrial tissue. SND has been reported less frequently in pedilatric patients with structurally normal hearts[2].

In this report, we review our experience in young patients who required implantation of a permanent pacemaker for treatment of SND during a 15-year-period.

Material and Methods

Between 1984-1999, 26 patients who were under 22 years of age underwent implantation of a permanent pacemaker for treatment of SND at our medical center. The total number of patients with permanent cardiac pacemaker implantation performed at our institution during this period was 140. This subset of patients represents 18.5% of all patients who required permanent pacemakers during this time. The mean age of the 17 male and nine female patients at initial implantation was 9.2±6 years (range 0.5 to 22 years).

The diagnosis of SND was based on the following electrocardiographic findings: 1) sinus pause or arrest for more than two seconds, 2)

sinus bradycardia (less than appropriate for age) 3) severe sinus dysrhythmia 4) slow escape rhythm 5) sinoatrial exit block (2° type I and II) 6) the bradycardia-tachycardia syndrome 7) sinus node re-entry tachycardia and 8) atrial muscle re-entry tachycardias.

The initial symptoms or signs of the patients were fatigue in 10, palpitation in four, syncope in four, dizziness in one and breath-holding spell in one. Six of the patients were asymptomatic at the time of pacemaker implantation. Indications for permanent pacing were SND with correlation of symptoms during age-inappropriate bradycardia in 22 patients and bradycardia-tachycardia syndrome with the need for long-term antiarrhythmic treatment other than digitalis in four patients. The 22 patients' Holter results were sinus arrest in six, pause more than 2.5 sec in six, sinus bradycardia + pause in four, sinus bradycardia in three, and atrial flutter/fibrillation in three (Table I).

Of the 26 patients, 18 (70%) had associated cardiovascular disease (Table I). Transposition of great arteries (TGA) was encountered in four patients. In 11 of 26 (34.6%) patients, SND developed after a cardiac operation. Surgical procedures included Mustard or Senning in four patients, Fontan operation in one, closure of the secundum atrial septal defect in five, and correction of endocardial cushion defect in one. Other surgical procedures, not strongly related with sick sinus syndrome, included repair of coarctaion of aorta, valvuloplasty for stenosis of aorta, closure of ventricular septal defect and resection of subaortic ridge, each in one patient. Eight (30%) patients had no cardiovascular abnormality.

Statistical analysis was performed using paired t tets. A p value <0.05 was considered significant.

Results

The 26 patients were followed for a total of 1,227 months (47±45, range 2-176,

Table I. Clinical and Pacemaker Data of Patients

Case	Associated cardiac disorders	Age at implantation (y)	Initial symptom or signs	Holter	Follow-up (m)	Mode	Medication
1 (YB)	None	12	Bradycardia	Sinus arrest	124	VVIR	
2 (MB)	None	2	Syncope	Sinoatrial block	176	DDDR	
3 (İK)	None	8	Bradycardia	Sinus bradycardia	110	VVIR	
4(İT)	None	11	Fatigue	Pause	101	VVIR	sotalol
5(AHE)	None	12	Syncope	S. Brady+Pause	28	VVIR	
6 (OÇ)	None	2	Breath-holding	S. Brady+Junc. rhythm	12	VVI	
7 (ZÖ)	None	0.5	None	Sinus bradycardia	37	VVI	
8 (KÇ)	None	8	None	Brady-Tachy snd	67	VVIR	
9(MEG)	TGA (Senning)	1	Bradycardia	Sinus arrest+pause	9	AAI	
10 (EK)	TGA-Mustard	21	Bradycardia	S. Brady+Pause	2	AAIR	
11(AÇ)	TGA (Senning)	0.75	Braycardia	Sinus arrest+pause	22	VVIR	
12 (AD)	TGA (Senning)	4	Fatigue	Atrial fibrillation	27	VVIR (Ep)	Quinidine, digoxin
13 (Mİ)	TA (Fontan)	15	Palpitation	Brady-Tachy snd	49	VVIR (Ep)	
14 (HK)	Closure of ASD+TVR	14	Palpitation	Brady-Tachy snd	58	VVI (Ep)	Amiodarone, digoxin coumadin
15 (SKo)	Closure of ASD+Repair of mitral cleft	11	Palpitation	Atrial Flutter	6	AAIR	
16 (AA)	Closure of ASD	8	Palpitation	Atrial fibrillation	120	VVIR	
17 (AI)	Closure of ASD	16	Syncope	Sinus arrest, J. rhythm	12	AAIR	
18 (HS)	Correction of ECD	8	Dizziness	Pause, Junc. rhythm	12	VVIR	
19 (SKr)	Closure of ASD+MVR	16	None	Pause	44	DDDR	
20 (SG)	VSD+SAS repair	9	None	S. Brady+Pause	28	AAIR	
21 (EO)	Aort coarctation repair	10	Fatigue	Sinus arrest	62	VVIR	
22 (HY)	AS operation	22	Syncope	Pause	2	AAIR	
23 (TK)	Myocarditis	12	Fatigue, CHF	Pause	39	DDDR	Digoxin
24 (SK)	Dilated cardiomyopathy	2	Fatigue, CHF	Brady-Tachy snd	41	VVI	Quinidine, digoxin.
25 (BD)	PFO+PSSVC	6	Fatigue	Pause	31	AAIR	
26 (MU)	Bicuspid Aorta	8	Bradycardia	S. Brady+Pause	8	VVIR	

TGA: transposition of great arteries, TA: tricuspid atresia, ASD: atrial septal defect, TVR: tricuspid valve replacement, ECD: endocardial cushion defect, MVR: mitral valve replacement, Subao: subaortic, PFO: patent foramen ovale, PLSVC: persistant left superior vena cava, CHF: congestive heart failure, S. Brady: sinus bradycardia, Brady-Tachy: bradycardia-tachycardia, Junc: junctional, VVIR: single chamber ventricular rate responsive pacemaker, DDDR: dual chamber rate responsive pacemaker, VVI: single chamber ventricular pacemaker, AAIR: single chamber rate responsive atrial pacemaker, Ep: epicardial, AS: aortic stenosis, Snd. sinus node dysfunction, AAI: single.

median 34). All symptomatic patients noted a resolution of symptoms after pacing had been performed, and they remained free of symptoms at the latest follow-up examination.

Twenty-three patients received the following transvenous pacing system: ventricular demand (VVI, VVIR) 12 patients (46%); atrial demand (AAIR) 8 patients (30%); and dual chamber (DDDR) 3 patients (11%) (Table I). Eight of 11 atrial leads (72%) and six of fifteen ventricular leads (40%) had screw-in mechanism. The remaining leads had tined fixation mechanism. There was no malfunction of leads.

Five patients initially received ventricular demand epicardial system. Two of them had high myocardial stimulation threshold and high lead impedance that necessitated replacement via transvenous approach. The remaining epicardial systems were good.

There was no atrial sensing or capture problem in atrial and dual chamber pacing systems. Atrioventricular synchronization was good in both groups.

Mean acute pacing thresholds during implantation and mean latest pacing thresholds for the endocardial atrial leads were 1.17±0.45 V and 1.65±0.75 V, respectively ($p>0.05$). Mean acute pacing thresholds and mean latest pacing thresholds for the endocardial ventricular leads were 0.98±0.45 V and 0.97±1.3 V, respectively ($p>0.05$). Mean acute impedance and mean latest impedance for the endocardial atrial leads were 560±177 Ohm and 591±165 Ohm, respectively ($p>0.05$). Mean acute impedance and mean latest impedance for the endocardial ventricular leads were 552±178 Ohm and 827±937 Ohm, respectively ($p>0.05$). Mean acute p wave voltage was 1.9±1.3 mV and the latest p wave voltage was 1.2±0.6 mV ($p>0.05$) (Table II).

Table II. Acute and Latest Pacemaker Measurement with Telemetry

	Implantation	Latest
Threshold		
Atrial leads	1.17±0.45 V	1.65±0.75 V
Ventricular leads	0.98±0.45 V	0.97±1.3 V
Impedance		
Atrial leads	560±177 Ohm	591±165 Ohm
Ventricular	552±178 Ohm	927±937 Ohm
P wave voltage	1.9±1.3 mV	1.2±0.6 mV

All but two of our patients were alive and asymptomatic. The deaths were not thought to be pacemaker related. The first patient died suddenly at home. He had a transvenous pacemaker system after operation for atrial septal defect. The second patient was operated for TGA and epicardial pacemaker system was removed four years after operation. He had taken quinidine and digoxin for atrial fibrillation with rapid ventricular response. The cause of death could have been either medication or life-threatening dysrhythmias.

Discussion

Sinus node disease has many names, all of which describe the same set of syndromes. Sinoatrial node disease is probably the most accurate, whereas sick sinus syndrome is possibly the most memorable. This condition is defined as an affliction of the sinoatrial node that either prevents impulse generation or prevents or delays the conduction of sinoatrial impulses to the surrounding atrial tissue. This affliction may be a pathologic process in or around the sinoatrial node, or it may be a pathophysiologic phenomenon of abnormal function of the autonomic nervous system that adversely influences impulse generation within the node or conduction out of it[3].

The clinical manifestations of SND are related directly to age, the function of the remaining conduction system and the underlying hemodynamic state. Poor feeding, lethargy, or signs of congestive heart failure may be associated with severe bradycardia in infats. In older children, bradycardia may manifest as general fatique, the inability to maintain the same level of activity as peers, or increased sleep requirement with or without change in activity. Fatigue was the most common sign in our patients. Syncope and palpitation were found in eight patients. Dizziness and syncope are difficult to detect in infants and young children. Also, it is imperative to evaluate the patient's rhythm when a child presents with unexplained seizures[4].

Although several types of classification of SND have been offered, the causes of SND in children are best classified as either nonsurgical or surgical[5]. When no other cause is found, SND is named as idiopathic. Thirteen of our patients were idiopathic. Five of 13 patients had associated cardiac disease not strongly related

with sick sinus syndrome. Familial occurrences have been reported but are probably uncommon[6]. Two of our patients were siblings. Acquired or familial myocardial diseases such as cardiomyopathies and inflammatory or ischemic diseases encompass a wide range of possible causes of SND[4]. One of our patients had dilated cardiomyopathy and another had viral myocarditis. Medications, particularly antiarrhythmic drugs, are an important cause of SND in children. Two of our patients with brady-tachycardia syndrome received antiarrhythmic medication.

Sinus node dysfunction occurs after several types of surgical procedure for congenital heart disease. Pathologic and electrophysiologic correlations have revealed that incisions, sutures, and progressive fibrosis in then area of the sinus node and sinus node artery are definite causes of SND in children who have undergone surgery for congenital heart disease. But, SND has also been found before operation in patients with congenital heart disease. The incidence is highest in patients who have undergone atrial repair (Mustard or Senning) for transposition of the great arteries[7]. Five of our patients who had undergone atrial repair (Mustard or Senning) for transposition of the great arteries received permanent pacemaker. Although patients with secundum atrial septal defect may have pre-existing SND, postoperative SND may also be found[8]. Five of our patients with secundum atrial septal defect had postoperative SND.

The true incidence of sudden death relative to SND is unknown. The problem of documentation relates to the distinct possibility of other life-threatening arrhythmias in the same patient. Flinn et al.[9] showed that the incidence of sudden death was 2.5% in 372 patients who had undergone Mustard repair for TGA; many of them had SND Gelatt et al.[10] reported that sinus rhythm was present in 77% at five years and in 40% at 20 years from records of 534 children who underwent the Mustard operation Kirjavainen et al.[11] showed that the probability of staying in sinus rhythm was 34% in patients with simple TGA and 7% in patients with complex TGA after Senning operation.

The diagnosis of SND can be suspected on the basis of a careful history and examination of the ECG. Yabek et al.[12] reported 74% of 30 children with various underlying heart diseases who were asymptomatic from SND. Thus, neither the history alone nor the ECG findings is reliable in making an accurate diagnosis of SND. In recent years, several non-invasive and invasive tests have been described in the evaluation of patients with suspected SND. A 24-hour ambulatory Holter monitor remains an important test in the diagnosis of suspected SND[13]. Transtelephonic recorders that are carried or worn by the patient are most useful in patients with intermittent symptoms, especially if the symptoms are not associated with abrupt syncope or if they occur with a brief prodrome. Electrophoysiologic tests were not performed for the evaluation of sinus node function in all patients[14].

Bradycardia-tachycardia syndrome (sinus bradycardia alternating with atrial flutter or reentrant atrial tachycardia) is an increasingly frequent problem in young patients following surgery for congenital heart disease. It is clear that long-term drug therapy deemed essential for the control of atrial flutter may result in symptomatic bradycardia in some patients, whereas in others the use of antiarrhythmic agents may potentially increase the risk of ventricular arrhythmias or sudden death in the presence of profound bradycardia. Thus, in young patients with recurrent arrhythmias associated with the bradycardia-tachycardia syndrome, permanent pacing should be considered as an adjunctive form of therapy[15]. Four of our patients who had bradycardia-tachycardia syndrome are symptom-free at follow-up after permanent pacemaker therapy. Two of four patients also received antiarrhythmic medication.

Sinus node dysfunction is not itself an indication for pacemaker implantation. Symptomatic bradycardia is considered an indication for pacemaker implantation, provided that other causes of the symptoms have been excluded. Alternative causes to be considered include seizures, breath holding, apnea, or neurally mediated mechanisms[4].

Single-chamber atrial pacemakers, with rate-responsive capability if appropriate, have been advocated for patients with SND but no evidence of atrioventricular (AV) block. Available data suggest that the incidence of atrial fibrillation in patients receiving atrial or

dual-chamber pacemakers may be lower than in patients receiving ventricular pacemakers[16]. Some studies showed a lower mortality in atrial-based pacemaker patients and others showed no significant difference. Short-term crossover studies in patients with SND have shown improved quality of life in dual-chamber versus ventricular pacing[17]. Most patients at our hospital who required pacemakers received ventricular demand epicardial or transvenous systems in previous years. When the screw-in leads were developed, we performed atrial or dual-chamber demand transvenous systems with rate-responsive capability.

In conclusion, SND has been diagnosed with increasing frequency in children with congenital heart defect, especially in patients who have undergone corrective cardiac surgery related with atrial tissue. The incidence is highest in patients who have undergone Mustard or Senning procedure for TGA. There is also a significant number of patients who are considered idiopathic because of no associated cardiovascular disease. The diagnosis of SND is difficult because of documentation. When pacemaker therapy is indicated, atrial demand endocardial systems are preferred.

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Tubular functions in familial Mediterranean fever

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SUMMARY: Akkuş S, Çalışkan S, Kasapçopur Ö. Tubular functions in familial Mediterranean fever. Turk J Pediatr 2002, 44: 317-320.

In this study, we aimed to evaluate renal tubular function in familial Mediterranean fever (FMF). Urinary N-acetyl-β-D glucosaminidase (U-NAG), β_2-microglobulin (U-β_2M) and microalbumin (Ua) levels were measured in children with different clinical stages of FMF (58 patients with FMF, 9 patients with amyloidosis secondary to (FMF). Control groups were healthy children (n=21), children with upper respiratory tract infection (URTI) (n=21) and with steroid sensitive nephrotic syndrome (SSNS) (n=18). U-NAG was significantly increased in patients with a recent diagnosis of FMF compared to patients with FMF on colchicine and to healthy controls. In patients with recently diagnosed FMF, a marked decrease in U-NAG, U-β_2M and Ua were determined after three months on colchicine therapy. On the other hand, U-NAG and Uβ_2M levels were increased in patients with FMF during attacks and then decreased in the post-attack period. U-β_2M in patients with FMF during attacks was significantly different from patients with URTI. Finally, U-NAG and U-β_2M were increased significantly in patients with FMF-amyloidosis and SSNS when compared with other FMF groups and healthy controls, respectively. In conclusion, the high U-NAG value in newly diagnosed patients compared to that of patients taking colchicine and the decline of U-NAG and U-β_2M levels after attack to the levels observed in colchicine users (without a significant change in Ua value) suggest that the renal injury early in the course of FMF might be dominantly at the level of the tubuli.

Key words: familial Mediterranean fever, renal tubular functions.

Familial Mediterranean fever (FMF) is an autosomal recessive disorder characterized by recurrent attacks of fever and polyserositis. The disease typically affects certain ethnic groups living in the Middle East and around the Mediterranean basin; mainly Sephardic Jews, Armenians, Turks and Arabs[1-4]. Colchicine is used for the treatment of this disease. In some cases that are not treated with colchicine, renal amyloidosis and proteinuria develop. Renal amyloidosis starting with proteinuria may progress into nephrotic syndrome and may finally result in chronic renal failure[1-6]. Additionally, in some cases, Fanconi's syndrome may develop as a consequence of amyloidosis[1-6].

Two previous studies demonstrated the presence of transient microalbuminuria and tubular proteinuria that became more prominent during attacks[7-8]. Therefore it may be suggested that microalbuminuria, tubular proteinuria and enzymuria can predict renal involvement in FMF, as is the case for some other renal diseases. In order to test this hypothesis, a prospective trial involving FMF cases was conducted.

Material and Methods

The patient enrollment started on the 1st of January 1998 and was terminated on the 1st of January 1999. Children with different clinical stages of FMF were enrolled into the study (58 patients with FMF and 9 patients with amyloidosis secondary to FMF). The study groups are summarized below.

A) Familial Mediterranean Fever Group

The diagnosis of FMF was based on Tel-Hashomer criteria in all children with FMF. The children with FMF were further divided into subgroups according to the clinical conditions.

1. FMF-Attack Free, Under Colchicine Treatment: This group consisted of children with FMF who were all attack-free, under colchicine treatment. The mean duration of colchicine use was three months.

2. Newly Diagnosed FMF Patients (FMF-ND):

2 a) Newly Diagnosed FMF Patients (FMF-ND) Pre-Colchicine Treatment: This group consisted of newly diagnosed patients who had not used colchicine before and who later responded to colchicine use under follow up. Urine specimens were taken from all 23 subjects before initiation of treatment.

2 b) Newly Diagnosed FMF Patients After Three Months' Colchicine Treatment: After three months from the initiation of colchicine treatment, urine specimens were recollected from some of the group 2A subjects.

3) FMF Subjects Who Experienced Attacks Under Colchicine Treatment:

3 a) FMF Patients with Attack: The FMF patients were seen at the time of the attack. All 14 children had fever and abdominal pain.

3 b) FMF Patients in Post-Attack Period: Urine specimens were recollected 15 days after the attack from the 3A subjects.

4) FMF Patients with Amyloidosis: Nine patients who had amyloidosis confirmed by either rectal or renal biopsy were recruited in this group. All subjects were in different stages of amyloidosis.

B) Control Groups

1. Healthy Children (HC): Twenty-one healthy children seen in the Pediatric Outpatient Department of Cerrahpaşa Medical Faculty, who were between 5 and 15 years of age, were recruited as healthy controls.

2. Children with Upper Respiratory Tract Infections (URTI): This group was designed to be the control group of the FMF subjects at the time of the attack. Twenty children who were seen at the Pediatric Outpatient Department with signs of URTI and who had fever above 38°C were recruited in this group. Urine specimens were collected from the chidren in the febrile period.

3. Children with Steroid Sensitive Nephrotic Syndrome (SSNS): This group was included in the study as the control group of FMF subjects with amyloidosis. All subjects were under the follow up of the Pediatric Outpatient Department. This group consisted of 18 SSNS patients who had normal C_3 levels and no macroscopic hematuria. None of the children had proteinuria at the time of the study.

