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RISK OF TUBERCULOSIS INFECTION IN EDIRNE PRIMARY SCHOOLS AND A REVIEW OF ITS EPIDEMIOLOGICAL INDICES*

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SUMMARY: Saltık A, Sungur İ, Agun K. (Department of Public Health, Trakya University Faculty of Medicine, Edirne, Turkey). Risk of tuberculosis infection in Edirne primary schools and a review of its epidemiological indices. Turk J Pediatr 33: 143-151, 1991.

The study population was defined as all first and fifth grade pupils from 17 primary schools located within Edirne Municipality. Of these 3188 pupils, aged 6 - 14 years, 569 had not received the BCG vaccine, and were tested for Tb with strengths of 1,3 or 5 TU PPD, and the reactions were evaluated. The annual risk of infection (AIR), a relatively new indice in Tb epidemiology, was determined in the different subgroups in which age, school grade and sex were taken into consideration. The results of each subgroup were compared with each other and with those reported in foreign countries. Global AIR was found to be 1.54 percent, a relatively very high value when compared with those reported in Syria and Egypt. The relative risk ratio was recorded as 22.4 percent in the Netherlands.

Another rate parameter which is almost as important as AIR is the annual variation in AIR. In Turkey this rate has been varying annually by an average of 5 % for over the past 21 years. These figures may be the result only of normal socioeconomic development rather than the effect of campaigns waged in the fight against Tb. We are convinced that under the conditions prevailing in Turkey, widespread, early vaccination with BCG is the best way to control Tb. *Key words: annual infection risk (AIR), AIR variation rate, Tb epidemiology, Tb immunization, strength of PPD.*

Tuberculosis (Tb) infection is still a major public health problem, particularly in developing countries. A report from the World Health Organization (WHO) released at the end of 1980 estimated that there were 3.5 million new cases and 1 - 2 million deaths each year from Tb in the world[1-4]. In Turkey, because of an efficient and long-term fight against the disease[5], Tb mortality has decreased from a rate that approached 300 per 100,000 in 1940 to 10 per 100,000, a rate that is almost the same as reported in Western countries. But can we really say that the fight against Tb has been won and that the infection is now under control?

Modern health services are of a very broad spectrum and it is therefore impossible to trace the source of diseases or health problems unless the occurrences of illness are taken into consideration on a community - wide basis.

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Thus epidemiological methods must be used for evaluating the past and present situation regarding the fight against Tb and control programs must be established, a task undertaken by the WHO Experts' Committee on Tuberculosis[4, 6, 7].

In Turkey, children and young adults should be screened for Tb. In the United States, however, Tb is becoming more and more a disease of the older non - white male[8-11].

In some countries, the actual mortality rate from Tb is unreliable due to a lack of well - organized recording systems. Patients who acquire chronic conditions resulting from irregular chemotherapy are included in the case rates of Tb which indicates that prevalence rates are not good indices. In addition in countries where health services and documentation are not adequate, a great many new cases of Tb are overlooked, and therefore, incidence rates are not good indices either.

There are two alternatives for determining Tb prevalence rates:

1– Examination of the sputum for the presence of tubercle bacilli.
2– Reactions to purified protein derivative (PPD).

The first method entails mass screening and laboratory facilities, while the second can be executed in a representative sample of a community in a definite region. Using the above - mentioned methods, preschool and primary school children comprise good subjects for the detection of Tb. The risk of infection is a relatively new concept. Classical indices are not very good indicators of Tb epidemiology[8]. The risk of becoming infected with tubercle bacillus in a one - year period has been determined by Styblo[4, 5, 12]. A one percent risk of infection indicates one hundred positive sputum specimens in a population of one hundred thousand. The formula below represents the annual infection risk (AIR) of Tb, which is not a function of socioeconomic conditions of related groups or community - based recording systems.

$$\text{annual infection risk (AIR)} = 1 - N^{1/y}$$

N represents the percentage of PPD (–) children without bacille Calmette-Guerin (BCG) at "y" years of age or "y" age group. For instance, at 7 years of age, let us suppose that the ratio of PPD negativity is 85 %.

$$AIR = 1 - (0.85)^{1/7} = 1 - 9.77 = 0.0229... \quad 2.29 \text{ percent.}$$

According to this logarithmic equation, it can be assumed that 2.9 percent of PPD (–) children might be infected in a one year period.

Primary school children are groups at special risk for Tb infection from many aspects. Clarifying the epidemiological characteristics of Tb in these population groups is therefore quite significant. The aim of this study is to review the indices of the epidemiology of Tb, to assess the risk of infection in five hundred and

sixty - nine children who had not been vaccinated with BCG and to formulate some resolutions. Two papers have been previously presented by the author on vaccination with BCG, and the delineation of allergy to BCG induced by different strengths of PPD[13, 14].