The demographic variables for the groups and the clinical stages of FMF cases are shown in Table I.

The second morning urine was collected from both the control and patient groups. Each urine sample was dispensed into three different test tubes and kept at +4°C [for albumin (Ua) measurement] and –20°C for N-acetyl-β-D glucosaminidase (NAG) and β_2-microglobulin (β_2M) measurements] for one month at most. NAG measurements were performed spectrophotometrically (Boehringer-Mannheim) with the use of krezolfonfleleinglucosaminidase as the substrate. β_2-microglobulin levels were measured with radioimmunoassay (DSL. Beta-2M kit). Micro albumin levels were assessed with radioimmunoassay technique "double antibody" kits (BDPS). Creatinine (c)

Table I. Demographic Characteristics and Clinical States of the Study Groups

Group	N	Male/female	Mean age at investigation (years)	Mean age at disease onset (years)
FMF-attack free C (+)	21	11/10	9.8±3.2	4.8±3.2
FMF-new diagnosis C (–)	23	12/11	8.7±4	6.1±4.2
FMF-new diagnosis after 3 months' colchicine treatment C (+)	11	6/5		
FMF-attack C (+)	14	7/7	8.4±3.3	4.1±2.9
FMF-post attack C (+)	11	4/7		
FMF-amyloidosis C (+)	9	3/6	15.4±1.7	5.3±3.6
Healthy children	21	13/8	9.7±2.7	
Upper respiratory tract infection	20	10/10	7.4±2.1	
Steroid sensitive nephrotic syndrome	18	11/7	6.2±4.4	
Total	148	77/71	9.06±3.9	

• C=on colchicine treatment; FMF: familial mediterranean fever.

levels were determined by Jaffe method. Urinary NAG, β_2-M and micro albumin levels were proportionate to creatinine in order to adjust for differences arising from urinary flow.

The weight, height, age at the time of study, gender, and the results of urinalysis were recorded for all children.

Statistical Methods: Student's t, Mann-Whitney U and Wilcoxon tests were used in the statistical analysis.

Results

The results of the study are summarized in Table II. No urinary tract infection was detected in any of the study subjects.

Comparison of Attack-Free Children and Healthy Children: The U-NAG/c, Uβ_2M/c and Ua/c ratios in attack-free children who were using colchicine were not significantly different from ratios determined in healthy controls.

Assessment of the Effect of Colchicine: In newly diagnosed children who had never used colchicine, the control values for U-NAG/c, U-β_2M/c and Ua/c ratios after the use of colchicine for three months were lower than the basal values; however, the difference between the two groups was not statistically significant. On the other hand, the newly diagnosed children who were randomised to the no colchicine treatment group had significantly higher ratios of U-NAG/c compared to asymptomatic patients treated with colchicine ($p=0.029$); the differences in U-β_2M/c and Ua/c ratios were not significant.

The Assessment of the Effect of FMF Attack: The patients with an FMF attack had statistically higher U-NAG/c ($p=0.0064$), U-β_2M/c ($p=0.0001$) and Ua/c ($p=0.036$) ratios compared to patients who were using colchicine but not experiencing an attack. Tubular markers (U-NAG/c and U-β_2M/c) were higher in patients suffering from an attack than in URTI patients; however, only the difference in U-β_2M/c ratio was statistically significant ($p=0.046$). The tubular markers (U-NAG/c and U-β_2M/c) in the post-attack phase were lower than in the attack phase ($p=0.05$ and $p=0.0008$, respectively).

The Assessment of the Effect of Amyloidosis: The values detected in children with amyloidosis were significantly higher than the values

Table II. Results of the FMF and Control Groups [median (range)]

Group	N	UNAG/c (U/L)	Uβ_2M/c (μgr/gr)	Ua/c (μgr/mg)
FMF-attack free C (+)	21	2.8 (0.9-168)	4.96 (1.4-65.1)	0.11 (0.02-0.37)
FMF-new diagnosis C (–)	23	4.9 (0.96-25)	6.8 (0-56.2)	0.12 (0.04-1.42)
FMF-new diagnosis after 3 months' colchicine treatment	11	3.6 (1.32-8.8)	4.1 (1.01-520.8)	0.11 (0.09-0.94)
FMF-attack C (+)	14	10.8 (0.12-86.3)	75.9 (9.8-404.04)	0.2 (0.07-0.6)
FMF-post attack C (+)	11	5.1 (1.22-16.8)	4.95 (1.22-225.8)	0.1 (0.07-0.06)
FMF-amyloidosis C (+)	9	89.9 (18.99-441.02)	236.9 (7.59-2498.7)	4.6 (0.2-44.52)
Healthy children	21	3.5 (1.05-7.88)	5.9 (1.36-108.5)	0.08 (0.0007-0.89)
Upper respiratory tract infection	20	3.5 (0-85.71)	16.4 (0.666)	0.2 (0.08-4.35)
Steroid sensitive nephrotic syndrome	18	45.05 (8.82-285.5)	333.4 (18.7-1373.6)	2.1 (1.02-3043)

* C=on colchicine treatment; FMF: Familial Mediterranean fever; UNAG: urinary N-acetyl-β-D glucosaminidase; c: creatinine; Uβ_2M: urinary β_2-microglobulin; Ua. urinary microalbumin.

detected in all other FMF groups. There was no statistically significant difference between the group with amyloidosis and the group with SSNS with regard to laboratory values.

Discussion

There were no statistically significant difference between the FMF group treated with colchicine and the healthy controls, with regard to U-NAG/c and U-β_2M/c values. However, the U-NAG/c ratio in the newly diagnosed cases who were not receiving colchicine treatment was significantly higher than in cases on colchicine treatment. Additionally, there was a trend toward a decline in tubular markers after three months of colchicine treatment. This suggests that tubular involvement is possible in FMF patients, and colchicine may have a protective effect on tubular function.

Patients experiencing an attack had statistically higher ratios of U-NAG/c and U-β_2M/c compared to asymptomatic patients treated with colchicine, and there was a significant decline in these values after the attack. These findings suggest a transient tubular impairment. Saatçi et al.[8] detected similar increases in β_2 microglobulinuria during attack. Detection of significantly higher levels of U-β_2M/c during attacks compared to URTI cases with fever suggests that this finding cannot be explained on the basis of high fever and inflammation alone and that additional factors can have a role as well.

Oren et al.[7] detected microalbuminuria in the first morning urine samples of some adult FMF patients receiving colchicine. In our study, the Ua/c ratios in FMF cases receiving colchicine treatment and in healthy children were not different. In newly diagnosed cases of FMF, there were no statistically significant changes after three months of colchicine treatment. These findings suggest that there is no early glomerular injury in FMF. On the other hand, the Ua/c ratio during attacks was significantly higher than the ratio detected in healthy children. Similarly Saatçi et al.[8] described a higher Ua/c ratio during attacks. One possible explanation for the increase in this ratio is fever. Accordingly, there were no significant differences between the values detected in URTI cases. Another possibility is the development of microalbuminuria secondary to tubular involvement during attacks.

In the group with amyloidosis, there was a marked increase in tubular markers, similar to the one observed in SSNS. The absence of a difference between the groups suggests that proteinuria is responsible for this increase.

In conclusion, the high U-NAG value in newly diagnosed patients compared to that of patients taking colchicine, and the decline of U-NAG and U-β_2M levels after attack to the levels observed in colchicine users (without a significant change in Ua value) suggest that the renal injury early in the course of FMF might be dominantly at the level of the tubuli. These findings are not enough to support the hypothesis that proteinuria and enzymuria have predictive value in FMF cases. A prospective trial is warranted in cases who are not receiving colchicine, but that does not seem feasible for ethical reasons.

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Childhood onset of narcolepsy-cataplexy syndrome in Turkey: clinical and genetic study

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Narcolepsy is a disabling sleep disorder characterized by excessive daytime sleepiness and abnormal manifestations of rapid eye movement (REM) sleep including cataplexy, sleep paralysis and hypnagogic hallucinations. It is known to be complex disorder in which both genetic predisposition and environmental factors play a role. In humans, susceptibility to narcolepsy is tightly associated with a specific HLA allele, DQB1*0602. In this report, we took advantage of the ongoing genetic study in Turkish narcoleptic patients to document clinical and genetic data of eight patients whose onset of symptoms were in the childhood period.

*Key words: narcolepsy-cataplexy syndrome, HLA DQB1*0602, excessive daytime sleepiness.*

Narcolepsy-cataplexy syndrome is among the leading causes of excessive daytime sleepiness (EDS) and is the most common neurologic cause. Despite a prevalence similar to that of multiple sclerosis, a socio-economic impact that may be as high as that of epilepsy, and the availability of effective treatments, knowledge about narcolepsy often remains limited even among neurologists[1].

Excessive daytime sleepiness, cataplexy, sleep paralysis, hypnagogic hallucinations and disturbed nocturnal sleep are the five cardinal symptoms of the narcolepsy-cataplexy syndrome. These symptoms vary greatly in severity and occurrence in narcoleptic patients. Daytime sleepiness is usually the most disabling symptom of the narcolepsy syndrome. Patients with narcolepsy report a continuous sleepiness that fluctuates and episodically becomes irresistible, with involuntary sleep attacks occurring in such unusual circumstances as talking, eating, standing and even walking. Sleepiness alone has a poor diagnostic value, because it is difficult to differentiate from that observed in other sleep disorders, as in obstructive sleep apneas, especially in adults. However, cataplexy, the second most disabling symptom, is almost pathognomonic for the syndrome. Cataplexy is defined as brief episodes of muscle weakness provoked by strong emotions. Typically laughter, happiness or anger produces brief attacks of knee buckling, head dropping, and/or jaw sagging that may escalate in total paralysis with collapse to the floor. The severity of cataplexy is variable and may range from unnoticeable loss of muscle tone in the legs to a complete episode of muscle paralysis. In most patients, cataplexy lasts for one minute or less (usually for just a few seconds) and consciousness is maintained during the episode. Isolated cataplexy without associated sleepiness is an exceptional occurrence that is most often observed in young narcoleptic subjects still developing the full-blown disorder. Sleep paralysis and hypnagogic hallucinations are not specific for narcolepsy and their diagnostic values are very poor. Sleep paralysis (SP) is seen in 60-80% of narcolepsy patients and is characterized by an inability to move at sleep onset or upon awakening. Hypnagogic hallucinations are reported in 40-60% of patients with narcolepsy and are the expression of the changing state of consciousness in which, as opposed to dreaming, elements of the normal awake mentation are still present. Such hallucinations may be simple (e.g., unformed sensations, geometric figures) or complex (e.g., faces, animals) and may involve one or more senses[1].

The diagnosis of narcolepsy is facilitated by the availability of an established diagnostic test, the multiple sleep latency test (MSLT)[2]. In this test, patients first undergo nocturnal polysomnography to eliminate other causes of daytime sleepiness such as obstructive sleep apnea. The following day, sleep is also recorded during five successive 20 minute naps separated by two-hour intervals. Latency to falling asleep and possible occurrence of rapid eye movement (REM) sleep episodes are then noted for each nap. Normally, MSLT sleep latency is more than 10 minutes and at most one REM sleep period (sleep onset REM period or SOREMP) is observed after an efficient night-time sleep. In contrast, narcoleptic patients have poor night-time sleep efficiency (% asleep/total bed time), a very short mean sleep latency (≤8 min) and multiple SOREMPs (≥2 in 5 naps).

Human narcolepsy-cataplexy is associated with centrally mediated hypocretin deficiency that has a tight association with HLA DQB1*0602[3]. Narcoleptic patients with well defined cataplexy appear to represent an etiologically pure nosological entity with a very high (85-100%) degree of association with HLA DQA1*0102 and DQB1*0602, mostly in the context of the DRB1*15, DQA1*0102, DQB1*0602 haplotype[4,5]. Also, significantly higher relative risk was reported for heterozygote combinations including DQB1*0301, DQA1*06, DRB1*04, DRB1*08, DRB1*11 and DRB1*12[3]. Three alleles, DQB1*0501 and DQA1*01 (non-DQA1*0102), were revealed to be protective[3].

Although considerable attention has been directed toward understanding narcolepsy in adults, few investigators have focused on childhood narcolepsy. Limited reports have emphasized that narcolepsy symptoms may begin in childhood. Retrospective studies suggest that about half of the adults with narcolepsy report experiencing symptoms during childhood or early adolescence[6]. Childhood narcolepsy is emerging as a significant clinical entity for several reasons. This disorder if frequently underrecognized and undiagnosed, leading to years of untreated symptoms during the important childhood years.

In this report, we took advantage of the ongoing genetic study of narcolepsy-cataplexy syndrome in Turkey to describe the onset of symptoms, and diagnostic and genetic data of narcoleptic patients whose narcoleptic symptoms manifested before puberty.

Material and Methods

All subjects reported here were seen prospectively at İstanbul University, Cerrahpaşa Medical School, Neurology Department, Sleep Disorders Unit between 1996 and 1999. The data of eight narcoleptic patients (5 female, 3 male) who were selected from 25 narcoleptic patients, with the inclusion criteria of onset of symptoms before the age of 16, were documented.

At the initial visit, the clinical evaluation included a complete history, general physical examination, complete neurological examination, administration of a sleep questionnaire and a narcolepsy inventory. The sleep questionnaire used was the Epworth Sleepiness Scale[7]. These were completed by a physician and the patients were to answer the questions with their close relatives when needed. The Epworth Sleepiness Scale is a questionnaire that asks patients to rate their sleepiness in each of eight different situations on a scale from 0 (never) to 3 (high chance)[7]. The total score in the Epworth scale can range from 0 to 24. The range from 0 to 7 was accepted as normal and above the score of 7 as an indicator of pathologic degree of sleepiness[7]. The narcolepsy inventory (Stanford Center for Narcolepsy Sleep Inventory) is a validated, 146-item questionnaire requesting details and specific examples for all narcolepsy symptoms experienced, with special emphasis on cataplexy[5].

All patients were studied with the same nocturnal polygraphic parameters. These recordings included electroencephalography (EEG) (C3/A2, C4/A1 of the international 10-20 electrode replacement system); chin electromyography (EMG); right and left electro-oculogram (EOG); nasal-oral airflow and oxygen saturation. The polysomnograms were scored using the standard international recommendations of Rechtschaffen and Kales[8]. These recordings were always performed after the patients were free of medication for at least 10 days.

MSLT was obtained following a nocturnal polysomnography. The MSLT recording included EEG as above, chin EMG, and right and left EOGs. For each MSLT, we recorded the mean sleep latency and the number of SOREMPs. Sleep latency was defined as the time in minutes from lights out to the first epoch (30 sec) of sleep. Sleep-onset REM sleep was scored when REM sleep occurred within 15 minutes of the first epoch of sleep.

HLA typing of all patients was performed with the great support of Emmanuel Mignot at Stanford University, Center for Narcolepsy. Inclusion criteria for HLMA typing: a) recurrent daytime naps or lapses into sleep occurring almost daily for three months, b) sudden bilateral loss of postural muscle tone in association with intense emotion (cataplexy) and c) less than 8 minutes sleep latency on a 5 nap MSLT. These patients also had to have ≥2 SOREMPs during MSLT and polysomnography combined. Blood drawings were performed from the patients and when possible their parents and controls. All subjects gave informed consent for blood drawings for HLA typing. Blood samples were sent to Stanford University at the same day of blood drawing, together with the data of polysomnography, MSLT and narcolepsy inventory.

Serological HLA typing was performed at the Stanford University Blood Center (Palo Alto, CA, U.S.A.) HLA DQB1 and DRB1 subtypes were determined using group-specific polymerase chain reaction (PCR) amplification and Sequence Specific Oligonucleotide (SSO) hybridisation as previously described by Mignot et al.[4].

The results were given as mean ± standard deviation.

Results

Narcolepsy-cataplexy syndrome was diagnosed in these eight patients (5 female, 3 male) based on the complaint of daytime sleepiness, the presence of cataplexy and their MSLT results. Their mean age on admission was 21±6.2 years. The age range was 12 to 28 years. The age of symptom onset was 12.9±3.3 years (range: 6 to 16).

Excessive daytime sleepiness was the first symptom in four patients. School-related complaints like inability to follow the lesson due to lack of attention and learning disabilities were more pronounced by these patients after the onset of symptoms.

Cataplectic attacks were the initial complaint in four patients. The emotional conditions that triggered cataplectic attacks are shown in Table I.

Table I. Emotional Conditions Triggering Cataplectic Attacks

Emotional Conditions	Patients* (n=8)
Laughter	8
Anger	5
Excitement	8
Surprise	8
Embarrassment	4
Stress	4
Being startled	4
Tension	2
Telling or hearing a joke	6

* The number of patients who had the emotion triggering cataplexy.

Three patients described hypnagogic hallucinations and only one patient had had both sleep paralysis and hypnagogic hallucinations during the childhood period. These symptoms appeared after the onset of the main symptoms, either excessive daytime sleepiness or cataplexy.

Epworth sleepiness score of patients was 21.5±1.9 (range: 18 to 23). The mean sleep latency of MSLTs was found to be 0.96±0.85 minutes (range: 0.5 to 3 minutes). Three patients had sleep onset REM period in each 5 nap MSLT. Three other patients had 4 SOREMPs in 5 naps. The remaining two patients had 3 and 2 SOREMPs, respectively.

The HLA analysis demonstrated that all patients were heterozygous for HLA DRB1*1501, DQB1*0602 except one. All HLA results of patients, their parents (if present) and their controls are given in Table II.

Discussion

This report is the first preliminary genetic and clinical data of narcolepsy-cataplexy syndrome in Turkey. The aim of this study was to focus

Table II. HLA DRB1 and DQB1 Subtypes in Narcoleptic Patients, Parents and Their Controls

Patient Number	Patient DRB1-DQB1	Father DRB1-DQB1	Mother DRB1-DQB1	Control DRB1-DQB1
1	0402/1501-0302/0602	0402/1104-0301/0302	1201/1501-0301/0602	0701/1601-0201/0502
2	1401/1501-0503/0602	0402/1501-0302/0602	1104/1401-0301/0503	0101/0101-0501/0301
3	0701/- -0201/-			0402/0701-0302/0303
4	1201/1501-0301/0602	1501/1502-0601/0602	0401/1201-0301/-	1101/1502-0301/0601
5	1301/1501-0602/0604	1501/1103-0301/0602	1301/1302-0603/0604	0701/0901-0203/0302
6	1101/1501-0301/0602			0101/0701-0501/0201
7	1104/1501-0301/0602			0403/1103-0305/0301
8	1104/1501-0301/0602			0101/1202-0501/0301

on the presenting features that are unique to childhood narcolepsy and provide especially the genetic information about Turkish narcoleptics. Studies of childhood narcolepsy in the medical literature are sparse. In 1960, Yoss and Daly[1] reported that 59% of 85 consecutive adults with narcolepsy experienced symptom onset by the age of 15. They reported 16 subjects with symptom onset before 15 years of age, including three children with onset by the age of three. In a retrospective review, Nevelet et al.[6] reported that excessive sleepiness presented before 15 years of age in 49% of adults with narcolepsy. In our patient population, 32% of narcoleptic patients had the first experience of narcoleptic symptoms before the age of 16, and the earliest onset of symptoms was found to be at the age of six. This lower percentage may be due to underrecognition of symptoms by both patients and their families before puberty.

In 1998, Gulleminault et al.[9] reported that cataplexy was an obvious symptom and clearly preceded observance of daytime sleepiness in several of his cases. They defined laughter as always being reported as a trigger af cataplexy. Of our cases, 50% had cataplexy as an initial symptom. Moreover, laughter, excitement and surprising conditions were common reports of our patients as causes of cataplectic attacks. Although these results are similar to the literature, study of a larger number of patients is still necessary in order to comment on Turkish narcoleptics. The early presence of cataplexy and frequency of complete association of HLA DR15 and DQB1*0602 must be emphasized in children with early onset of the disease[4,5]. The Turkish results confirm the positive association of the HLA DRB1*1501 and DQB1*0602. In Turkey a similar association was found in patients with multiple sclerosis in that myelin degeneration led to this disease, while in narcolepsy-cataplexy syndrome[11], degeneration in hypocretin neurons was present.

In 1988, Young et al.[12] analyzed clinical and polysomnographic data in patients with narcolepsy, comparing eight children at the age of 15 or younger with a comparable adult group. The pediatric group showed greater daytime sleepiness and a higher frequency of SOREMPs than the adult group, as measured by MSLT. The results of MSLT and Epworth score revealed greater daytime sleepiness in our patient population.

Even though the overwhelming majority of the adult cases of narcolepsy and cataplexy are idiopathic, narcolepsy has occasionally been shown to be associated with brain tumors and various other brain lesions, mostly around the third ventricle[13] and in the pons[14]. The situation is more complex in children, especially those younger than six to eight years of age. In an analysis of 97 reported cases of childhood narcolepsy by Challamel et al.[15], a significant proportion of very young children with narcolepsy and cataplexy had other associated disorders. Niemann-Pick disease type C and D and diencephalic tumors were diagnosed in this group. Therefore, narcolepsy-cataplexy syndrome carries greater importance when symptoms begin during childhood.

In conclusion, narcolepsy-cataplexy syndrome is only recently beginning to be recognized in Turkey. The increasing number of Turkish patients will provide new insights into our patient population and help to generate various medical and psychological treatment approaches in narcoleptic children as well as in adults.

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Evaluation of bone mineral density in chronic glue sniffers

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SUMMARY: Dündaröz MR, Sarıcı SÜ, Türkbay T, Baykal B, Kocaoğlu M, Aydın Hİ, Gökçay E. Evaluation of bone mineral density in chronic glue sniffers. Turk J Pediatr 2002; 44: 326-329.

Although acute and chronic toxic effects of inhalant (glue) abuse have been well demonstrated on many organ systems, the effects on the skeletal system and bone mineral content of young people with this addiction have, to our knowledge, not yet been investigated by bone mineral density measurement. In the present study bone mineral density was measured by the dual-energy X-ray absorptiometry method in 25 children and adolescents with inhalant abuse and compared with that of a control group (n=30) to detect whether there was any delay in bone development or any decrease in bone mass. Chronological age, height and weight, serum calcium, phosphorus and alkaline phosphatase levels of the study group were not significantly different from those of the control group ($p>0.05$), whereas bone mineral density was significantly reduced in the study group ($p=0.001$). Teenagers with glue vapor abuse may carry an increased risk of future fracture even though the exact mechanism(s) responsible for the toxicitiy of glue vapor on bone metabolism remains to be determined. To ascertain the exact component of glue responsible for bone demineralisation may be of value in proposing a change in the composition of the glue. Education and/or rehabilitation programs currently have the greatest importance in preventing and overcoming the harmful effects of this public health problem which is so common in young children and adolescents.

Key words: addiction, adolescent, bone mineral density, children, glue, inhalant abuse.

Inhalant abuse is one of the most common problems of young children and adolescents in developing countries. Desired effects with intentional inhalation of a volatile substance appear to be euphoria, tranquillity, relaxation and hallucination. Glue/adhesive sniffing is the most common form of volatile substance abuses since these substances are different from other drugs in that they are not sold illegally[1].

There are significant morbidity and mortality, both organic and psychosocial, associated with inhalant abuse. Both acute and chronic toxic effects in neuropsychiatric, urological, hematological, cardiovascular, pulmonary and gastrointestinal systems, including death due to cardiac arrhythmia or pulmonary and cerebral edema, encephalopathy syndromes, cerebral damage, optic atrophy, peripheral neuropathy, muscle weakness, gastrointestinal disturbances, or hepatic and renal damage well have been documented[2-4]. However, toxic effects of inhalant abuse on the skeletal system and bone mineral content of young people with this addiction have, to our knowledge, not yet been evaluated by bone mineral density (BMD) measurement although childhood and adolescence are critical periods of bone mineralization and skeletal development.

In the present study, BMD was measured by the dual-energy X-ray absorptiometry (DXA) method in children and adolescents with inhalant abuse and compared with that of a control group to detect whether there was any delay in bone development or any decrease in bone mass.

Material and Methods

This study was performed between September 2000-May 2001 at the Government's Education and Rehabilitation Center of Children in

Ankara, where inpatient and outpatient chemical dependency treatment and prevention programs are conducted. The study group consisted of 25 boys whose ages were between 13 to 19 years (mean 14.9±2.5 years). These cases had overcome the habit at least one month previously (range 1 to 3 months, mean 2.3 months), and the duration of abuse before rehabilitation was between 2 to 5 years (mean 3.2 years). All the cases were smokers, and none took any antioxidant drugs (vitamin C, vitamin E, selenium, etc.) prior to or during the study. The parents and authorities were informed about the study and informed consent was obtained from the cases and/or their parents.

Thirty healthy volunteer adolescents of similar age (mean 15.1±2.9 years) without any addiction were chosen as the control for each case in the study and control groups, brief history was taken, and a complete physical examination was performed. Blood samples for complete blood count, serum calcium, phosphorus and alkaline phosphatase (ALP), and renal and liver function tests were obtained, and a urinalysis was performed. Cases with any signs or symptoms of any acute or chronic illness were excluded from the study.

Body weight and height measurements of the cases in the study were taken carefully by the same experienced personnel. BMD was measured by DXA method[5,6] at L2-L4 levels of lumbar vertebrae using the DXA Norland (Fort Atkinson, WI, USA) XR-36 densitometer. Spinal BMD was measured in supine position; the scan time ranged between 3 to 5 minutes, and results for spinal BMD measurements were expressed in g/cm^2.

Mann-Whitney U test was used in statistical analysis of the data.

Results

Chronological age, anthropometric measurements, serum calcium, phosphorus and ALP levels, and BMD measurements of the study and control groups are shown in Table I. Chronological age, height and weight, serum calcium, phosphorus and ALP levels of the study group were not significantly different from those of the control group ($p>0.05$), whereas BMD was significantly reduced in the study group ($p=0.001$) (Table I).

Discussion

The effects and toxicity of various components of volatile substances on skeletal development and mineralization have been investigated in only a few studies, most of which have been performed on animals. Subchronic toluene exposure caused inhibition of skeletal growth (torso length, rump width) in weanling male rats which became relatively shorter in length and narrower in girth as they grew, compared to controls[7]. In mouse embryos and fetuses maternal exposure to toluene during pregnancy was associated with a higher incidence of the presence of 14 ribs, suggesting the teratogenic toxicity of toluene on the skeletal system[8]. In two other studies, delayed ossification of sternebrae after inhalation of benzene in rats[9] and skeletal variations due to oral or inhalational benzene exposure in mice and rabbits[10] were reported. In a study conducted at a forensic medical institute, where bone age measurement is routinely performed in determination of the identities of prosecuted glue vapor abusers, a significant bone age retardation has been demonstrated in this population when compared to healthy controls[11]. The present study is the first one

Table I. Comparison of the Data Obtained in the Study and Control Groups*

	Study group (n=25)	Control group (n=30)	p value
Chronologial age (years)	14.9±2.5	15.1±2.9	p>0.05
Height (cm)	162.3±11.2	165.1±10.8	p>0.05
Weight (kg)	53.3±8.9	54.7±9.4	p>0.05
Serum calcium (mg/dl)	9.41±0.7	9.35±0.5	p>0.05
Serum phosporus (mg/dl)	5.23±0.5	5.19±0.9	p>0.05
Alkaline phosphatase (mU/ml)	251±25	249±21	p>0.05
Bone mineral density (g/cm^2)	0.7065±0.116	0.8755±0.182	p=0.001

* Values are given as mean ± SD.

investigating the bone mineral content by DXA method and reporting a significantly reduced BMD in adolescents who chronically abused glue vapor. DXA is a new method that permits BMD to be measured accurately and directly. Lumbar spinal BMD measurements are preferred in this method. The spine is formed mainly of trabecular bone, which has higher surface-to-volume ratio and is more active metabolically than other regions of the skeleton. Because of the greater rapidity of bone turnover in the trabecular compartment than in the cortical one, the spine is thought to be a more sensitive site than the long bones of the upper and lower extremities for evaluating the effects of various stimuli on bone mineral status of the body[5,6,12].

Bone mineral content is influenced by genetic, hormonal and exogenous factors such as physical activity, diet, certain medications, and exposure to sunlight. Cases in both the study and control groups in our study had relatively the same degree of physical activity and were living in the same region, being exposed to relatively the same amount of sunlight, although we could not determine the objective degrees and/or amounts of these two parameters. Moreover, anthropometric measurements and serum calcium, phosphorus and ALP, which indirectly reflect growth, development and nutritional status, were not significantly different between the study and control groups in this study.

The other factors which might possibly affect bone mineralization in this study were alcohol consumption and smoking. However, there are some controversies about the effects of these habits on bone mineral density values. A detrimental effect of chronic alcohol abuse on the skeleton has been shown in two studies[13,14]. whereas two other studies[15,16] have suggested a beneficial effect of light-to-moderate alcohol consumption, and some other studies[17-19] have demonstrated no significant effects. Regarding smoking, some negative effects on the skeletal system have been reported[15,17], wehereas two other studies[18,19] found no obvious effects on bone mineralization. Most of the cases in our study group were smokers and consumed alcohol although we could not quantify amounts. Thus, to demonstrate the exclusive effects of glue sniffing on bone mineralization, it would certainly be more appropriate, if possible, to compare our study group with a control group consisting of cases who merely smoked and consumed alcohol but did not abuse any volatile substances.

Results of the present study indicate that teenagers with glue vapor abuse may be at risk for developing ostoopenia, possibly due to the toxic effects of glue vapor on the skeletal system. BMD peaks by the age of 20, makes a long plateau, and decreases after the age of 40. A lower peak bone mass in adolescence has been suggested to be associated with a greater risk of osteoporosis and fracture in the future[20,21]. Teenagers with glue vapor abuse, therefore, may carry an increased risk of future fracture even though the exact mechanism(s) responsible for the toxicity of glue vapor on bone metabolism remains to be determined. Commercially available glue products generally contain toluene, benzene, xylene, trichloroethylene, tetrachloroethylene, methylene chloride, trichlorethane, carbon tetrachloride, acetone, naphtha, and n-hexane. To determine the exact component of glue responsible for bone demineralisation may be of value in proposing a change in the composition of the glue. Education and/or rehabilitation programs currently have the greatest importance in preventing and overcoming the harmful effects of this public health problem which is so common in young children and adolescents.

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Comparison of thicknesses of the myocardial fibers of anencephalic and normal human fetuses

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When heart transplantation is needed in newborns, brain death should be confirmed, and the heart should not be exposed to hypoxia. The anencephalic newborn has been presented as a donor in heart transplantation. It is important, therefore, to evaluate possible morphological differences in the hearts of anencephalic cases. In this study, muscle fibers were studied in 10 anencephalic and 10 normal fetuses (27-35 weeks) and the results were compared. Random samples were taken from the upper 1/3 of the right ventricle's posterior wall and processed for light microscopic examination. Thicknesses of the 100 myocardial muscle fibers for each fetus were evaluated. There was statistically no significant difference between the anencephalic and normal fetus groups and the sex groups. Morphological features of the transplant probably affects the performance of the heart after operation. The anencephalic fetuses could be unique donors for heart transplantation.

Key words: anencephaly, heart transplantation, fetal heart, light microscopy, human fetus.

In this study we aimed to compare the thicknesses of the myocardial muscle fibers in anencephalic and normal fetal hearts. It is important to evaluate the possible morphological changes in anencephalic cases, and no microscopic study on this subject exists in the literature to our knowledge.

Use of anencephalic fetuses as donors in heart transplantation has been presented in the literature[1-3]. Heart transplantation in newborns is accepted as an effective treatment for congenital heart diseases such as cardiomyopathies and endocardial fibroelastosis and in some cases of tricuspid atresia, which are not compatible with life[1,4-7-9]. In such a procedure, brain death should be confirmed, and the heart should not be exposed to hypoxia. However, it is not easy to find a donor that meets both conditions, and this lack of suitable donors is the most important factor restricting heart transplantation[1,2,4-6,7,10].

Material and Methods

In this study 10 anencephalic (8 female, 2 male) and 10 normal fetuses (6 female, 4 male), aged between 27 and 35 weeks, were used. The fetuses were fixed with 10% formalin solution.

For the estimation of the fetal age, foot lengths were measured and compared with Mercer's scale[11].

Atrium walls were removed to obtain tissue samples from the ventricular walls. Random samples were taken from the hearts. Tissue specimens were taken from the upper 1/3 of the right ventricle's posterior wall, including all three layers of the heart, for the evaluation of the ventricular wall thickness. Tissues were embedded in paraffin blocks. Five-micron thick sections were taken and stained with H&E.

On each slide, 100 heart muscle fiber thicknesses were measured. The measurements were taken from around the nuclei where the

muscle fibers thicknesses were largest. The arithmetic mean for each slide was taken, so in each group 10 arithmetic means were attained. Results were statistically compared both for the groups and for sexes. Factorial analysis of variance technique was used.

Results

The evaluation of the muscle fibers of the anencephalic and normal human fetuses by light microscopy showed that there were no obvious differences between the two groups. The muscle fiber density and structure of the two groups were found similar. Cytoplasmic solidity, nuclei and striated structure of the heart muscle fibers were of normal appearance (Figs. 1, 2).

Data obtained from the statistical analysis of myocardial fiber thickness of the anencephalic and normal fetus groups are given in Table I. The mean ± SE mean values of the myocardial muscle fiber thikness were 50.25±3.16 μ (SD=8.93) and 38.01±4.85 μ (SD=6.86) for female and male fetuses, respectively, in the anencephalic group. The mean±SE mean values were 43.04±1.87 μ (SD=4.59) for female and 43.60±4.02 μ (SD=8.03) for male fetuses in the normal fetus group. Minimum and maximum values were 43.25 μ-69.32 μ for female anencephalic fetuses and 33.16 μ-42.86 μ for male anencephalic fetuses. Minimum and maximum values of the normal fetus group were 37.66 μ-49.76 μ and 38.76 μ-55.54 μ for normal female and male fetuses, respectively.

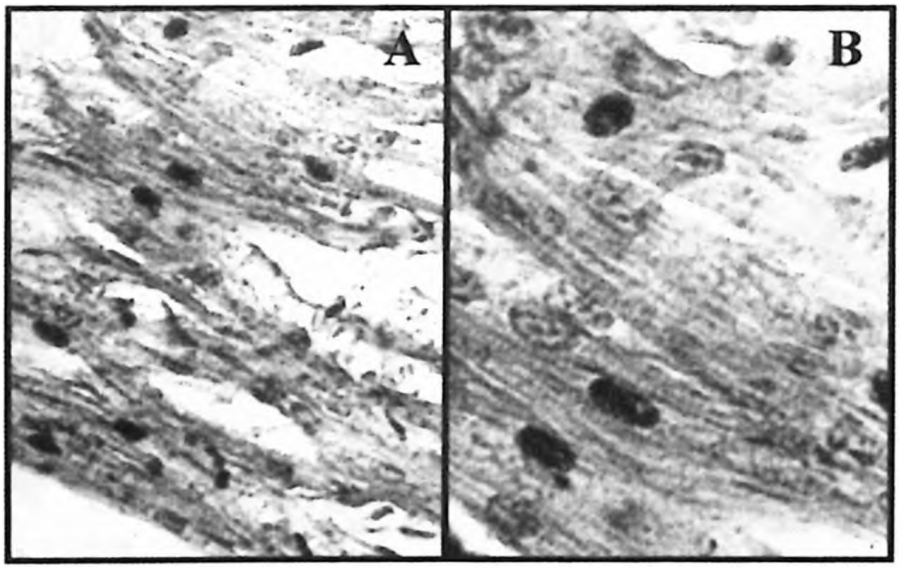

Fig. 1. Muscle fibers of the anencephalic fetus under the light microscope (A: X200 and B: X400, HE stain).

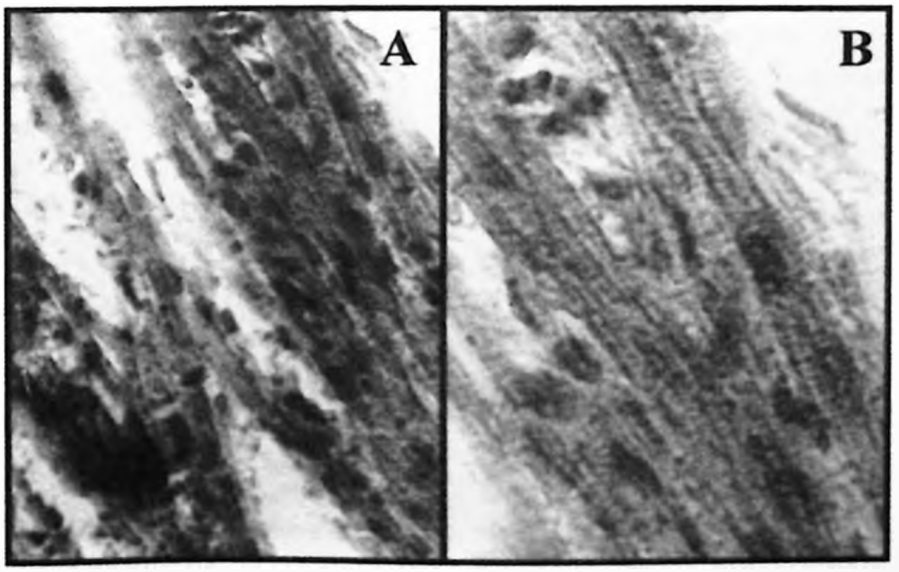

Fig. 2. Muscle fibers of the normal fetus under the light microscope (A: X200 and B: X400, HE stain).

Table I. Statistical Comparison of Myocardial Fiber Thickness Between Anencephalic and Normal Fetuses and Sexes

Group	Sex	Mean±SE mean (μ)	Standard deviation (μ)	Minimum (μ)	Maximum (μ)
Anencephalic	female	50.25±3.16	8.93	43.25	69.32
	male	38.01±4.85	6.86	33.16	42.86
Normal	female	43.04±1.87	4.59	37.66	49.76
	male	43.60±4.02	8.03	38.76	55.54

Statistically, no significant differences were found between the two fetus groups (p=0.196) or between genders (p=0.159). Furthermore, fetus group sex interaction was not statistically significant (p=0.169). Therefore, differences between anencephalic and normal fetus groups were not significant according to sex. Likewise, differences between gender groups were not significant for the anencephalic and normal fetus groups.

Discussion

Although a high number of organ anomalies are seen in anencephalic newborns, a specific heart anomaly is not mentioned in the literature[12-14]. There may be morphological differences between anencephalic and normal fetal hearts as a result of certain abnormalities at different developmental stages.