Material and Methods

The study population was defined as all first and fifth grade pupils from 17 primary schools located within the Edirne Municipality (n = 3188). Junior high school students were excluded from the study because the BCG vaccination programs implemented by the Ministry of Health are restricted to the first and fifth grades[15]. The Mantoux test procedure consisting of strengths of 1, 3 or 5 TU PPD with RT 23 Tween 80 supplied by the RESAMENS - Health Ministry (Refik Saydam Public Health Institute) was administered to three age-randomized sub-groups, respectively who had not received BCG vaccination (n = 569). Teams from the Dispensary for the Fight Against Tuberculosis, The State Chest Hospital and related Departments at Trakya University Faculty of Medicine carried out the tests.

Seventy-two hours later, the diameter of the induration was measured and recorded along with other necessary information like sex, school grade, and age. Pupils with a history of respiratory infection and/or fever within the past fortnight were excluded. PPD reactions and the effectiveness of vaccination with BCG were evaluated in a total 3188 children, 1725 (54.1 %) first graders and 1463 (45.9 %) fifth graders whose ages ranged between 6 - 14 years. In another paper, the author reported on the efficacy of BCG vaccination and its interaction with allergy to tuberculin in 2619 children of the same study population who had been vaccinated at least once with BCG (82.2 %)[13].

The aim of this presentation is basically to assess the risk of infection in the five hundred and sixty - nine children who had not received BCG vaccine. AIR was calculated separately by computer for these subjects according to age and sex as previously illustrated.

Results

The composition of the study group is presented in TABLE I.

TABLE I: The Distribution According To Age and School - Grade of 569 Children Who Had Not Received BCG Vaccine

Grades	Girls No. (%)	Boys No. (%)	Total No. (%)
Grade I	222 (49.2%)	229 (50.8%)	451 (100.0%)
Grade V	62 (52.5%)	56 (47.5%)	118 (100.0%)
Total	284 (49.9%)	285 (50.1%)	569 (100.0%)

The number of fifth grade pupils in the sample is decreased because of the fact that by the time school - children reach the fifth grade, many have already been vaccinated with BCG. Table II shows, according to sex and school grade, the status of immunization of those subjects vaccinated at least once with BCG. Although there were no significant differences between vaccination ratios of both sexes in the first and fifth grades, there was a significant increase in both sexes of first and fifth graders vaccinated at least once.

TABLE II: Sex and Grade Distribution (%) of 3188 Pupils With Regard To BCG Vaccination Status

	At Least One BCG Scar			
Sex	First Grade	Fifth Grade	Total	Without BCG
Girls	73.8 (%)	91.7 (%)	81.1 (%)	18.9 (%)
Boys	73.9 (%)	92.3 (%)	83.7 (%)	16.3 (%)
Total	**73.9 (%)**	**91.9 (%)**	**82.2**	**17.8**
			(n = 2619)	**(n = 569)**

In the study subgroups, the average ages for assessing AIRs are represented in TABLE III.

TABLE III: Mean Ages ± SD in Study Subgroups

PPD (TU) Test applied	First Grade		Fifth Grade		Total	
	no.	m.age ± SD	no.	m.age ± SD	no.	m.age ± SD
1	82	7.1 + 1.1	20	11.9 ± 1.6	102	8.0 ± 1.3
3	121	7.2 + 0.9	34	11.3 ± 1.1	155	8.1 ± 1.0
5	248	7.2 + 1.2	64	11.7 ± 1.4	312	8.1 ± 1.3
Total	**451**	**7.2 ± 1.1**	**118**	**11.6 ± 1.3**	**569**	**8.1 ± 1.2**

For example when calculating AIR for a given 6 - 10 age group, the age of a related group can be expressed as (6 + 10) / 2 = 8. But, since junior high school students were excluded and also because of the gap between the number of pupils from the first to the fifth grades, (451 and 118, respectively), mean ages were calculated separately for each subgroup. Girl and boy ratios were very similar in both grades, thus no importance was given to sex in the calculations.

TABLE IV, shows the percentages of PPD (+) children without BCG vaccination which were used for calculating AIRs in the study subgroups. As is seen,

increased doses of PPD result in a greater possibility of the occurrence of natural infection allergy (Fig. 1).

TABLE IV: Percentages of PPD Positivity in 569 Pupils Without BCG Who Were Administered 1, 3 or 5 TU PPD

PPD Test Applied	First Grade			Fifth Grade			Grand Total		
	Girls	Boys	Total	Girls	Boys	Total	Girls	Boys	Total
1 TU	9.09	13.16	10.98	0.00	25.00	10.00	7.14	15.22	10.78
3 TU	12.96	7.46	9.92	6.25	22.20	14.71	11.43	10.59	10.97
5 TU	15.32	8.06	11.69	20.59	20.00	16.95	16.46	10.39	13.46
Total*	**13.51**	**8.73**	**11.09**	**14.55**	**21.43**	**15.13**	**13.38**	**11.23**	**12.30**

* Total percentages are determined according to the number of cases in each TU sub-group. For example for 1, 3 and 5 TU sub-groups, respectively [(102x10.78)+(155x10.97)+(312x13.46)]/569=12.30.