In postnatal life, progressive increase in heart load (in pathological states such as in hypertension and anemia, and in physiological states such as in strenuous exercise) causes myocardial fiber hypertrophy as a compensatory response. There is an increase in the rate of protein synthesis, the amount of protein synthesis in each cell, the size of mycoytes, the number of sarcomeres and mitochondria and consequently the mass and size of the heart due to the compensatory mechanism[15]. Likewise, in the fetal period brain tissue takes an important percentage of the blood pumped by the heart. In anencephalic newborns absence of any vascular bed of the brain is thought to decrease the heart load. In anencephalic fetuses, as the need for blood is less, the heart load should also decrease. Thus, it is possible that the myocardial fibers might undergo morphological and ultrastructural changes.

Effect of the cerebral cortex via the autonomic nervous system and the medulla has been shown on the fetal heart rate and the rhythm of the heart. Although only the medulla controls the fetal heart rate according to the studies performed in anencephalic and normal fetuses intrauterinely at 27-28 weeks, it has been shown later that it is under the control of the developing cerebral cortex[16-18]. Studies have also shown that the fetal brain affects the diurnal rhythm of the fetal heart rate and contributes to synchronous maternal-fetal rhythm. It has also been reported that the cerebral cortex is the origin or a transmission route for the response of fetal heart rate to acoustic stimulation[18-20]. For these reasons, there may be differences in the establishment of the fetal heart rate between anencephalic and normal fetuses, especially after the 27th week of intrauterine life.

Furthermore, removal of CSF (cerebrospinal fluid) or air invasion into the cerebral ventricles and impairment of intracranial hydrodynamics cause electrocardiographic alterations[21]. Neuronal stimulation not only affects the heart rate, but also the strength of contractibility[22]. In view of recent data, absence of brain tissue may affect functions of the heart. However, investigations of how these effects are reflected in the myocardial fibers, which are the functional units of the heart, do not exist in the literature.

General morphological features of the transplant probably affects the performance of the heart after the operation and may assist heart surgeons in determining the donor. In this study, thicknesses of the myocardial fibers were studied, and no differences between the two fetus groups was found. The results indicated that neither decrease in the heart load nor deficiency in autonomic innervation was reflected in the myocardial fiber thickness in the upper 1/3 posterior wall of the right ventricle. Considering the effects of the cerebrum on the heart, the anencephaly itself seems to be the reason for certain pathological changes. The results of this study showed that there is no statistically significant difference

between the thicknesses of the muscle fibers of normal and anencephalic fetuses in the right ventricle's posterior wall. However, these results may not be similar for the other regions of the heart, and thus, further investigation is needed in these two groups for other walls of the heart.

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Munchausen syndrome by proxy: a case report

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Munchausen syndrome by proxy (MSBP) is a serious form of child abuse, which is characterized by a child with symptoms and signs of an illness that have been fabricated by the mother. Here, we present a case of MSBP, who at four months of age was brought to our hospital by her mother because of rectal bleeding. The patient underwent many invasive procedures until the diagnosis of MSBP was finally considered. The mother reported no rectal bleeding for almost a year, during follow-up at the well-child outpatient department. At 19 months of age, another episode of rectal bleeding occurred, when a bloody diaper was presented to the attending physicians. The blood group and DNA analysis of the blood in the diaper confirmed the diagnosis. The case was reported to the social services and the patient was placed in the custody of her father's sister. The mother is still undergoing treatment in our psychiatry department.

Key words: Munchausen syndrome by proxy, factitious illness, rectal bleeding.

Munchausen syndrome by proxy (MBSP), first described by Meadow[1], is a serious form of child abuse which is characterized by a child with symptoms and signs of an illness that have been fabricated by an adult, usually the mother. Fabrication of those signs and symptoms lead doctors to perform many unnecessary and painful investigations and treatments on the child. The victims are usually under six years of age[2]. These are difficult cases to manage and the condition may result in the death of the victim. In the ones who survive, as they grow older, there is a tendency to participate in the deception and to believe that they are disabled[3].

The awareness of medical staff about child abuse is a new emerging concept in Turkey[4] and MSBP, being a special form of child abuse, is unknown by many physicians. We aim to call attention to the disease in our country, by presenting a case of MSBP. To our knowledge this is the first reported case in Turkey.

Case Report

G.E., now a 21-month-old girl, had been brought to our hospital approximately 1.5 years previously due to rectal bleeding. At four months of age, her mother, claiming passage of a bloody stool, brought her to the emergency department at 11:30 p.m. Physical examination was unremarkable except for a small anal fissure. A complete blood count, stool examination including cultures and abdominal ultrasonography revealed normal findings. The rectal bleeding was thought to be due to the anal fissure and the patient was sent home with appropriate recommendations. During the next three months, she was brought to the emergency department every two-three weaks, mostly at nighttime, with the same complaint. Because of the recurrence of the complaint, she was admitted to the pediatric ward for observation and investigation. All of the haematological values including complete blood count, ferritin, prothrombin time, and activated partial thromboplastin time were within normal limits. There was no occult blood in her stool. A radionuclide scan for Meckel's diverticulum was negative. Gastrointestinal system endoscopy and biopsy revealed normal findings. Radiographs of small and large bowel were normal and she was subsequently discharged. Fourteen days after her discharge, when she was seven months old, she was brought to the emergency room at 10:30 p.m. This time her mother supplied a diaper soiled

with blood, as hard evidence for rectal bleeding. Physical examination revealed the presence of a small amount of blood in her perineum, but no sign of injury was evident around her vagina or anus. She was readmitted for observation. During the hospital stay the mother appeared suspicious, because of her exaggerated willingness to let her baby undergo medical investigations, while being completely indifferent to her suffering. She explained that this was her second marriage and G.E. was her fourth child. Complaining about her current husband, she characterized him as an alcoholic. She claimed to have experienced domestic violence in her previous marriage, with her three previous babies dying around 8-10 months of age with similar symptoms. She expressed difficulty remembering their exact age of death, as well as the given diagnoses and even their names. When asked to present their medical records, she replied that this was impossibile, as they were destroyed by the recent earthquake in Adapazarı State Hospital. Her indifference alarmed the staff. Moreover, there were many inconsistent points in her story about times and places. Although claiming to be a police officer, investigations revealed that she actually was a housewife. An intern remembered her undergoing an appendectomy the previous years. Her file indicated that she was readmitted two days after her discharge and remained in hospital for another month because of wound infection, which failed to improve despite appropriate treatment. The intern further added that her physicians had assumed that she had intentionally infected herself. These facts led to the consideration of the diagnosis of MSBP.

The baby was moved to the intensive care unit to separate her from her mother. A psychiatric consultation was obtained concerning the parenting behavior of the mother. During the three psychiatric interviews, one of which included her husband, then most prominent feature was the inconsistency in both the personal history of the mother and the disease of her child. The content of her personal information including her own developmental, educational, marital, parental and professional history, was highly variable. When she was slightly confronted with these discrepancies, she managed to find new explanations without any sign of anxiety, which in turn were not compatible with the previous history. Psychometric tests including MMPI and Rorschach did not reveal any gross psychopathology, except for a tendency for somatization and depressive features, together with a defensive pattern.

Her husband appeared nervous and irritable about the interview and was highly uncooperative. He also had a defensive pattern about their marriage and about his wife, and was unwilling to volunteer clear information concerning the issues at hand. He denied any addiction or problems regarding alcohol consumption and refused further treatment. The case was reported to the social services. A home visit was performed, where the living conditions were found far from satisfactory, the flat being dirty and smelling of alcohol.

The baby was observed in the hospital for 15 days without subjecting her to any invasive procedure. During this time she was separated from her mother for three days while she was kept in the intensive care unit. Otherwise she stayed with her mother in a regular hospital room, with three patient beds that prevented the mother's remaining alone with her child. No rectal bleeding was observed during her hospital stay. At the time of discharge, the mother was informed that no organic disease could be found in the baby, but that she would be followed closely in the healthy children outpatient department. She was urged to feel free to consult her daughter's physicians whenever there was a problem, in order to discourage her from seeking medical attention elsewhere. Consequently she was followed by monthly visits for a year. The mother denied any problems including rectal bleeding. Normal developmental milestones were reached.

At 19 months of age, her mother, citing a large amount of rectal bleeding, brought G.E. to the emergency room one night at 01:00 a.m. Although a diaper with bright red, unclotted blood was presented, the baby looked healthy and her hemoglobin level was normal. The blood in the diaper was tested for blood type and was found to be 0 RH (+), while the baby's blood group was A Rh (+). This incompatibility was confirmed by the criminology laboratory, which revealed that the DNA analysis of the blood in the diaper was different from the patient's blood. This last incident confirmed the diagnosis of MSBP.

The mother was confronted with the situation. She remained calm and defended herself by pointing out that she was not responsible, but that her husband or mother-in-law might have tried to mislead the physicians. When she was told that her baby would be kept in the hospital until the situation was resolved and that social services would then decide on the future of the baby, she exhibited no emotional response. On the other hand, the father displayed disbelief and anger. He menacingly referred to his wife's previous operation and wound infection, and blamed the physicians for the still more ineffective handling of the child's illness. He accused them of treating his wife as a psychotic and himself as an alcoholic. He insisted that her daughter should be immediately discharged and refused to contact the social services, but later changed his mind and was persuaded to do so. The social services decided to put the child under custody of her father's sister. The mother was admitted to the psychiatry ward, where she confessed that she created the entire scenario about the rectal bleeding of her daughter. She said that it was her own menstrual blood in the beginning, but in the last incident she used a tube of blood she found in the hospital. According to her, the reason for this fabrication was a dysfunctional family life. She accused her husband of drinking alcohol, and of abusing her physically when he was drunk. She related that she needed to go out of the house whenever there was a quarrel with her husband, and found the hospital a safe enough environment to escape to.

Discussion

Asher[5] coined the term Maunchausen syndrome in 1951, in his description of patients who consistently produced false stories or symptoms about themselves to obtain needless hospital investigations and treatments. In 1977, Meadow[1] described another form of this syndrome where the parents, usually the mother, caused their children to undergo harmful hospital procedures by fabricating symptoms, which was called Munchausen syndrome by proxy. Since then, many cases have appeared in the literature[6-8].

The most common presentations seen in MSBP include any form of bleeding (hematuria, hematochezia, hematemesis, etc), seizures, central nervous system (CNS) depression, apnea, diarrhea, vomiting, fever and rash[9,10]. Our patient was brought to us with one of the most common presentations of MSBP, namely gastrointestinal bleeding. While this symptom justifies the mother's taking the child to the emergency room and is easy to fabricate, it presents a dilemma for the attending physicians, due to the fact that in order to exclude organic disease, invasive procedures must be ordered. The medical personnel contribute indirectly to the damage inflicted, by resorting to painful investigative procedures as experienced in our case up until her fifth admission.

There are some specific features of MSBP which lead to the correct diagnosis[11,12]. First, the illness is prolonged, unexplained and repetitive; observations and investigations are inconsistent with parental reports or the condition of the child. In our case, although the mother brought the baby to us every 2-3 weeks for almost 1.5 years with the complaint of rectal bleeding, the baby looked unusually healthy for a child bleeding so massively. All investigations performed to find the etiology of the bleeding were normal. Second, symptoms and signs begin only in the presence of the mother. They are conspicuously absent when the baby and mother are under strict supervision. In our case, the mother was the only witness of the bleeding. Others saw only the bloody diaper she presented. When the baby stayed in the hospital and when they were under supervision, there was no bleeding reported by the mother. Third, mothers are unusually calm for the severity of illness and not as worried as the nurses and doctors about their child's illness. Our patient's mother was very eager to stay in hospital, but she looked as if she had no anxiety about her baby's illness. She accepted every invasive procedure for her baby without any sign of worry. Fourth, there is a history of unexplained or unusual illness or death in previous children. Our mother had a very similar story that could not be confirmed, due to the unavailability of either birth or medical records of these babies. As no records were available, this information was supposed to be a part of the scenario fabricated by the mother to convince the doctors about the gravity of her daughter's illness. Fifth, more than half of the mothers have some features of Munchausen syndrome themselves[9]. The hospital records of the mother revealed an unusually prolonged wound infection after

appendectomy. However, her surgeon believed that the wound had been infected intentionally. Sixth, the fathers are usually extremely unsupportive of their wives and unaware of the fabrication of the illness[9]. Our patient's father abused alcohol. The mother admitted that she was an unhappy woman. In their last presentation to the emergency room, she stated that she had left her husband because he was drinking too much, but later had been persuaded by him to come back with a promise to cease drinking.

Some authors classify such mothers into three groups: active inducers, help seekers and doctor addicts[13]. The first group actively induces the symptoms in their children. Help seekers have social problems such as domestic violence, unhappy marriage, etc, and they use their children to avoid these problems. They are more open to psychotherapeutic intervention. The third group, doctor addicts, being more antagonistic, suspicious and paranoid, are obsessed with the goal of obtaining medical treatment for non-existent illnesses of their children. The mother of our case was classified as a "help-seeker", taking into consideration her relation with her alcoholic husband. We believe that our concern about her problems and the psychological support we provided helped her to deal with them for almost one year.

The patients with MSBP usually do not show a gross psychopathology as in this case. Proposed explanations for the illness behavior in MSBP remain to a large degree speculative. The underlying motivations for this disease are probably heterogenous and multifactorial. Expect for the search for nurturance, secondary gains, and the need for power and superiority, it is proposed that the patient with a poor sense of self can achieve a personal identity with the sick role and the pseudologia phantastica (pathological lying). The predominant feature of the mother of our patient was also pathological lying. She described herself as a police officer in the hospital and gave a history of loss of three children. The latter was serving her needs by making her doctors assume that they were faced with an unusual case of bleeding, resulting in increased attention to the baby; while the former was helping her to create the sense of identity in which she was no longer an abused wife with lifelong difficulties, but a working lady with a sick child, deserving the care of the hospital staff.

To confirm the diagnosis of MSBP, some authors recommend the use of a covert video surveillance[14], while others claim that it is unethical and should be used only in restricted circumstances[15]. We preferred to separate the mother and child or to keep them under close supervision instead of video surveillance. If the symptom of MSBP is bleeding of any origin, it is suggested to examine the blood group in the specimen[16]. In our patient, the blood grouping and DNA analysis of the blood in the diaper confirmed the diagnosis.

The management of MSBP consists of confronting the mother. Many cases respond to that kind of therapy[11]. The aim of the confrontation is to understand and respect the meaning of the symptoms in order to help. In our case, after the diagnosis was firmly established, the mother was confronted and was admitted to the psychiatry ward, where she is still under treatment. During the mother's hospitalisation period, her husband also accepted treatment. Another approach is to separate the child from the family by placing her under custody. This is especially recommended in more dangerous situations, where the abuse involves suffocation or poisoning, the child is under five years of age, or there is a history of sibling death or overt Munchausen syndrome in the mother herself[17,18]. Because our case fulfilled most of these criteria, she was reported to the social services. Interviews with all the members of the family were obtained that led to the decision to place the infant in her aunt's custody. She has been living with her aunt's family for three months now and feels safe and happy with them. After psychiatric treatment of the mother, family reunification has been tried for certain cases, but long-term follow up is necessary to ensure the child's safety[19].

Most of the mothers we meet during our practice tell the truth about their babies and their observations are very valuable. We must listen to them carefully and act accordingly, but rarely we may meet such mothers who try to deceive their physicians. If our aim is to supply a good quality of life to all children, we must be careful in the situations listed below:

- Prolonged or recurrent symptoms and/or signs which are irrelevant to patients' general health state and cannot be explained by medical professionals.

- When the mother is the only witness of the symptoms and the symptoms and/or signs disappear in the absence of the mother.
- Admissions of other children within the same family with similar features.
- Unexplained child loss within the same family.
- Mothers who are unusually calm about the illness of their children and eager to let their children undergo invasive procedures or hospitalizations.
- Mothers with unexplained physical or psychiatric diseases, symptoms or signs.

In such cases, the diagnosis of MSBP should be considered before performing invasive procedures on the child.

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Neonatal onset propionic acidemia without acidosis: a case report

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Propionic acidemia is an inherited disorder of organic acid metabolism characterized by a spectrum of clinical and biochemical findings. The usual presentation is life-threatening ketoacidosis and hyperammonemia. In this report we present a neonate with propionic acidemia presenting with prominent neurologic problems without ketoacidosis. The patient had a serum ammonia level of 3,500 µg/dl which was effectively lowered to normal values in 48 hours by peritoneal dialysis, with remarkable improvement in neurologic status. However, she developed Candida albicans peritonitis, and sepsis and died of cardiorespiratory failure. Infants who have an early onset propionic acidemia have a high mortality and morbidity rate. In conclusion, propionic acidemia should be in the differential diagnosis of patients with neurologic symptoms and hyperammonemia with or without acidosis.

Key words: propionic academia, neonate, peritoneal dialysis.

Propionic acidemia is a rare disorder of organic acid metabolism, caused by a deficiency of propionyl-coenzyme A carboxylase[1,2]. The reported incidence in Turkey is approximately 1/50,000-100,000[3,4]. Several clinical patterns of presentation have been described in neonates, older infants and children[3-5]. The disorder is frequently manifested with poor feeding, vomiting, lethargy, hypotonia, metabolic ketoacidosis and hyperammonemia in the neonatal period. Both early-and late-onset disease cause permanent neuroloic problems with markedly delayed development, seizures and cerebral atrophy[5,6]. The plasma ammonia level is related to the severity of the disease[1], and the duration of the hyperammonemic coma is an important prognostic factor for long-term sequelae. Thus, it should be treated immediately and effectively by hemofiltration or peritoneal dialysis[7,8].

We aimed to discuss the clinical and metabolic findings and management of early onset propionic acidemia, and report a patient who presented as a pure neurologic disease without acute episodes of acidosis.

Case Report

The infant, born at term by cesarean section C/S (because of a former C/S) after an uncomplicated pregnancy, was the second child of healthy consanguineous parents (first-degree relatives). Her birth weight was 3,500 g and Apgar scores were 8 and 9 at 1 and 5 minutes. Postnatal course was remarkable for progressive hypoactivity. At 13 days of age she was referred to our institution for feeding difficulties, poor sucking, 500 g weight loss, hypotonia, and hypoactivity.

On examination her vital signs were stable. Neurologic exam revealed a lethargic infant who could be awakened only by stimulation. She had minimal spontaneous activity but did not have any focal deficits. Sucking and rooting reflexes were absent and she had a weak gag reflex. Deep tendon reflexes were hypoactive. She had decreased flexor and extensor head tonus as well as decreased axial tonus. The rest of the physical examination was normal except for oral moniliasis.

On the day of admission to the neonatal intensive care unit she had an apnea and desaturation followed by an irregular respiratory

pattern. She was intubated and put on the respirator. After a few hours she had a left focal seizure and phenobarbital was started. Cranial ultrasonography was normal. EEG revealed a disorganized background activity. Following the sepsis work-up, intravenous antibiotics (ampicillin and netilmicin) were initiated empirically. Cerebrospinal fluid examination was normal. Blood, urine and cerebrospinal fluid cultures were negative.