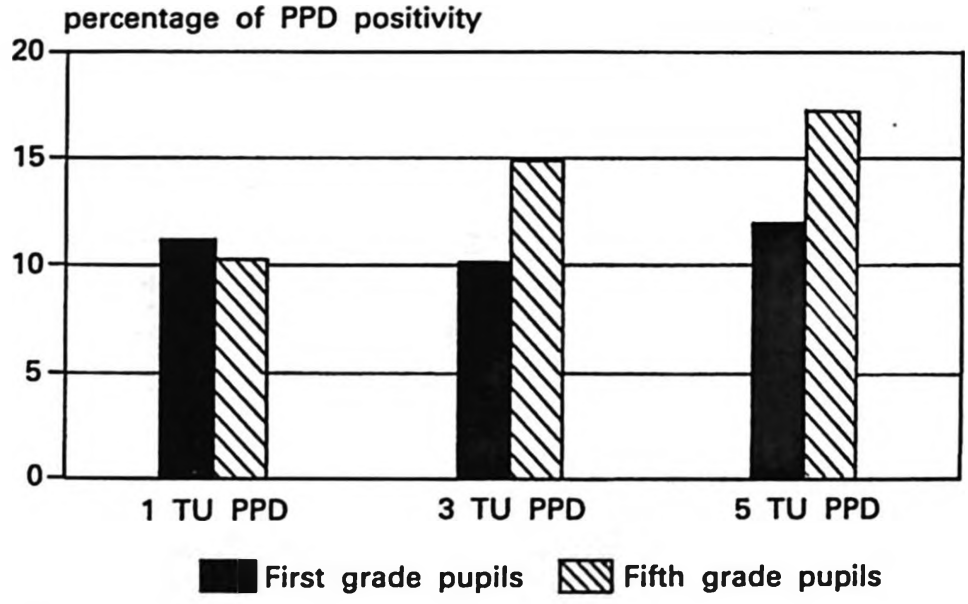

Fig. 1: Percentages of PPD positivity in 569 pupils who had not received BCG vaccine and were given 1, 3, and 5 TU PPD.

According to these data, PPD reactions are not dependent on the school grade, in other words, age and sex of pupils, with the exception of subjects given dosages of 5 TU. Table V shows AIR values for both first and fifth graders who received three different PPD dosages in order to determine the changes in AIR values according to dose (Fig. 2).

TABLE V: The Risk of Tb Infection in 569 Children Who Had Not Received BCG Vaccine in Relation to TU Subgroups

Grades	Tb Risk of Infection (%) According to Applied PPD Dose			In General
	1 TU	3 TU	5 TU	
First Grade	1.62	1.44	1.71	1.62
Fifth Grade	0.88	1.40	1.57	1.40
Total	**1.62**	**1.42**	**1.67**	**1.61**

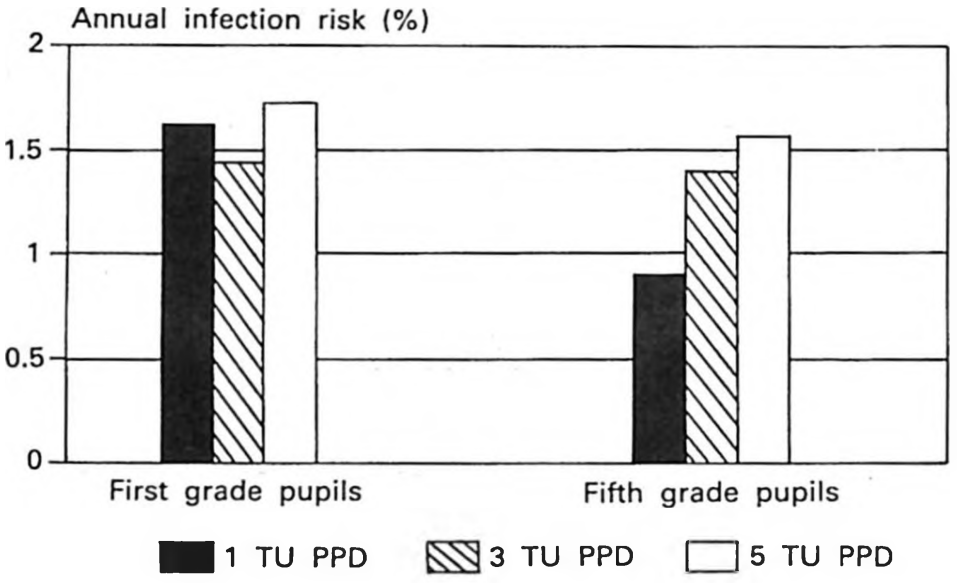

Fig. 2: Risk of Tb infection in 569 pupils who had not received BCG vaccine in relation to TU subgroups (%).

As seen from TABLE V, the AIR value increases according to PPD dosage and the possibility of delineating natural infection allergy rises[12, 14, 16, 17]. AIR is about 1.62 percent in the first grade and 1.40 percent in the fifth grade, which is to be expected because an inverse relationship exists between age and AIR. Additionally, the ratio of PPD (−) pupils without BCG vaccination gradually decreases with time because of natural infection allergy.