Laboratory investigations revealed hematocrit of 38.7%, white cell count 3,500/mm^3 with 20% neutrophils and 80% lymphocytes, and platelet count 162,000/mm^3. Serum electrolytes, glucose, kidney function tests, liver enzyme levels, T_3, T_4 and TSH were all within normal limits. Urinalysis revealed moderate ketonuria. Serum ammonia concentration was 3,500 µg/dl (normal: 79-129 µg/dl). Arterial blood gas analysis and serum lactate concentration were normal (Table I). Blood and urine amino acids and urine organic acids were sent to the laboratory. Meanwhile, since serum ammonia level was very high and the clinical condition of the patient was critical, peritoneal dialysis was started to resolve the hyperammonemia. Twenty-four hours after the initiation of peritoneal dialysis, the baby started to improve neurologically.

Table I. Laboratory Findings

Parameter	Results	Normal values
Leukocyte	3,500	5,00-20,000/mm^3
Thrombocyte	40,000	150,000-450,000/mm^3
Glucose	72	50-110 mg/dl
[HCO_3^-]	20	20-25 mEq/L
Anion gap	24	<25
Lactate	1.8	0.5-2.2 mmol/L
NH_3	3,500	79-129 µg/L
Urine ketone	(+)	(-)

On peritoneal dialysis serum ammonia levels decreased to 1,200 µg/dl at the end of the first day and to 175 µg/dl at the end of the third day, but seizures and leukopenia (WBC: 2,200/mm^3) persisted. She developed thrombocytopenia (thrombocyte: 40,000/mm^3) and needed several platelet transfusions and GCSF treatment as well.

Urine organic acid study established the diagnosis of propionic acidemia with characteristic metabolites screened by gas chromatography-mass spectrometry. (Urinary propionylglycine=17.7 mmol/mol creatinine, 3 hydroxypropionate=230 mmol/mol creatinine, 2 hydroxybutyrate=27 mmol/mol creatinine). Carnitine and biotin was started and serum ammonia level was measured daily. Despite the diagnosis of propionic acidemia, metabolic acidosis had never been observed during the hospitalisation period.

On the sixth day of admission, due to malfunctioning of peritoneal dialysis catheter serum ammonia level rose to 385 µg/dl. When the catheter was replaced with a new one and peritoneal dialysis was continued, ammonia level decreased to 87 µg/dl. In the following days, despite neurologic improvement, the clinical condition worsened. The microscopical examination of peritoneal dialysis fluid showed 60 WBC/mm^3 and yeast cells, thus intravenous fluconasole was started. Peritoneal dialysis fluid culture yielded Candida albicans and the clinical condition deteriorated further. The antifungal therapy was changed to amphotericin B. Peritoneal dialysis was continued in the meantime since hemodialysis was not possible for the patient because of hemodynamic instability.

On the 17th day of admission, she was still on respirator when pulmonary hemorrhage occurred and her clinical status deteriorated. The infant died of cardiorespiratory failure the next day in spite of all supportive therapy.

Discussion

Propionic acid is an intermediate metabolite of isoleucine, valine, threonine, methionine, odd-chain fatty acids and cholesterol catabolism[7]. It is normally carboxylated to methylmalonic acid by propionyl-CoA carboxylase (PCC), which requires biotin as a cofactor. Propionic acidemia is an autosomal recessive disease caused by PCC deficiency[7]. Concentrations of propionic acid, methylcitric acid and propionylglycine are markedly elevated in the plasma and urine of infants with propionic acidemia. Definitive diagnosis can be established by measuring PCC activity in cultured fibroblasts or leukocytes. Our patient had significant elevation of characteristic metabolites in her serum and urine but the measurement of PCC activity in cultured fibroblasts was not possible during her hospitalisation.

The clinical manifestations of the disease are variable even within the same family, but generally the earlier the onset, the higher the

mortality and morbidity[5,8,9]. Hyperammonemia, ketoacidosis and thrombocytopenia are the hallmarks of propionic acidemia[7,10]. Less frequently the patient present later in life with mental retardation, choreoathetosis and seizures without acute attacks of ketoacidosis[7,10-12]. While metabolic acidosis is a key feature of early onset propionic acidemia, there are several reports of patients where metabolic acidosis was not a persistent finding[11-14]. Nyhan et al.[11] reported two patients with propionic acidemia presenting with prominent neurologic symptoms suggesting a disease of basal ganglia without episodes of ketoacidosis. These children had spastic quadriparesis, choreoathetosis and seizure disorder. Propionyl-CoA carboxylase activity was 5% of the control in each patient. Similarly, our patient had never had ketoacidosis but exhibited the clinical symptoms of hyperammonemic coma, neutropenia and thrombocytopenia.

Mild-to-moderate mental retardation is common in patients with propionic acidemia even with good compliance to therapy. Neurodevelopmental deficits are usually due to recurrent episodes of hyperammonemia and acidosis as well as long-term exposure of brain to abnormal metabolites[5].

Early diagnosis and treatment are important for better long-term prognosis[5]. Treatment of acute attack includes rehydration, correction of acidosis and prevention of the catabolic state by provision of adequate calories. Very ill patients with severe acidosis and hyperammonemia require peritoneal dialysis to remove ammonia and other toxic compounds[7,15]. Moderate hyperammonemia is common in propionic acidemia due to inhibition of urea cycle enzyme N acetyl glutamate synthetase by propionyl-CoA[16]. In our patient, ,serum ammonia level was extremely high (3,500 µg/dl) at presentation possibly due to the delay in referral of the patient (at 13 days of age) to our hospital. In our patient peritoneal dialysis was very effective in reducing the serum ammonia with concomitant clinical improvement.

Patients with propionic acidemia have frequent infections, as a result of which an acute metabolic decompensation may follow[17,18]. In our patient Candida albicons peritonitis and fungemia progressed to multiorgan failure. The patient did not respond to any therapy and died of cardiorespiratory failure.

We conclude that propionic acidemia should be included in the differential diagnosis of patients with neurologic symptoms and hyperammonemia with or without acidosis. Infants with propionic acidemia should be followed at tertiary care centers with a dialysis unit to treat hyperammonemia. Even with intensive therapy mortality is high during acute attacks.

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Metastatic endodermal sinus tumor: CT appearances

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A one-week-old boy had undergone resection of a sacrococcygeal benign cystic teratoma. At the age of 12 months, he had a serum alpha-fetoprotein level of 139,000 IU/ml and a recurrent pelvic mass which was removed, and the microscopic examination revealed endodermal sinus tumor. Postoperatively, massively enlarged inguinal lymph nodes and abdominal distention developed. Computerized tomography displayed enlarged inguinal lymph nodes, metastatic lesions in the liver, and a pelvic recurrent mass. He received BEP (bleomycin, etoposide, cisplatin) chemotherapy regimen, and a complete remission was achieved with a normal serum alpha-fetoprotein. Close follow-up and serum alpha-fetoprotein monitoring are mandatory after the resection of a sacrococcygeal teratoma.

Key words: endodermal sinus tumor, sacrococcygeal teratoma, computerized tomography, alpha-fetoprotein.

In children the sacrococcygeal region is the most common location for nongonadal germ cell tumors[5], and sacrococcygeal teratoma (SCT) is the most common tumor of the newborn[1]. Although these tumors are mostly benign, some may contain immature elements and malignant transformation may develop. Here, we report a case with a resected mature SCT which recurred at the age of 12 months as an endodermal sinus tumor (EST) with extensive abdominal disease and rather extraordinary images on computerized tomography (CT).

Case Report

A one-week-old boy had been seen in another hospital due to a sacrococcygeal mass, at which time he had a preoperative serum alpha-fetoprotein (AFP) level over 300 IU/ml and had undergone a total excision of the mass together with the coccyx. Histopathological diagnosis had been benign cystic teratoma. At the age of 12 months he was admitted to the same hospital and a pelvic CT displayed a predominantly cystic, recurrent sacrococcygeal mass which was removed totally: histopathological examination revealed EST. Two weeks after the operation, abdominal distention, hepatomegaly and massively enlarged inguinal lymph nodes were noted (Fig. 1) and he was referred to our hospital. The patient was reevaluated and an abdominal CT displayed a recurrent pelvic mass, inguinal enlarged lymph nodes and metastatic lesions in the liver (Figs. 2 and 3); serum AFP level was 139,000 IU/ml. Thoracic CT was normal and abdominal ultrasound findings were similar to abdominal CT. After receiving six cycles of BEP regimen (bleomycin, etoposide, cisplatin), abdominal CT displayed no evidence of tumor and serum AFP level was normal. However, in the following months, serum AFP level increased steadily, and the patient is still under treatment due to recurrent disease.

Discussion

The prognosis of SCTs depends on the age of the patient, surgical resectability of the primary tumor and the histological grading[4]. Patients under two months of age who have had a complete surgical excision have a favourable outlook, especially when the histological grading is low and tumor lacks malignant elements[4]. EST is the most common histological type of malignancy that develops within SCTs[5]. Recurrences occur in about 3% to 10% of cases and usually develop in about two years after the initial resection[1-3]. The presence of a malignant

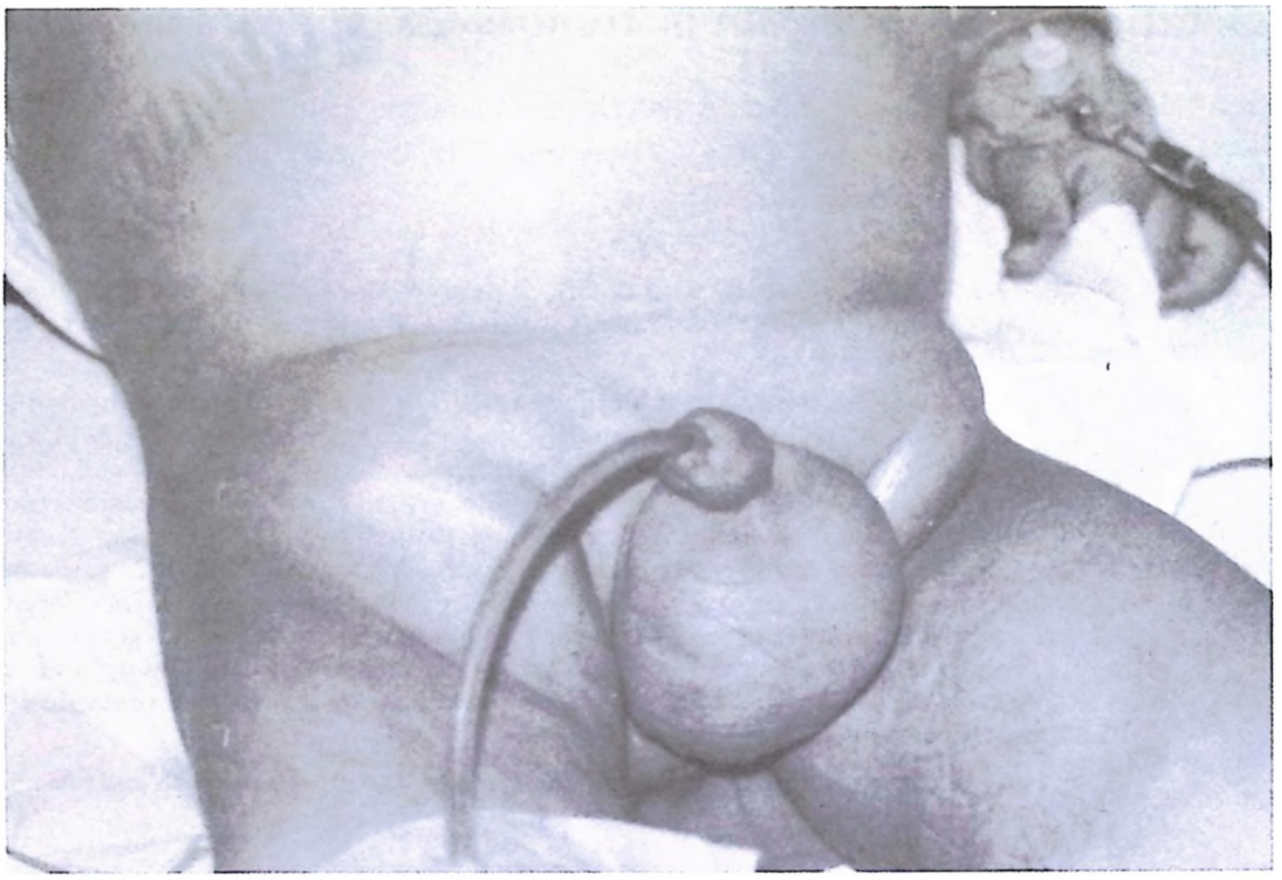

Fig. 1. Clinical appearance of the patient with abdominal distention and enlarged inguinal lymph nodes.

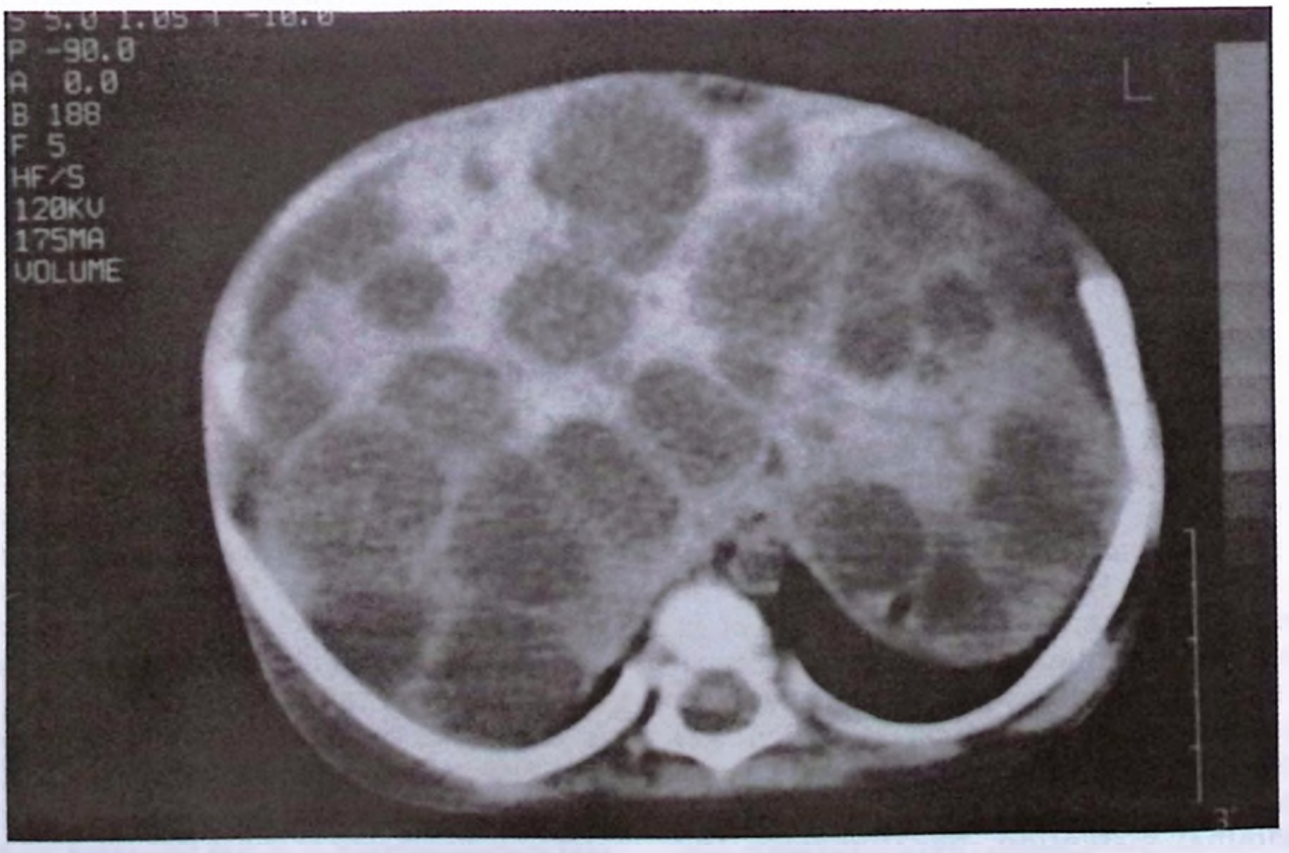

Fig. 2. Computerized tomography shows multiple metastatic lesions in the liver.

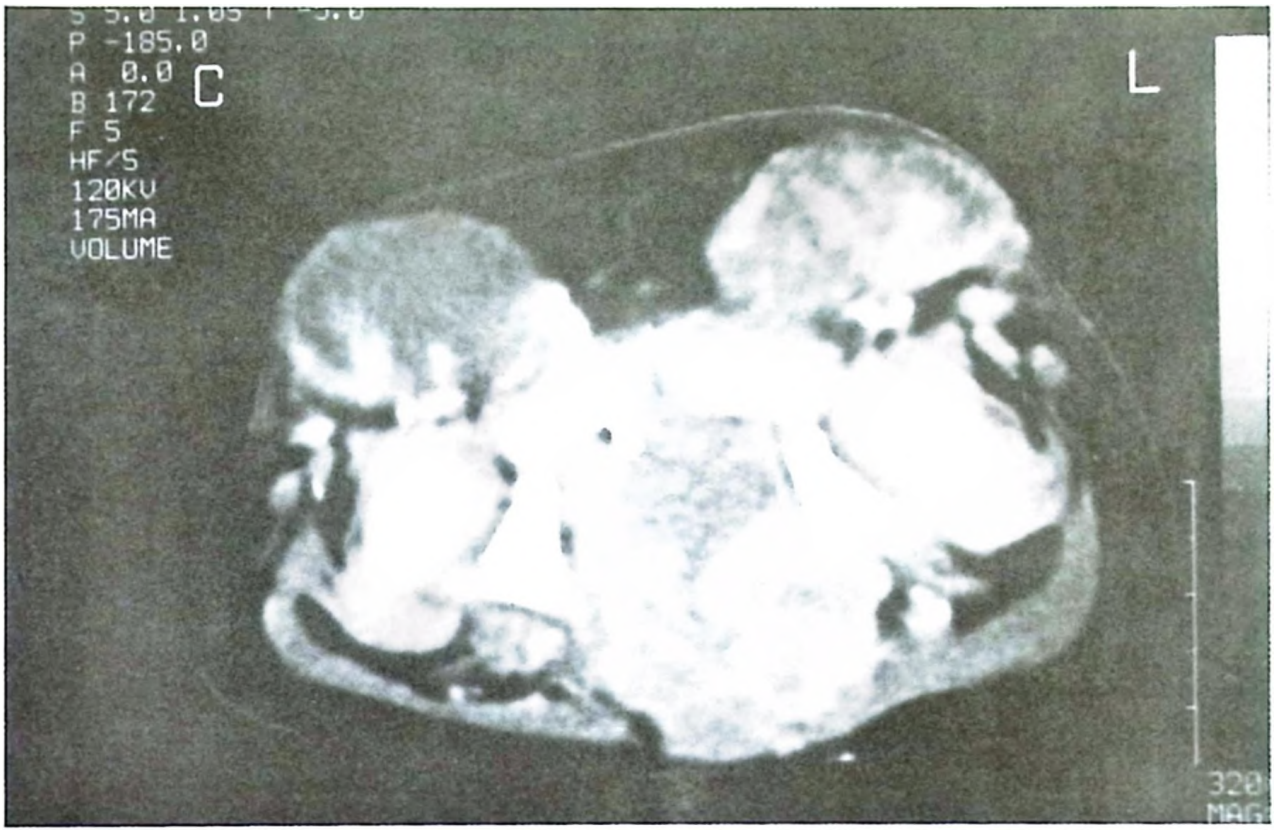

Fig. 3. Pelvic computerized tomography displays huge metastatic inguinal lymph nodes and a pelvic mass.

component is commonly associated with an elevated serum AFP level[2,5]. In a newborn patient, it may be difficult to evaluate serum AFP level as a tomur marker since it is already elevated in the neonatal period.