The AIR value of 1.61 percent listed in TABLE V for the entire group may be calculated more precisely. Considering the relative magnitude of the first and fifth grades in the total 569 pupils, real or effective AIR can be calculated more precisely as:

$$(1.62 \times 0.79) + (1.40 \times 0.21) = 1.57$$

One can easily assume by this AIR value that there may be 1.57 x 50 = 79 (per 100 thousand) smear (+) patients within a period of a year[4, 5, 12].

In TABLE VI, the AIR values calculated above were compared to the AIR values in other countries using relative risk coefficients.

TABLE VI: AIR Rates in Various Countries and Relative Risk Coefficients

Country	1950 - 53	1982 - 84	Relative Risk
Syria	3.0 %	0.25 %	6.28
Egypt	2.8 %	0.40 %	3.90
The Netherlands	–	0.07 %	22.40
Turkey:			
– (0 - 3) age, (1964)	1.8 %	0.65 %	–
– (0 - 6) age, (1964)	2.0 %	0.63 %	–
Presented study	–	1.57 %	1.0

Discussion

The data obtained are more than striking. In WHO projects, it has been reported that AIR which was three percent in 1950 fell to 0.25 percent in 1983. In the Netherlands it was 10 percent in 1910 and fell to 0.07 percent by the end of 1984[4]. In comparison, the relative risk ratios have been reported as 6.3 in Syria, 3.9 in Egypt, and 22.4 in the Netherlands (Table VI; Fig. 3).

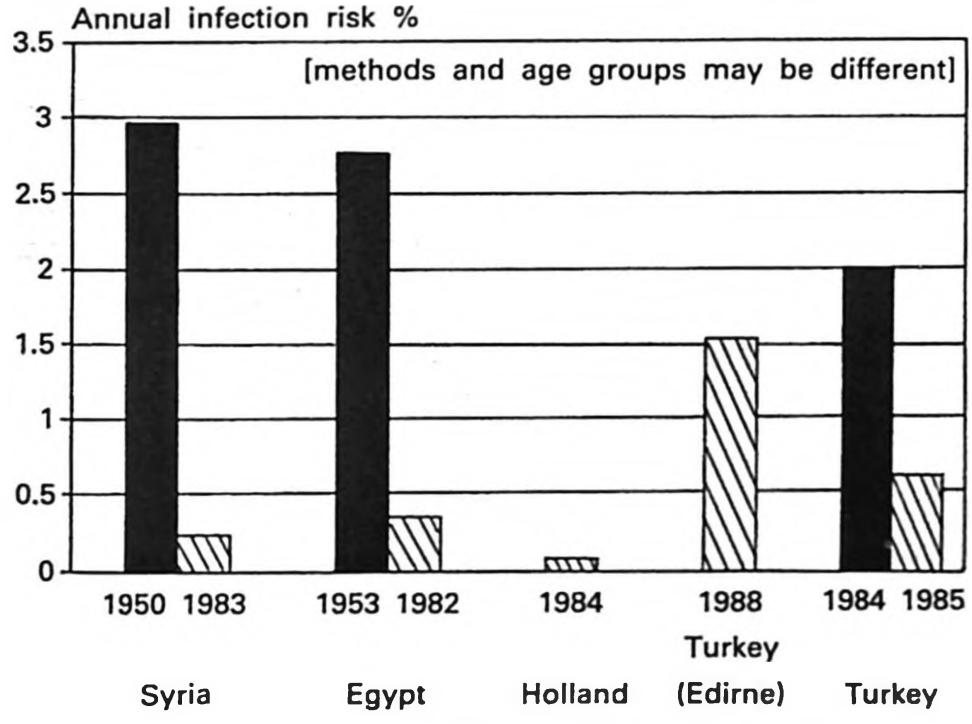

Fig. 3: Annual risk of Tb infection in various countries and in Edirne.

The official statistics on the number of Tb cases in Turkey is much lower than expected, which means that screening programs are limited. Thus, we cannot realistically abandon BCG vaccination programs and substitute them with active patient surveillance, as is implemented in Western countries. In Western countries, the infection reservoir was reduced to the present level only after BCG vaccination programs and other social and economic measures had been strictly implemented[18].

What is even more important than AIR itself, is the fact that the rate of AIR varies annually. An average annual (geometric) drop of 5.3 percent in AIR values has been reported in Turkey over the past twenty - one years[12].

Even though no great strides have been made in combatting Tb in Turkey, some improvement has been observed as a result of socioeconomic development[12]. Rates have been reported to be decreasing at an average of 7 percent in Syria, 13 percent in the Netherlands (1940 - 1984), and from 10 percent (1910 - 1940) to 6 percent (1950 - 1983) in Egypt[4]. In the United States a decreasing average annual rate of 8.5 percent has been noted, and it is envisaged that the rate of incidence might drop to 1 per million towards the year 2010[10]. These rates, which are dropping more than 5 percent annually, indicate the effectiveness of Tb control programs.