Our patient was not under regular follow-up after the initial operation. We did not have information regarding initial preoperative radiologic or other investigations. The massively enlarged inguinal metastatic lymph nodes and the liver filled with multiple metastatic lesions made the CT images rather extraordinary. Clinically and radiologically, we achieved complete remission and serum AFP was normal after chemotherapy. The use of multiagent chemotherapy regimens has resulted in significant improvement in the prognosis for children with EST, and overall survival has improved significantly even in children with advanced disease. After total resection of a primary SCT, an elevated AFP level is a reliable marker for a recurrence of EST. Tumor recurrence could have been detected earlier if our patient had been under regular follow-up previously. Close follow-up at least for three years with frequent physical examination, serum AFP monitoring and diagnostic imaging is necessary for all children who have undergone excision of SCT in the newborn period[1,5].

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Generalized lichen planus in childhood: is dapsone an effective treatment modality?

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SUMMARY: Başak PY, Başak K. Generalized lichen planus in childhood: is dapsone an effective treatment modality? Turk J Pediatr 2002; 44: 346-348.

Childhood lichen planus is generally atypical in appearance, prolonged in duration and resistant to therapy. Moreover, the risk in administration of systemic drugs because of several adverse effects limits their use and effective therapy remains a problem in this age group. We report a case of generalized lichen planus in a nine-year-old boy with oral mucosa and nail involvement who was treated with dapsone.

Key words. lichen planus, childhood, treatment, dapsone.

Lichen planus (LP) is a rare inflammatory disease of unknown etiology and its incidence peaks in adulthood. Only 2% to 3% of all cases were reported to be described in children under 14 years, and they may present atypical clinical findings[1-4]. Mucosal involvement and nail abnormalities are rare and management of treatment is difficult in the pediatric patient[5,6]. Generalized lichen planus was observed in a nine-year-old boy and treated with dapsone during a period of approximately one years.

Case Report

A nine-year-old boy presented with a three-month history of itchy lesions on the trunk and limbs. There was no family, atopy, drug or vaccination history except for unsuccessful response to systemic steroids previously used for his skin lesions. On examination, brown to violaceous, slightly scaly papules all over the trunk, and coalescing in the lumbar region and limbs were observed (Fig. 1a). Besides leukokeratotic buccal mucosal involvement, the first toenails of both feet appeared dull and opaque with slight longitudinal ridging (Fig. 2a, b). Laboratory analysis including complete blood count, urinalysis, biochemical and hormonal parameters as well as markers for hepatitis were in normal ranges. Mycologic examination of the toenails was negative and radiology revealed no constitutional abnormalities. Histopathologic findings of the biopsy from the skin lesions were consistent with LP (Fig. 3). Dapsone, 50 mg daily (1.5 mg/kg/day) was started. Itching subsided within one month and disappeared in the second month of treatment. Skin lesions, especially on the trunk, started flattening after three months. At the end of eight months of therapy, there were still active lesions present on the limps. The dosage was increased to 75 mg daily (2.5 mg/kg/day) and new lesions stopped appearing within two months. The lesions were totally cleared at the end of five months of treatment with the same dosage, leaving hyperpigmented macules on the lower extremities (Fig. 1b).

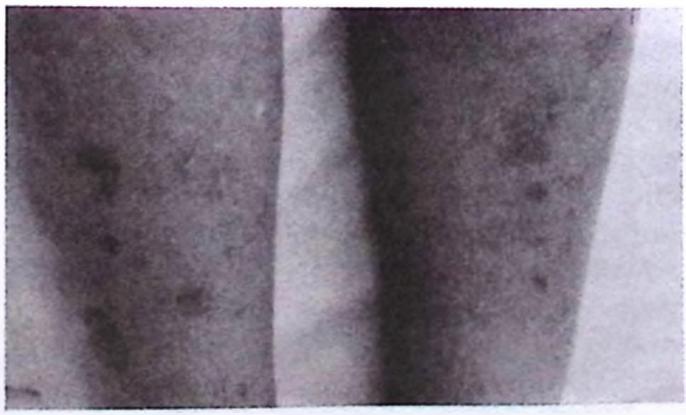

Fig. 1a: Lesions on the legs before treatment.

Poster presentation at the 9th EADV Congress, Geneva 2000.

Fig. 1b. Posttreatment view of the legs.

Fig. 2a. Buccal mucosa involvement.

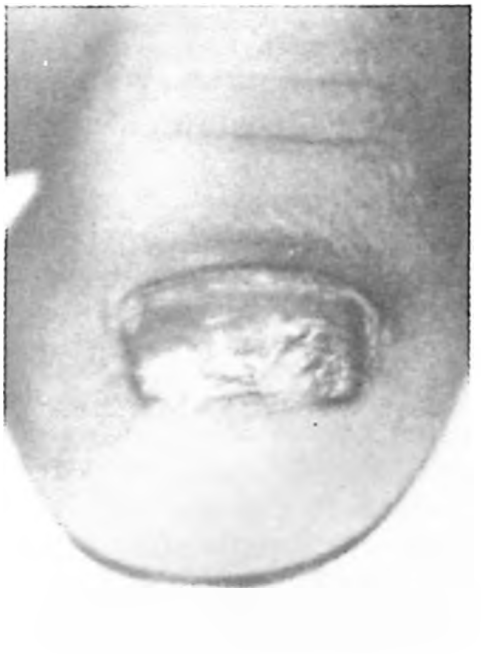

Fig. 2b. Nail abnormalities.

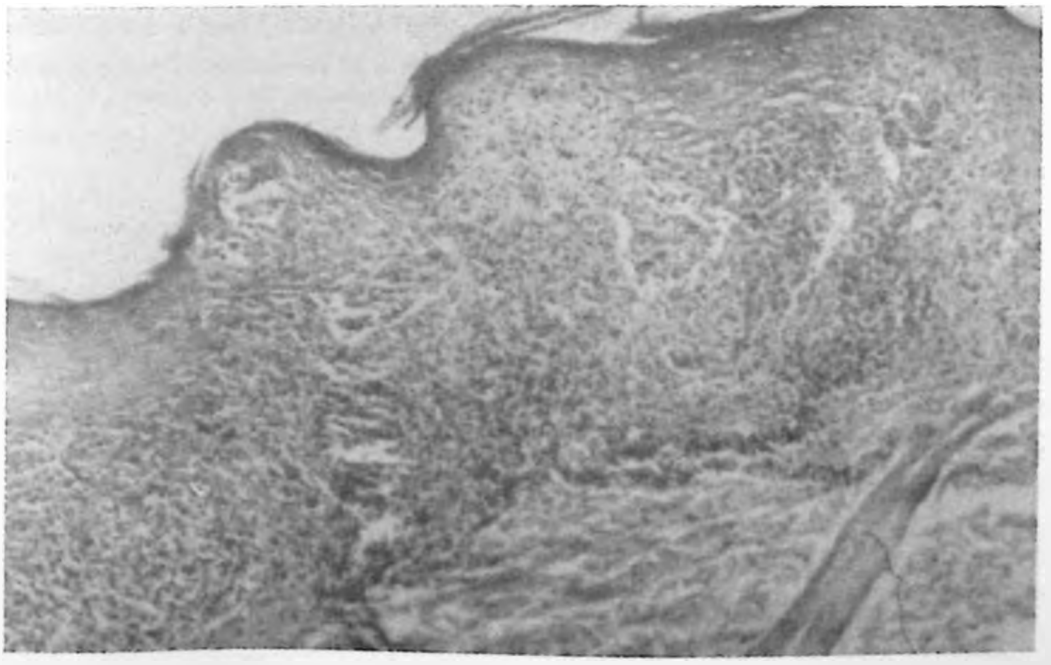

Fig. 3. Lichenoid infiltration, irregular rete ridges of epidermis and focal hypergranulosis (H&E, x40).

Discussion

The incidence of classic LP lesions with violaceous, polygonal, flat-topped papules has been reported to occur in childhood cases by Kanwar[2] and Kumar et al.[6] at rates of 76% and 56% respectively. In contrast, Milligan et al.[3] experienced only one classical case out of six presenting with childhood LP. Our case was a classical generalized LP with prominent Köbner's phenomenon.

Oral LP is rare in children. Very few of the children with cutaneous lesions were reported to have oral LP simultaneously. Of 17 patients, only one had violaceous lesions over the lips in Kanwar's series[2]. In the study of Kumar et al.[6], oral mucosal involvement with 20-nail dystrophy was detected in only one patient among 25 children. Three girls, having buccal and/or lingual mucosal LP without skin involvement were reported. Two of them were manifested with erosive lesions and one was in reticulopapular pattern[7].

Lichen planus (LP) of the nails in children has been rarely described[2,8]. Nail abnormalities characterized by longitudinal ridging, pterygium and scarring atrophic dystrophy were reported in four children by Colver et al.[9]; longitudinal ridging and onycholysis were described by Peluso et al.[10] in the absence of skin or mucous membrane lesions. Twenty-nail dystrophy of childhood due to LP was also reported[11]. However, in the series of Milligan et al.[3], only one patient was described to have skin lesions associated with nail changes. To our knowledge, our case was different than the previous reports with diffuse skin involvement accompanied by oral mucosal and nail lesions. However, a nail matrix biopsy would have been helpful to confirm the nail involvement.

Therapeutical approach consists of systemic antihistamines and corticosteroids[1-3,10], acitretin[5], dapsone[6] and griseofulvin[5] for extensive involvement. Considering the adverse effects of oral steroids and the risk of premature epiphyseal closure due to acitretin in children, dapsone was preferred in this case.

Lichen planus (LP) may follow a prolonged time-course in children, and there are some variants resistant to conservative therapies[3,5]. Although it was reported that 94% of cases cleared in less than a year as in adults[2], we had to continue the treatment up to one year, increasing the dosage because of the progression of the lesions. We observed difficultly in controlling the lesions particularly on the lower extremities. Oral mucosal lesions were cleared and nail changes were slightly improved with dapsone therapy. In addition, no adverse effects were detected. We concluded that dapsone may be an alternative treatment modality, especially to alleviate pruritus, and that it is safe as well, even in long-term use. However, treatment of a series of patients or blinded and controlled studies would be necessary to prove that this treatment modality for childhood lichen planus is safe and effective.

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Physiotherapy results in a baby with congenital lymphedema: a follow-up study

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SUMMARY: Akbayrak T, Çıtak İ, Demirtürk F, Kerem M, Akarcalı İ. Physiotherapy results in a baby with congenital lymphedema: a follow-up study. Turk J Pediatr 2002; 44: 349-353.

Physiotherapy results of a 6.5-month-old baby with congenital lymphedema in the lower limbs are presented in this study. Her motor developmental level and reflexes were evaluated and test results did not show any abnormal findings. Assessment of limb volume included circumferential and volumetric measurements, and photographs showing the changes in macroscopic view are presented. The physiotherapy program consisted of manual lymphatic massage, remedial exercises, multilayered inelastic compression bandaging, meticulous skin care and education of parents. The treatment lasted for 2.5 months, five days per week. Evaluations were done immediately after treatment and six months after treatment. The evaluations showed reduction in swelling after the treatment and this reduction continued during the follow-up period. It can be concluded that this physiotherapy program reduces the swelling in a baby with congenital lymphedema. Further studies are required in order to see the effectiveness of this therapy program in a greater number of subjects.

***Key words:** congenital lymphedema, manual lymphatic massage, remedial exercises, bandaging.*

Lymphedema is the chronic swelling of one or more extremities resulting from a malformation of the lymphatic system, causing lymph stasis and accumulation of interstitial fluid[1,2].

Lymphedema can be classified as primary (idiopathic) or secondary[1,3]. Primary lymphedema is related to in utero developmental defect and affects 1.15/100,000 persons younger than 20 years of age[1]. Although in reality present at birth, it is described according to age at presentation: congenital (present at birth to 1 year), praecox (from 1-35 years), and tarda (occurring after the age of 35 years)[2]. Primary lymphedema leads to disabling and disfiguring swelling of the extremities[4].

Lymphedema, regardless of the etiology, is essentially incurable and can a negatively effect physical and psychological well being[5]. Different therapy approaches exist for lymphedema which serve to contain swelling[6]. The objectives of treatment are to reduce swelling, restore shape, and prevent inflammatory episodes, e.g. recurrent cellulitis[6]. There are essentially three main approaches to lymphedema treatment: physical therapy, drug therapy and surgery[6]. The principle of physical therapy is to reduce excessive capillary filtration and improve drainage of interstitial fluid and macromolecules from congested regions to normally draining lymph node sites[6]. This is achieved through a combination of compression, exercise and massage[6].

This study presents the early and long-term physiotherapy results of a 6.5-month-old baby with congenital lymphedema in her lower limbs.

Case Report

The swelling of the lower limbs of the baby was first noticed by her parents at two months of age. She was examined by pediatricians and diagnosed as congenital hymphedema. She was then referred to Hacettepe University, School of Physical Therapy and Rehabilitation, Connective Tissue Manipulation and Pediatric Rehabilitation Units.

Physical characteristics of the subject were recorded (Table I). The subject was initially evaluated with regard to motor developmental and reflex levels as described by Bobath[7], and the test results did not show any abnormal findings.

Evaluation of the limb volume consisted of volumetric measures using limb submersion in water, and circumferential measures of limbs at various points[8-11] (Table II). The evaluations were performed at first visit, after the 2.5-month treatment course and at the follow-up visit (6 months after treatment), in order to assess changes in the amount of swelling. Changes in macroscopic view of the lower limbs of the baby were followed with photographs (Figs. 1-3).

The physiotherapy program included manual lymphatic massage, multilayered inelastic compression bandaging, remedial exercises and meticulous skin care[6,12,13]. The treatment lasted for 2.5 months, five days per week. The mother was also educated about these methods and was advised to repeat them at home after each treatment session and during the follow-up period.

The pre-treatment, posttreatment and follow-up values of circumferential measurements are presented in Table II and volumetric measurement values in Table III. The values reflected reduction in swelling. This reduction can also be observed from the photographs (Figs. 1-3). Motor developmental and reflex levels were repeated after the treatment and at the follow-up visit. Test results of all evaluations showed no abnormal findings.

Table I. Physical Characteristics of the Subject During the Follow-Up Period

Physical Characteristics	Height (cm)	Weight (g)	Head circumference (cm)
First visit	69	8550	43
After treatment	71	8750	44
Six Months after treatment	75	9400	46

Table II. Circumferential Measurement Values (cm)

Measurement levels	Before treatment (R)	After treatment (R)	Control (R)	Before treatment (L)	After treatment (L)	Control (L)
Malleolus	14	14	14	15.5	15	13.3
M+2 cm	15.1	15	13.2	15.6	15	13.2
M+4 cm	18	16	15.5	18.5	17	15.2
M+6 cm	19.1	18	17	19.5	18	16
M+8 cm	18.5	18	17.3	18.6	17.5	17.5
M+10 cm	–	–	16.5	–	–	17
M+12 cm	–	–	16.5	–	–	16.5
M+14 cm	–	–	19	–	–	17.5
Knee	20	20	19.1	20.1	20	19
K+2 cm	22.5	21	19.1	21	21	19.2
K+4 cm	23	23	21	24	22.3	20.2
K+6 cm	25.5	23.6	22	25	23	21.2
K+8 cm	–	–	22	–	–	21.2
K+10 cm	–	–	22.5	–	–	22.5

M: Malleolus.
K: Knee.
R: Right lower limb.
L: Left lower limb.

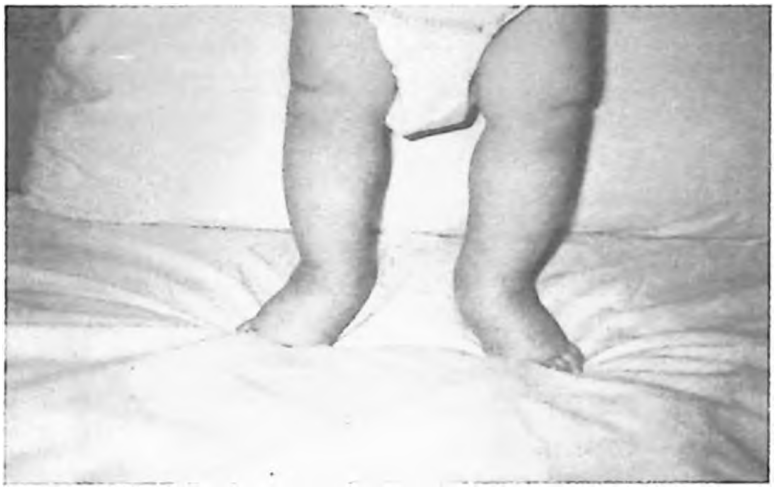

Fig. 1. Macroscopic view of the subject before treatment.

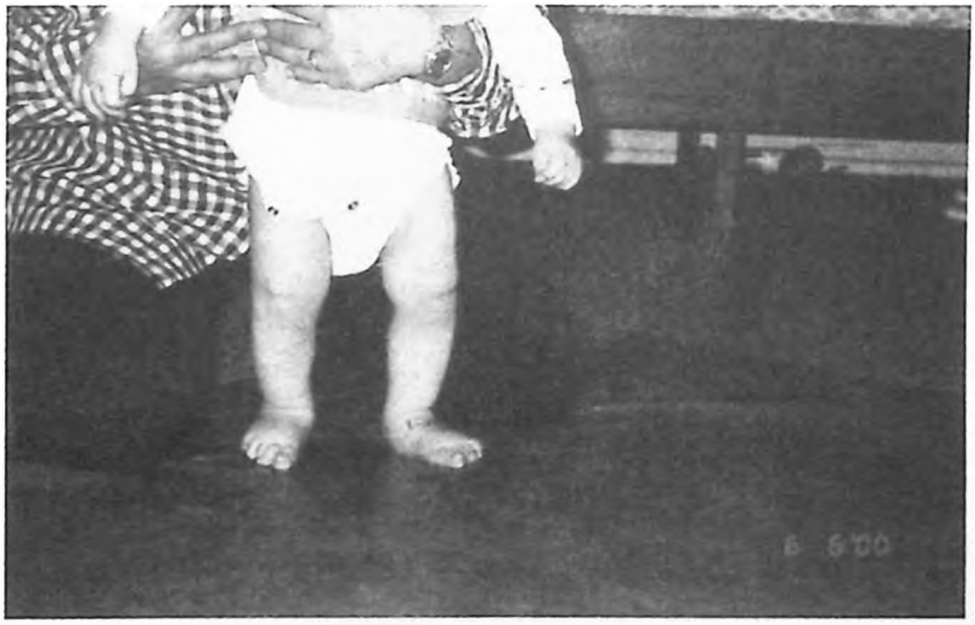

Fig. 2. Macroscopic view of the subject after treatment.