When compared to the average in Turkey, Edirne has better socioeconomic parameters. In spite of these relatively good living conditions, even if only present in the urban center, a remarkably high AIR has been reported in this province. Can it be said, therefore, that the average annual decrease of 5 percent in AIR values over the past twenty - one years did not result from campaigns waged to fight against Tb but rather from socioeconomic development? If this assumption is incorrect, then socioeconomic development in Turkey is at a standstill. What is more probable is that both factors equally influenced the drop in AIR.

Early, widespread and efficient vaccination programs should be given precedence, which should be followed by prompt implementation of other recommendations by WHO and adapted to Turkey's needs and conditions.

In conclusion, it is impossible to ignore Winslow's adage: "Tb can be defeated only in countries where poverty has been eliminated and there is adequate nutrition".

We plan further studies to monitor AIR trends in Edirne through longitudinal epidemiological surveys.

REFERENCES

1. *WHO Tech Rep Ser.* No 652, 1980, Geneva, Switzerland.

1. *WHO Tech Rep Ser.* No 671, 1982, Geneva, Switzerland.

3. Killpatrick GS. Global tuberculosis. In Hobson W (ed). *Theory and Practice of Public Health,* (4th ed). New York: Oxford, U Pr, 1975, pp. 341-354.
4. Notes from Dr. Bleiker's speech. *Infection Risk in Some of the Middle-East Countries and Some Indicators of Tuberculosis,* "Middle-East Region Meeting on the Fight Against Tuberculosis". October 1985, Istanbul.
5. Özcan C. Türkiye'de Tüberkülozun Bugünkü Durumu. *Tüberküloz.* Türkiye Sağlık ve Tedavi Vakfı konferans kitapçığı, December 9, Ankara, s. 3-10.
6. Barker DJ. *Practical Epidemiology* (3rd ed). Edinburgh: Churchill Livingstone, 1984.
7. Vaughan JP, Morrow RH (eds). *Manual of Epidemiology for District Health Management.* Geneva: WHO, 1989.
8. Styblo K, Meijer J, Sutherland I. The transmission of tubercle bacilli: its trend in a human population. *Bull Int Union Tuberc* 42:1, 1969.
9. Styblo K. Tuberculosis control and surveillance. In Fenley DC, Petty TL (eds). *Recent Advances in Respiratory Medicine.* London: Churchill Livingstone, 1986, pp. 77-108.
10. Rieder HL, Cauthen GM, Comstock GW, Snider DE Jr. Epidemiology of tuberculosis in the United States. *Epidemiol Rev* 11:79, 1989.
11. Dowdle WR. A strategic plan for the elimination of tuberculosis in the United States. MMWR 38 (Suppl 3): 1, 1989.
12. Koçoğlu F. *Verem Savaşı.* Ankara: Hacettepe Üniversitesi Tıp Fakültesi, Halk Sağlığı Anabilim Dalı Yayınları, 1986.
13. Saltık A, Sungur İ, Agun K. Edirne merkez ilkokullarında BCG'li 2619 öğrencide tüberkülin allerjisi ve aşı etkinliği. Presented at *Social Pediatrics Day,* 21 February, 1990, İzmir.
14. Agun K, Saltık A, Sungur İ, et al. Edirne merkez ilkokullarında 1, 3 ve 5 TU PPD ile BCG'siz çocuklarda tüberküloz allerjisi araştırması. *Türkiye Solunum Araştırmaları Derneği XVII. Ulusal Kongresi*'nde sunulmuştur. 25-29 Eylül 1989, İzmir.
15. Sağlık Bakanlığı Temel Sağlık Hizm. Gn. Md. *Aşı Uygulama Rehberi,* Ankara, 1987.
16. Furcolow ML. Quantitative studies of tuberculin reaction of tuberculin sensitivity and its relationship to tuberculosis infection. *Public Health Rep,* (Washington) 56:1082, 1941.
17. Murtagh K. Unreliability of the Mantoux test using 1 TU PPD in excluding childhood tuberculosis in Papua New Guinea. *Arch Dis Child.* 55:795, 1980.
18. Horwitz D, Palmer CE. Epidemiological basis of tuberculosis eradication. 2. Dynamics of tuberculosis morbidity and mortality. *Bull WHO* 30:609, 1964.

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ROTAVIRUS DIARRHEA IN NEWBORN INFANTS*

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SUMMARY: Akıncı A, Teziç T, Gür İ, Çetin H, Hatun Ş. (Department of Pediatrics, Dr. Sami Ulus Children's Hospital, Ankara, Turkey). Rotavirus diarrhea in newborn infants. Turk J Pediatr 33: 153-157, 1991.

In this study, the frequency of rotavirus infection and also the relation of rotavirus pathogens to necrotizing enterocolitis were investigated in newborns with diarrhea. We observed that rotavirus is a very important agent as a cause of nosocomial infection and also that it has a role in the development of NEC. *Key words: rotavirus, diarrhea, necrotizing enterocolitis.*

Diarrhea is still one of the leading causes of infant mortality[1,2]. In developing countries, enterotoxigenic *E. coli* and rotavirus infections are the most prevalent pathogens known to cause diarrhea[1,3]. This study describes the clinical and laboratory findings and prognosis of diarrhea due to rotavirus infection in newborn infants. We also compared our findings with those reported in the literature.

Material and Methods

This study was performed on 70 newborn infants hospitalized for several reasons in the Neonatal Unit of Dr. Sami Ulus Children's Hospital between January - March 1990. Table I lists the reasons for admission to the neonatal unit. Patients passing normal stools at admission and then passing loose, watery and frequent stools 3 - 6 days after admission were considered to have diarrhea, and were categorized into three groups according to symptoms.

1– Mild diarrhea: Newborn infants with diarrhea but without complications or signs of dehydration.

2– Moderate diarrhea: Newborn infants with diarrhea accompanied by fever, vomiting and dehydration.

3– Severe diarrhea: Newborn infants with diarrhea accompanied by dehydration and such symptoms and signs as: lethargy, hypotonia, apneic spells, blood in the stool, X - ray findings of pneumatosis intestinalis, abdominal distension, prominence of bowel movements and indication of perforation.

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Stool, blood and urine cultures were taken from all patients. Hemograms and serum electrolytes, and BUN and creatinine levels were determined. The ELISA (Rotazyme) test was used to determine rotavirus in the stool samples. Stool samples of the personnel working in the newborn unit, water samples, and the oral rehydration solution (ORS) used in the unit were analyzed. The patients were hospitalized between 2 - 19 days. There was no significant difference found between the hospitalization period of rota (+) and rota (−) patients ($p > 0.05$) (Table II). Clinical data of the patients are given in Table II.

Results

Of the 17 rota (+) patients, three had mild, six had moderate and eight had severe diarrhea. Of the 53 rota (−) patients, 29 were described as being mildly ill, 10 moderately and 14 severly ill. The patients' laboratory findings are shown in Table III. The stool cultures of each of the two rota (+) infants with severe diarrhea, yielded *Shigella, E. coli* and *Klebsiella,* while the stool cultures of the 14 rota (−) infants with severe diarrhea yielded *Klebsiella* in two and both *Shigella* and *Klebsiella* in one. Pathogens were not detected in any of the remaining rota (−) infants with severe diarrhea.

The blood cultures obtained from the rota (−) infants grew the following: Staphylococcus (one patient), *E. coli* (two patients) and *Klebsiella* (two patients). No pathogens were isolated from the blood cultures of the rota (+) patients. Microscopic analysis of stools demonstrated leukocytes in six rota (+) patients and eleven rota (−) patients. Other laboratory findings are given in Table III.

Discussion

Rotavirus is one of the leading causes of diarrhea in newborn infants[1-5]. There is a strong correlation between rotavirus infection and symptoms such as fever, vomiting and diarrhea in the first six months of life. Asymptomatic rotavirus infections have been also reported in newborns. Besides the asymptomatic cases, there are also severe cases such as those with manifestations as necrotizing enterocolitis (NEC)[4, 6, 7]. In our 17 rota (+) patients, three had mild, six had moderate and eight had severe diarrhea. In four of the eight severe cases, NEC developed with pneumatosis intestinalis which was demonstrated on the X - ray studies. In one of these cases, there was a history of perinatal asphyxia and exchange transfusion, both of which are considered risk factors for the development of NEC. Three rota (+) patients with a history of prior exchange transfusion did not develop NEC (Table II). However, the other three patients who developed NEC were not at risk for this disease. *E. coli* and *Klebsiella* were isolated in the stool cultures of one of these four patients, while in the remaining patients no bacterial pathogen was cultured. Previous studies have shown that

TABLE I: The Cause of Admission to the Neonatal Unit

Admission Causes	Number
Hyperbilirubinemia	58
Respiratory distress	2
Convulsion	1
Diarrhea	2
Sepsis	1
Anoxia	1
Epidermolysis bullosa	1
Neonatal tetanus	1
Lower respiratory tract infection	1
Meconium aspiration	2
Total	70

TABLE II. The Clinical Data of the Patients

	Rotavirus (+) (n:17)			Rotavirus (−) (n:53)		
	Mild (n:3)	Moderate (n:6)	Severe (n:8)	Mild (n:29)	Moderate (n:10)	Severe (n:14)
Prematurity	–	–	–	1	3	3
Median birth weight (gr):						
1500-1999	–	–	–	1	3	3
2000-2499	–	–	–	8	4	3
2500-2999	1	2	3	12	1	6
3000-3499	1	3	5	5	–	2
3500-	1	1	–	3	2	–
Median duration of hospitalization (days)	2	5	19	3	5	17
Median length of diarrhea (days)	2	5	14	3	5	12
Breast-feeding	3	3	8	12	8	11
Median duration of breast-feeding (days)	2	3	3	4	3	3
Colostrum-feeding	3	3	8	12	8	11

the ELISA method can yield false (+) results in positive stool cultures, which has been explained by non - specific binding of rotavirus - antibody to bacteria[8]. In our study, the detection of rotavirus in two patients with positive stool cultures, could have possibly been due to the sensitivity of the ELISA method. In the severe rota (+) cases, leukocytosis and toxic granulation of polymorphonuclear leukocytes were strikingly higher than the rota (−) patients ($p < 0.05$; Table III). The leukocytes in the stool smear of rota (+) patients were also higher than the rota (−) patients. This can be explained by the enteroinvasive character of rotavirus[9, 10]. In the patients with moderate and severe diarrhea, BUN and creatinine levels were higher than in the patients with mild diarrhea (Table III).