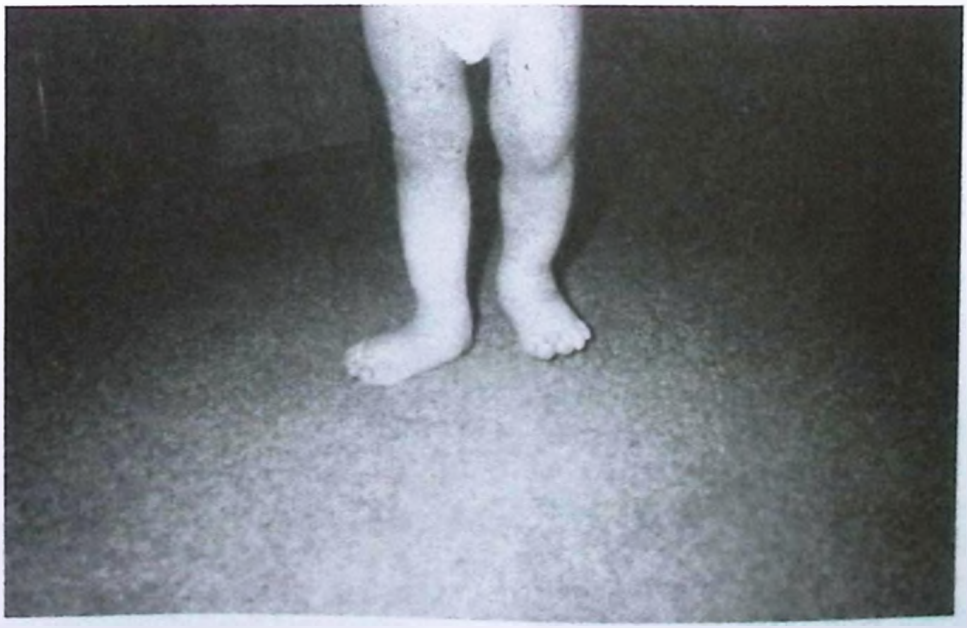

Fig. 3. Macroscopic view of the subject six months after treatment.

Table III. Volumetric Measurement Values (ml)

Volumetric values	Right lower limb	Left lower limb
First visit	250	250
After treatment	200	175
Six months after treatment	125	125

Discussion

Studies about congenital lymphedema are usually related to the diagnosis, etiology, pathogenesis, evaluation and surgical management of the problem[14-16]. Recent foreign literature about physiotherapy results has usually been related to post-surgical lymphedema[17-22].

The reported patient was diagnosed as congenital lymphedema. When the baby was referred to us, we planned a physiotherapy program in the light of the related literature. To our knowledge, there has not been any study performed in Turkey investigating the results of physiotherapy programs in such a case. We performed the physiotherapy program as outlined in the case report section and found improvement as shown in Tables II and III.

Ohkuma[10] investigated the effectiveness of microwave and elastic dressing in 30 cases of primary and secondary lymphedema. He concluded in his study that treatment of lymphedema by microwave and elastic dressing appears satisfactory, particularly when given at an early stage of the disease. Although the case characteristics and therapy approaches of our study differed from the work of Ohkuma, we also had the advantage of starting therapy at an early phase, and this may be one of the factors of success in our case.

Ko et al,[13] reported that complete decongestive physiotherapy, including manual lymphatic massage, multilayered inelastic compression bandaging, remedial exercises, and meticulous skin care, is a highly effective treatment for both primary and secondary lymphedema[13]. Our study included similar approaches for reducing the limb volume of the baby. We noticed that manual massage was well accepted by the baby, so this method can be used easily by therapists and parents. The reaction of the baby to bandaging was not as positive as it was to the massage; however, we advised the parents to use it as it was taught, while carrying the baby daily and during bed rest. Since the case was an infant, remedial exercises were presented as playing activities and tactile stimulation was used for facilitation of the pumping activity of muscles. Skin care was important because the skin of the lower limbs was dry and stretched. We thus advised the parents to keep the skin moist and clean.

To our knowledge, this is the first study investigating a physiotherapy program result in such a case. Nevertheless, further studies are required in order to see the effectiveness of this therapy program in a greater number of subjects.

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Visceral leishmaniasis and Coombs' positive hemolytic anemia: a rare association in an infant treated with liposomal amphotericin B

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SUMMARY: Dilber E, Erduran E, Işık Y. Visceral leishmaniasis and Cooms' positive hemolytic anemia: a rare association in an infant treated with liposomal amphotericin B. Turk J Pediatr 2002; 44: 354-356.

Visceral leishmaniasis is a worldwide, disseminated intracellular protozoal infection that usually manifests by fever, hepatosplenomegaly, anemia, thrombocytopenia, leukopenia and hypergammaglobulinemia. Although anemia is a usual finding, Coombs' positive hemolytic anemia has rarely been reported in association with this disease. Pentavalent antimonials have been the preferred treatment for this disease for decades, but increasing numbers of treatment failure with antimony are being reported. Liposomal amphotericin B is a new drug which is highly efficacious in the treatment of visceral leishmaniasis and produces minimal toxicity. Here we report an infant with visceral leishmaniasis associated with Coombs' positive hemolytic anemia who was successfully treated with liposomal amphotericin B.

Key words: visceral leishmaniasis, Coombs' positive hemolytic anemia, liposomal amphotericin B.

Visceral leishmaniasis (VL) is a protozoal infection that infects and multiplies in macrophages of liver, spleen and bone marrow[1]. It usually manifests by fever, hepatosplenomegaly, anemia, thrombocytopenia, leukopenia, and hypergammaglobulinemia, and may cause a lethal disease if untreated.

Anemia, leukopenia and thrombocytopenia are the main hematologic abnormalities commonly seen in VL[1,2]. It has been confirmed that during the active phase of VL , the erythrocyte life span is shortened[2,3]. At this time, erythrocytes have been shown to be agglutinated by anti-complement and anti-non-γ-globulin (direct Coombs' test) sera. It has been postulated that an autoimmune mechanism was the likely explanation for the reduced erythrocyte survival in kala azar[3]. The pentavalent antimonials are the first-line drug in the treatment of VL[1,4]. They may cause serious toxicity especially on the heart and kidney and there is also increasing resistance to these drugs[4-6]. Liposomal amphotericin B (L-AmB) is a highly effective anti-leishmanial drug and causes less toxicity[4,7-10]. It has been successfully used especially in the treatment of resistant cases[4-6].

Here we report successful treatment with L-AmB of an infant with VL associated with Coombs' positive hemolytic anemia and discuss the effect of L-AMB in the treatment of VL.

Case Report

An eight-month-old girl was admitted to our clinic from Torul-Gümüşhane because of intermittent fever, anemia and hepatosplenomegaly. On admission she was pale and had a distended abdomen. Temperature was 37.6°C, liver was palpable 4 cm below the right and spleen 7 cm below the left costal margin. The laboratory investigation revealed a hemoglobin of 5 g/dl, leukocytes $9x10^9$/L, with 56% neutrophils, 40% lymphocytes, and 4% monocytes, platelet count of $50x10^9$/L, and reticulocyte count of 12%. Coombs' test was positive, albumin was 1 g/dl and globulin was 5.6 g/dl.

Initially, high dose methylprednisolone (HDMP-30 mg/kg for three days, 20 mg/kg for four days) was initiated for the treatment of Coombs' positive hemolytic anemia. During one week of treatment no improvement was noted either in clinical findings or laboratory

parameters, and a slight intermittent fever also appeared. At this time, bone marrow aspiration was done and multiple Leishmania amastigotes were shown in macrophages (Fig. 1). She was then given intravenous L-AmB at a dose of 3 mg/kg daily as an infusion over an hour and continued for 30 days. With this treatment hemoglobin increased to normal value and Coombs' positivity disappeared on the 25th day; platelets increased to normal on the 5th day. No Leishmania amastigote was seen on bone marrow aspirate at the 30th day of L-AmB treatment. At this time liver and spleen were palpable 1 and 2 cm below the costal margin, respectively. During treatment a transient hypokalemia was noted. Five months after completion of treatment the patient was healthy and all laboratory tests were normal.

Discussion

Visceral leishmaniasis is a disseminated intracellular protozoal infection that occurs worldwide. Infection of the macrophage of the reticuloendothelial system results in VL that is clinically present as fever, hepatosplenomegaly and pancytopenia[1-3]. Anemia, leukopenia and thrombocytopenia are the main hematologic abnormalities commonly seen in VL. The anemia appears to be due to a combination of factors, including hemolysis, marrow replacement with Leishmania-infected mononuclear phagocytes, hemorrhage, splenic sequestration of erythrocytes and hemodilution. In addition, reversible myelodysplasia has been reported in association with VL[11]. Although anemia is a usual finding, Coombs' positive hemolytic anemia has rarely been reported in association with VL[2,3,12]. In a previous report it was suggested that an autoimmune mechanism was the likely explanation for reduced erythrocyte survival in kala azar[2]. In that study the red blood cells of three patients with kala azar gave a positive anti-non-γ-globulin reaction, and agglutination with anti-complement sera was also demonstrated in two patients at the time of proven reduced erythrocyte survival. In our case the association of VL and Coombs' positive hemolytic anemia was not recognized initially, and HDMP was initiated for the treatment.

In Turkey, VL is endemic in the southeast region[13]. Another region where VL is sporadically reported is Torul-Gümüşhane[13]. In a previous report, it was suggested that a landslide that occurred in 1988 could have led to the development of climate conditions favourable to the growth of sandflies in this region[14].

Pentavalent antimonial agents have been the preferred treatment for VL for decades, but an

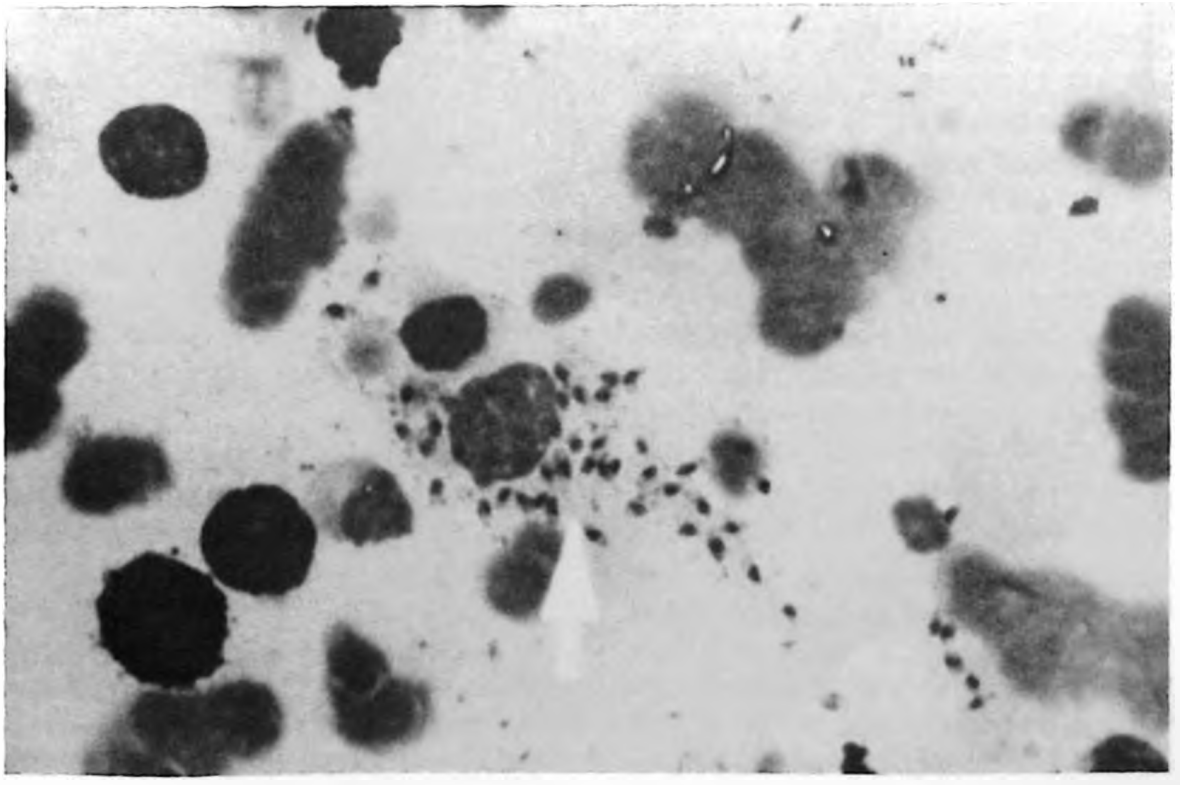

Fig. 1. Multiple leishmanial amastigotes in bone marrow aspiration.

increasing number of treatment failures with this drugs are being reported throughout the world[4-6]. These drugs may also cause significant toxicity, particularly to the heart and kidney[6]. One of the alternative drugs that has been shown to be the most active anti-leishmanial agent in use is L-AmB[4,7-10]. It is a new and well tolerated drug for VL. It is also highly lipophilic and selectively concentrates in reticuloendothelial tissue, the site of disease in the case of VL[7,8]. It is an important alternative especially in patients who did not respond to conventional pentavalent antimony therapy given alone or in combination with other agents[7]. After an initial HDMP treatment, L-AmB was initiated and continued for 30 days. With this treatment laboratory abnormalities improved and hepatosplenomegaly decreased. Treatment with L-AmB may cure leishmanial infection even in a shorter time. In this case initial HDMP treatment may have prolonged the treatment period. The only side effect noted in our case was transient hypokalemia.

This is one of the youngest patients with VL associated with Coombs' positive hemolytic anemia who was successfully treated with L-AmB. We believe this drug may be the first choice of treatment in VL even in very young infants.

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Osteochondritis dissecans in a patient with hyperimmunoglobulin E syndrome

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SUMMARY: Kılıç SŞ, Sanal Ö, Tezcan İ, Ersoy F. Osteochondritis dissecans in a patient with hyperimmunoglobulin E syndrome. Turk J Pediatr 2002; 44: 357-359.

Hyperimmunoglobulin E syndrome (hyper-IgE) is a rare immunodeficiency disease associated with recurrent pyogenic infections, chronic eczematoid dermatitis and osteopenia.

We present here a 13-year-old girl with hyperimmunoglobulin E syndrome, who developed osteochondritis dissecans (OCD) of the lateral femoral condyle, which is rare. Osteopenia, which is frequently associated with hyper IgE, may predispose the patient to the development of OCD.

Key words: bone abnormalities, hyperimmunoglobulin E syndrome, osteochondritis dissecans, osteopenia.

The hyperimmunoglobulinemia E (hyper-IgE) syndrome is a rare primary immunodeficiency characterized by markedly elevated serum IgE levels, chronic dermatitis, recurrent sinopulmonary infections, recurrent severe skin abscesses generally caused by Staphylococcus aureus, coarse facial features and growth retardation[1]. Osteochondritis dissecans (OCD) is a disease characteristically affecting the epiphysis, with subsequent separation and fragmentation of the subchondral bone in association with disruption of the overlying articular cartilage[2]. Although bone abnormalities such as osteopenia and recurrent fractures are present in most patients with the hyper-IgE syndrome, OCD has not been reported yet in these patients.

Here we present a patient having hyper-IgE syndrome who developed osteochondritis dissecans o the lateral condyle of the right femur.

Case Report

The female patient was first admitted to Hacettepe University İhsan Doğramacı Children's Hospital at two years of age with the complaints of recurrent pneumonia, skin abscesses and oral candidiasis. She was the third child of nonconsanguineous parents, and her two brothers were healthy. Physical examination showed coarse facial appearance (Fig. 1) and multiple skin abscesses. Her serum immunoglobulin G, A, and M levels were normal with an elevated IgE of 2,500 IU (4-269 IU/ml). In vitro lymphocyte proliferative responses to mitogens, neutrophil chemotaxis, and antibody responses to polio antigens were found to be normal. Absolute eosinophil count was 464/mm^3. Bone density was found more than two standard deviations below the normal values for age and sex. She was put on prophylactic daily trimethoprim-sulfamethoxasole therapy. Her growth and development were within normal limits and she experienced occasional minor skin abcesses and upper respiratory tract infections during follow-up. At age 11 years, the patient was hospitalised for the treatment of right superior lobe abscess of the lung, and was treated with antistaphylococcal antibiotics.

She was admitted to the hospital with the complaints of pain, swelling and locking on her right knee at 13 years of age. Clinical examination revealed effusion of the right knee. X-rays of the right knee (Fig. 2) showed a fragmentation of the lateral condyle of the femur. The loose body was removed surgically. When she was seen two months after surgical

Fig. 1. Characteristic facial appearance of our patient with hyperimmunoglobulin E syndrome.

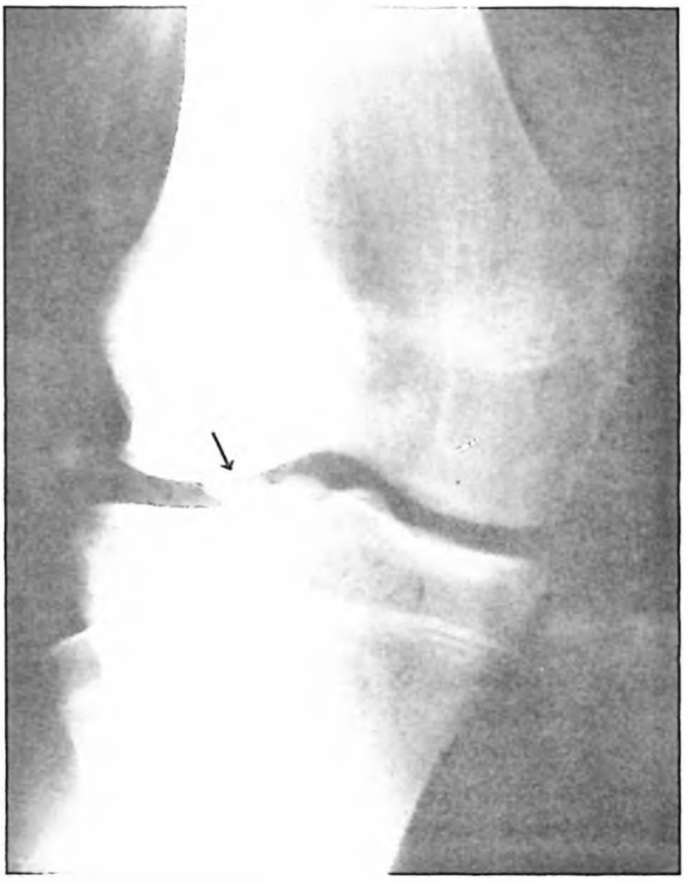

Fig. 2. Plain X-ray showing osteochondritis dissecans of the lateral femoral condyle.

intervention she did not have any complaints and clinical examination was normal. A full range of movement of right knee was possible.

Discussion

Osteochondritis dissecans (OCD) has been called "quiet necrosis". The peak frequency occurs during the teenage years, with a mean age of 15 years. The frequency in males has been reported to be two or three times more than that of females[3]. The right knee is involved slightly more often than the left, and in 7-25% of cases the lesion is bilateral[4]. All prior reports have documented that the intercondylar aspect of the medial femoral condyle was the predominant site of this lesion, with osteochondritis in the lateral condyle accounting for approximately 15-20% of all affected knees[5]. The clinical management of OCD depends largely on the mechanical stability of the osteochondral fragment. Surgical removal or internal fixation of the fragment is indicated when the lesion is dignosed before or after epiphyseal closure and there is objective evidence of looseness of the fragment as well as functional disability[6]. Surgical treatment relieved the symptoms in our patient

There are many theories regarding the cause of the disease, and ischemic necrosis, trauma, congenital alterations in bone or cartilaginous development (or both), endocrine alterations (Fröhlich's syndrome, thyroid or sex hormone abnormalities) and genetic predisposition have been implicated[3]. We could not detect any trauma history, genetic predisposition or endocrine abnormalities in our patient.