TABLE III: Laboratory Findings of the Patients

	Rotavirus (+) (n:17)			Rotavirus (−) (n:53)		
	Mild (n:3)	Moderate (n:6)	Severe (n:8)	Mild (n:29)	Moderate (n:10)	Severe (n:14)
Hb (gr/dl)	10.9±1.8	13.4±1.1	14.6±2.1	12.6±1.1	16.6±2.1	15.0±2.3
Hct	35.6±5.4	37.2±8.2	37.8±4.4	34.4±4.4	38.2±6.1	37.9±3.8
Leukocyte (mm3)	10.000±1.200	18.000±6.100	32.000±10.800 ($p < 0.05$)	9.500±3.900	21.500±5.750	19.000±1.500 ($p < 0.05$)
Toxic granulation	–	1	6 ($p < 0.05$)	–	2	3
Na (mEg/L)	142±10	138±8	135±7.5	138±11	140±10.8	138±11
Urea nitrogen (mg/dl)	18.6±7.6	20.5±6.5	23.8±8.5	10.4±4.7	13.2±7.9	28.6±10.1
Creatinine (mg/dl)	1.01±0.3	1.2±0.8	2.2±0.7	1.9±0.1	2.4±0.18	2.3±0.8
PNL (%) in faces (percentage per high-power field)	70±17	70±15	80±22	55±12	60±20	65±15

• Values are given as mean ±50.

Dehydration was regarded as having caused the elevation in BUN and creatinine levels. It is a well known phenomenon that breast milk protects the infant from rotavirus[11]. Fourteen of our rota (+) patients were breast - fed approximately three days before they were admitted to the hospital. This paradox can be explained by the short duration of breast - feeding which is insufficient for the development of protective antibody against rotavirus. Our patients support recent reports about the severity of rotavirus infections during the newborn period. As a result of this study, we can say that the clinical course of rotavirus infections ranges from mild to severe gastroenteritis and also that it is the leading cause of NEC during the newborn period and infancy.

REFERENCES

1. Gurwith M, Wenman W, Hinde D, et al. A prospective study of rotavirus infection in infants and young children. *J Infect Dis* 144:218, 1981.
2. Ho MS, Glass RI, Pinsky PF, Anderson LJ. Rotavirus as a cause of diarrheal morbidity and mortality in the United States. *J Infect Dis* 158:1112, 1988.
3. DuPont HL. Rotaviral gastroenteritis-some recent developments. *J Infect Dis* 149:663, 1984.
4. Champsaur H, Questiaux E, Prevot J, et al. Rotavirus carriage, asymptomatic infection, and disease in the first two years of life. I. Virus shedding. *J Infect Dis* 149:667, 1984.
5. Truant AL, Chonmaitree T. Incidence of rotavirus infection in different age groups of pediatric patients with gastroenteritis. *J Clin Microbiol* 16:568, 1982.
6. Champsaur H, Henry-Amar M, Goldszmidt D, et al. Rotavirus carriage, asymptomatic infection, and disease in the first two years of life. II. Serological response. *J Infect Dis* 149:675, 1984.
7. Rotbart HA, Nelson WL, Glode MP, et al. Neonatal rotavirus-associated necrotizing enterocolitis: case control study and prospective surveillance during an outbreak. *J Pediatr* 112:87, 1988.
8. Ceyhan M, Yeniay I, Kanra G, Ciliv G. Rotavirus viral RNA electrophoresis in infants with diarrhea and the importance of rotavirus in the etiology of gastroenteritis in infants in Ankara, Turkey. *DOGA TU Tip ve Ecz D* 10:246, 1986.
9. Kovacs A, Chan L, Hotrakitya C, et al. Rotavirus gastroenteritis. Clinical and laboratory features and use of the Rotazyme test. *Am J Dis Child* 141:161, 1987.
10. Dearlove J, Latham P, Dearlove B, et al. Clinical range of neonatal rotavirus gastroenteritis. *Br Med J [Clin Res]* 286:1473, 1983.
11. Rahman MM, Yamauchi M, Hanada N, et al. Local production of rotavirus specific IgA in breast tissue and transfer to neonates. *Arch Dis Child* 62:401, 1987.