In Lindholm's series[7], osteoporosis was observed in three out of 20 patients with OCD. One of them had immobilization for four months; the others had osteonecrosis due to continuous corticosteroid therapy or chronic alcoholism. None of the reported risk factors was presented in our patient but she was diagnosed as hyper-IgE syndrome associated with osteopenia. Leung et al.[8] suggested that monocytes from patients with hyper-IgE syndrome are activated to resorb bone via products of the prostaglandin synthase

(cyclooxygenase) pathway. The activation of cells in the monocyte-macrophage family to resorb bone may contribute to osteopenia observed in hyper-IgE syndrome. Fifty-seven percent of the patients with hyper-IgE have bone fractures[9]. It seems that osteopenia caused by hyper-IgE syndrome is a favourable etiologic factor of OCD.

Both pediatricians and orthopedists caring for patients with hyper-IgE syndrome should be aware of the associated bone abnormalities.

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Ankyloblepharon filiforme adnatum (AFA) associated with trisomy 18

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SUMMARY: Tüysüz B, Ilıkkan B, Vural M, Perk Y, Ankyloblepharon filiforme adnatum (AFA) associated with trisomy 18. Turk J Pediatr 2002; 44: 360-362.

Ankyloblepharon filiforme adnatum (AFA) is a rare congenital anomaly which is characterized by single or multiple strands joining the upper and lower lids. Its etiology is unknown. A girl with a gestational age of 38 weeks is presented. It was interesting to find an association of AFA with hypoplasia of hair, eyelashes, eyebrows and nails and a karyotype of trisomy 18 in this case.

Key words: ankyloblepharon filiforme adnatum, trisomy 18.

Ankyloblepharon filiforme adnatum (AFA) is a rare congenital anomaly which is represented by single or multiple strands of tissue, joining the upper and lower eyelids[1]. Although this anomaly has been reported as an isolated malformation, it may also be found in association with other anomalies or as a part of a defined syndrome[2]. AFA associated with trisosmy 18 was first reported by Clark and Patterson[3], and six other cases have been published[4,5]. We present a case with AFA, whose karyotype revealed a trisomy 18 syndrome and in whom we found clinical signs of both trisomy 18 and Hay-Wells syndrome of ectodermal dysplasia.

Case Report

A girl was born by cesarean section at 38 weeks gestation to a 42-year-old gravida 7 para 6 mother as a first child from the second marriage of the mother. Two of her brothers from the first marriage had died, and their etiologies are still unknown. Her mother had four healthy children from the first marriage. The pregnancy was uncomplicated but the baby had Apgar scores of 0 at 1 minute and 6 at 5 minutes. After a thick meconium had been aspirated from the larynx she was intubated and ventilated by airbag. Birth weight was 2650 g (10^{th} percentile), length was 45 cm (3^{rd} percentile), and head circumference was 34 (50^{th} percentile).

Sparse hair, absent eyebrow and eyelash, broad nasal bridge, short palpebral fissure, two narrow bands on the right and one thicker band on the left eye joining the upper and lower eyelids (Fig. 1), low-set ears, micrognathia, hypoplastic nails (Fig. 2), hypoplasia of labia majora, erosion between anus and external genitalia, hyperpigmentation of anal and perineal regions (Fig. 3), short and dorsiflexed hallux, rocker-bottom feet (Fig. 4) and 3/6 systolic murmur were detected on physical examination.

Cranial ultrasonography showed a cystic expansion of the posterior fossa and renal ultrasonography was normal except for a minimal degree of ectasia of the right kidney. On echocardiography ventricular septal defect (VSD), atrial septal defect (ASD), and patent ductus arteriosus (PDA) were detected. Peripheral lymphocyte karyotype in 20 different metaphases showed 47, XX, +18 (Fig. 5). The infant died on day 10 after a cardiac arrest.

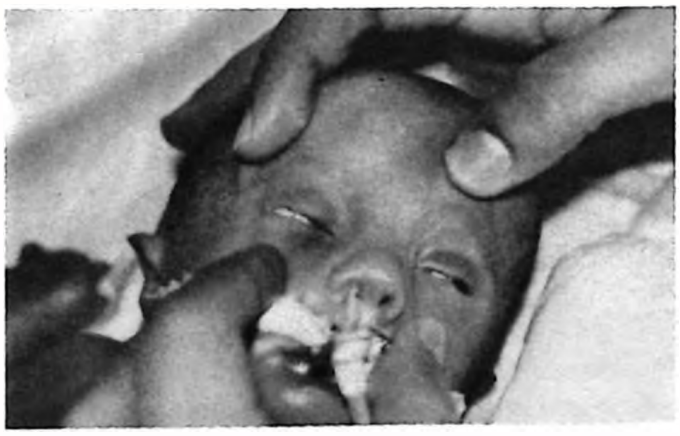

Fig. 1. Facial anomalies including absent eyebrows and eyelashes, low-set ears and strands of tissue joining the upper and lower eyelids.

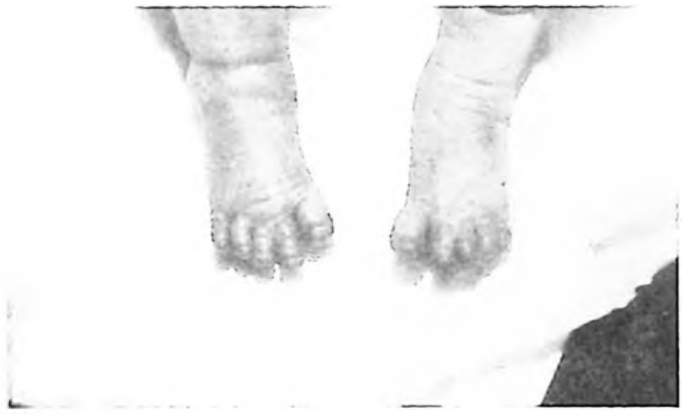

Fig. 2. Hypoplastic nails.

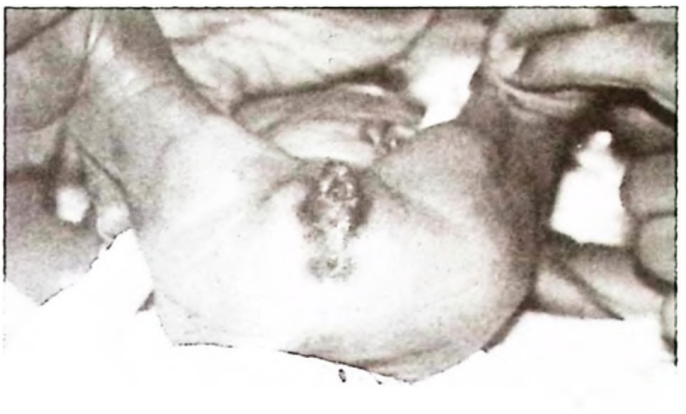

Fig. 3. Erosion between anus and external genitalia, hyperpigmentation of anal and perineal regions.

Fig. 4. Short, dorsiflexed hallux and rocker-bottom feet.

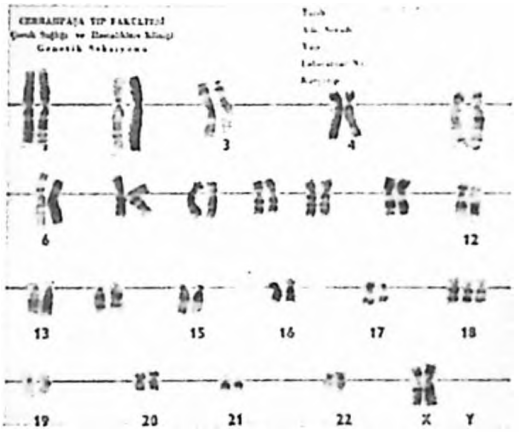

Fig. 5. Karyotype of the patient.

Discussion

Classification of AFA into four subgroups by Rosenman et al.[2] is as follows: Group I includes sporadic cases without any associated defects. Group II is also sporadic, but AFA is found in association with cardiac, gastrointestinal and central nervous system (CNS) defects. Group III represents cases of AFA in association with ectodermal syndromes (ankyloblepharon-ectodermal dysplasia-clefting-AEC syndrome). Group IV represents cases of AFA with cleft lip and palate defects in the patient or in the extended family. The seven previous reports describing the association with trisomy 18 suggested a fifth subgroup of AFA[5], which is also how we classify our case. AFA may also be associated with Fraser's syndrome, Van der Woude syndrome and popliteal pterygial syndrome[6,7].

In our patient, association of AFA with hypoplasia of hair, eyelashes, eyebrows and nails suggested Hay-Wells syndrome of ectodermal dysplasia. Presence of perineal erosion and hyperpigmentation in the same region supported our diagnosis. When we reviewed other syndromes in association with AFA, we found that clinical signs of our patient, such as short palpebral fissure, nail hypoplasia, severe cardiac malformations, short and dorsiflexed hallux and rocker-bottom feet had also been reported in trisomy 18, but typical cranio-facial signs (narrow bifrontal diameter, prominent occiput) and hand malformations (clenched hand, tendency for overlapping of index finger over third, and fifth finger over fourth) of trisomy 18 were absent in our patient. Ocular abnormalities reported in trisomy 18 are numerous[8,9]. The most commonly found are short palpebral fissures, ptosis, epicanthus, mongoloid and anti-mongoloid palpebral fissure, abnormally long or sparse eyelashes and thick lids. But AFA has been described in only seven trisomy 18 syndromes.

The etiology of AFA is unknown. A number of theories have been proposed. The currently

accepted theory is that the condition is due to an interplay of temporary epithelial arrest and rapid mesenchymal proliferation, allowing union of the lids at certain points. The association of AFA with the cleft lip and palate, popliteal pterygium, ectodermal dysplasia and vaginal erosion may be explained by a delay in temporary overlapping and apoptosis during the development of mesenchymal and ectodermal tissues[10]. This suggests a common defect in the mechanism that regulates tissue fusion at multiple sites during development. It has also been postulated that in trisomy 18, there is an abnormal cellular proliferation and hyperplasia[11]. Interestingly, since in our patient we found the clinical signs of both Hay-Wells syndrome of ectodermal dysplasia and trisomy 18 syndromes, and since most of the associated anomalies (congenital heart abnormality, cleft palate, nail hypoplasia) of AFA are found in trisomy 18, we thought that the common etiology of these anomalies could be related to the 18th chromosome.

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Anomalous left coronary artery from the main pulmonary trunk: physiologic and clinical importance of its association with patent ductus arteriosus and pulmonary hypertension

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SUMMARY: Kılıç A, Elshershari H, Özkutlu S. Anomalous left coronary artery from the main pulmonary trunk: physiologic and clinical importance of its association with patent ductus arteriosus and pulmonary hypertension. Turk J Pediatr 2002; 44: 363-365.

Origin of the left main coronary artery from the pulmonary trunk is an extremely rare, fatal, but potentially treatable anomaly. Left ventricular perfusion with desaturated blood with low pressure from the pulmonary artery results in early death. Associated pulmonary hypertension can help to prevent ischemia. We present a four-month-old boy with this anomaly associated with patent ductus arteriosus and pulmonary hypertension.

Key words: anomalous left coronary artery, pulmonary hypertension.

Origin of the left coronary artery (LCA) from the pulmonary trunk (Bland-White-Garland syndrome) is an extremely rare but important anomaly because early death is often the natural outcome. In this report, we present a four-month-old case with this entity hat had been protected from early neonatal death by associated patent ductus arteriosus (PDA) and pulmonary hypertension.

Case Report

A four-month-old boy was admitted to our hospital with the chief complaints of cyanosis upon crying, dyspnea and seizures. The neonatal period had been uneventful and these symptoms had begun after the age of three months. The history did not reveal any abnormalities of feeding, easy fatigability or excessive sweating with minor effort.

The child appeared well nourished, with length and weight measurements over 90th percentile for age. Physical examination was normal except for mild peripheral cyanosis. Complete blood count and blood gas analysis were normal. The chest X-ray showed moderate cardiomegaly with increased pulmonary vascularity. Electrocardiography showed normal sinus rhythm, marked right axis deviation, right ventricular hypertrophy, ST segment depression in precordial leads V_{4R} and V_1, and absent Q waves in left precordial leads V_4-V_6.

On echocardiography, the right heart chambers and main pulmonary artery were extremely enlarged and a second-degree tricuspid regurgitation with a peak velocity of 4.6 m/sec was observed, which led to the diagnosis of pulmonary hypertension. Fractional shortening was 0.39; all other aspects of the examination were normal. Echocardiographic evaluation of the coronary ostia was not performed, and the patient was subjected to cardiac catheterization to identify the cause of pulmonary hypertension.

On catheterisation, the pulmonary arterial pressure was 71/42, mean 56 mmHg; aortic pressure was 97/37, mean 58 mmHg; and the left atrial pressure was not elevated. The pulmonary artery O_2 saturation was 77%. Ductal patency had been demonstrated by the passage of the catheter retrogradely from the aorta. The ascending aortogram did not show the patent ductus arteriosus which might have been because of the evident pulmonary hypertension. The origin and course of the right coronary artery was as normal in the aortic root injection, but the left coronary artery was not seen. A main pulmonary artery angiogram demonstrated the anomalous left coronary artery from the

pulmonary trunk with anterograde flow into the anomalous left coronary artery from the main pulmonary artery (Fig. 1). With these findings, a diagnosis of Bland-White-Garland syndrome with patent ductus arteriosus and pulmonary hypertension was made. The patient was scheduled for dissection and reimplantation of the left main coronary artery to the appropriate sinus of Valsalva when he was lost suddenly just before surgery.

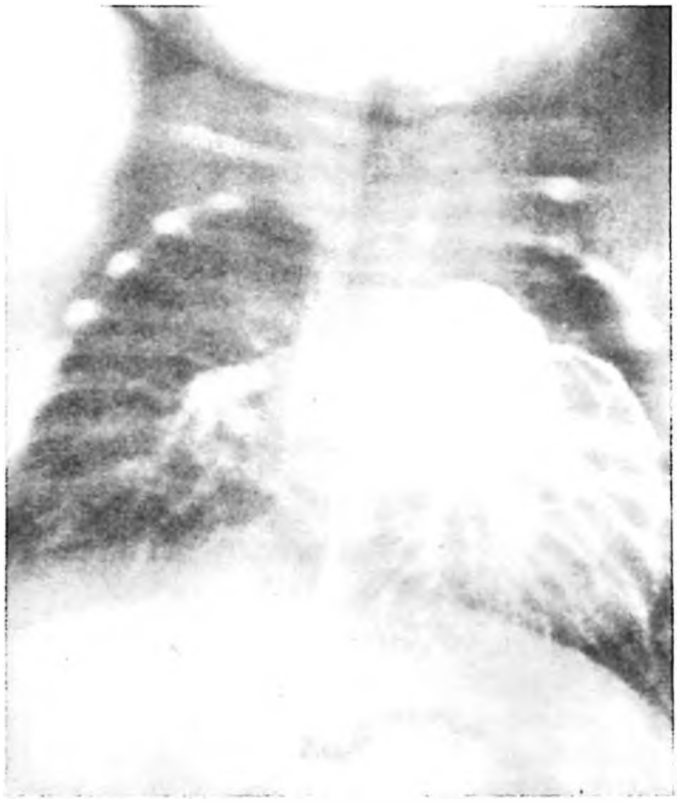

Fig. 1. Angiographic view of the left main coronary artery originating from the pulmonary trunk.

Discussion

Anomalous origin of the LCA from the pulmonary trunk is an extremely rare anomaly, having been found in only 24 out of 23,249 coronary angiograms done in older children and adults[1]. Of all children born with this anomaly, about 87% present in infancy and about 65 to 85% die before one year of age from intractable congestive heart failure, usually after two months of age[2].

This anomaly is well tolerated in fetal life, because pressures and oxygen saturations are similar in the aorta and the pulmonary artery. After birth, the pulmonary artery contains desaturated blood at a rapidly falling pressure; accordinly, the left ventricle is perfused with desaturated blood at low pressures. At first, ischemia is transient and occurs only with exertion, such as feeding or crying, but further increase in myocardial oxygen demand leads to infarction of the anterolateral left ventricular free wall. This causes congestive heart failure, which is often made worse by a dilated mitral ring or infarction of the anterior papillary muscle. Myocardial ischemia stimulates the development of collateral circulation between the two coronary artery systems. The severity of the symptoms and survival beyond this point will depend on the adequacy of this collateral circulation. In about 15% of these patients, myocardial perfusion can sustain myocardial function at rest or even during exercise and these cases can reach adult life.

This anomaly is often isolated, but has been associated with PDA, ventricular septal defect, tetralogy of Fallot, and coarctation of the aorta. The presence of an associated congenital heart disease may either mask the clinical picture of the anomalous LCA with its own symptoms or provide higher pressure perfusion to the coronary circulation via the pulmonary artery, such as in the presence of a PDA and pulmonary hypertension or pulmonary hypertension alone. A large arterial duct will maintain adequate left coronary perfusion and thus delay presentation until the duct is ligated. Indeed in some cases it could be a cause of sudden deterioration or cardiac arrest[3,4].

Pulmonary hypertension not associated with congenital heart malformations, pulmonary parenchymal disease, left atrial hypertension, hypoventilation, or other known causes of pulmonary hypertension, are encountered rarely in adults, and even less frequently in infants and children. Newborns with increased pulmonary vascular resistance and right-to-left shunting are usually considered to have persistent pulmonary hypertension of the newborn as in our case, whereas older infants and children are said to have "primary" or "unexplained" pulmonary hypertension[5].

On physical examination, there may or may not be evidence of congestive heart failure. Radiologically, there is marked cardiomegaly and pulmonary edema. Thallium-901 myocardial perfusion imaging shows reduced uptake in the anterolateral ischemic region. Because the patient usually presents with an anterolateral

infarct, there may be abnormal Q waves in leads I, aVL, and V_4-V_6. There may also be abnormal R waves or R wave progression in the left precordial leads. However, these findings are not specific, because they are encountered in cardiomyopathies or in myocardial infarcts from other causes as well. Our case showed, instead, right axis deviation, right ventricular hypertrophy, and right ventricular strain which are the electrocardiographic (ECG) findings of associated pulmonary hypertension.

Echocardiography is replacing cardiac catheterisation as the standard method of diagnosis. Abnormal attachment of the LCA can be seen, and the direction of coronary arterial blood flow toward the great artery establishes the diagnosis. Enlarged right coronary artery, size and function of the cardiac chambers, regional left ventricular wall motion abnormalities, mitral regurgitation and echogenicity of te papillary muscles and adjacent endocardium can be demonstrated as well. Cardiac catheterisation and angiography are employed only if echocardiographic results are uncertain[6]. However, we emphasized the importance of evaluating the coronary ostia in every patient on echocardiographic examination in order not to miss a potentially treatable anomaly.

This anomaly can be treated with effective surgery. The principal surgical methods are ligation of the origin of LCA to prevent pulmonary-coronary steal, ligation of the origin of LCA with reconstitution of flow with a subclavian arterial or saphenous venous graft, direct reimplantation of the origin of LCA to the aorta, or creation of an aortopulmonary window and a tunnel that directs blood from the aorta to the left coronary ostium. The late results after surgery are fairly good. The heart becomes smaller, congestive heart failure abates, left ventricular shortening fraction improves, and mitral incompetence tends to regress[7].

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