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THE EFFECT OF EPIDERMAL GROWTH FACTOR ON SERUM ZINC LEVELS*

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SUMMARY: Demirsoy S, Erbaş D, Hasanoğlu E. (Departments of Pediatrics and Physiology, Gazi University Faculty of Medicine, Ankara, Turkey). The effect of epidermal growth factor on serum zinc levels. Turk J Pediatr. 33: 159-161, 1991.

Epidermal growth factor is a potent stimulator of the growth of epithelial and mesenchymal cells. In this study the effect of epidermal growth factor on the cells of developing mice intestine and on the serum zinc concentrations were assessed. Higher serum zinc concentrations (840.21 ± 187.82 mg/dl) were found in the mice given epidermal growth factor (n: 10) as compared to the values obtained in the controls (347.55 ± 108.88 mg/dl), (n: 12) ($p < 0.05$). *Key words: epidermal growth factor, serum zinc level.*

Epidermal growth factor (EGF), a small polypeptide hormone first isolated from mouse submandibular gland, is one of the most biologically potent and best characterized growth factors[1, 2]. It plays an important role in the growth of a wide variety of cultured cells, both epithelial and mesenchymal[3]. It affects cells by cell receptors which exhibit tyrosine kinase activity[4]. Thus EGF is an extremely essential polypeptide for growth and development[5].

Our study was designed to demonstrate the effect of EGF on the gastrointestinal tract and the serum zinc level of mice.

Material and Methods

This study was carried out on a total twenty-two newborn mice. Ten of the mice were injected with 10 mg/kg/day EGF subdermally for a period of ten days. The twelve controls were injected with 10 mg/kg/day physiologic serum subdermally also for a period of ten days following birth.

At the end of the tenth day, the mice were sacrificed by decapitation and blood samples were collected. Serum zinc levels were determined by using atomic absorption spectrophotometry. Statistical analysis was carried out by the Student's *t* test.

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Results

The body weights of the subjects and the controls at birth and on the tenth day, and the serum zinc levels of the mice who had been given EGF and the controls are shown in Tables I and II. The serum zinc levels of the mice given EGF were found to be significantly higher than those of the controls ($p < 0.05$).

TABLE I: Body Weight (g) and Serum Zinc Levels (mg/dl) of Mice Given EGF and Control Group

	Mice Given EGF			Control Group		
No.	Birth Weight	Body Weight 10th Day	Zinc Level	Birth Weight	Body weight 10th Day	Zinc Level
1	1.59	5.83	845.30	1.49	4.70	213.56
2	1.73	4.73	544.80	1.59	4.88	289.15
3	1.48	6.05	1011.30	1.77	4.99	595.79
4	1.35	5.50	547.39	1.69	5.56	459.36
5	1.69	5.35	1100.83	1.43	6.25	208.45
6	1.78	5.28	633.98	1.34	6.12	345.48
7	1.55	5.79	759.54	1.66	6.36	416.17
8	1.94	5.13	990.21	1.58	6.48	318.42
9	1.85	4.89	845.60	1.94	5.56	256.55
10	1.78	4.70	656.65	1.87	5.43	318.42
11				1.46	4.88	347.45
12				1.39	4.74	386.85

TABLE II: Comparison of the Mean Serum Zinc Levels of Mice Given EGF and Controls (mg/dl)

	Serum Zinc Level
Mice given EGF n: 10	840.21 ± 187.82
Control n:12	347.55 ± 108.88 $p < 0.05$

Discussion

Zinc is a constituent of several vitally important enzymes, and plays an important role in nucleic acid synthesis. It prevents the lipid depolarization of the cell membrane[7]. It has been reported that submandibular gland extract increases serum zinc levels and accelerates wound healing in adult mice[8].

The postnatal development of the small intestine of laboratory animals offers an interesting model to examine the possible role of EGF in the proliferation and differentiation of absorptive cells. EGF influences the maturation of the digestive function of absorptive cells by inducing a precocious appearance, sucrase activity and a premature increase of several brush border activities[9]. Intestinal mucosa has EGF receptors during the suckling period in neonate mice. EGF promotes the maturation of duodenal epithelial cells in cell culture[9]. According to our results, serum zinc levels of the mice given EGF were higher than the controls, which suggests that EGF may increase zinc absorption with increasing maturation and enzyme activity of the small intestine.

REFERENCES

1. Cohen S. Isolation of a mouse submaxillary gland protein accelerating incisor eruption and eyelid opening in the newborn animal. *J Biol Chem* 237:1555, 1962.
2. Cohen S. Epidermal growth factor. *J Invest Dermatol* 59:13, 1972.
3. Carpenter G, Cohen S. Epidermal growth factor. *Ann Rev Biochem* 48:193, 1979.
4. Gonzales CR, Konemitsu M, Boynton LA. Epidermal growth factor induces the accumulation of calpactin II on the cell surface during membrane ruffling. *Cell Motil Cytoskeleton* 15:34, 1990.
5. Carpenter G. Epidermal growth factor. *Handbook Experimental Pharmacology* 57:89, 1981.
6. Buckley A, Davidson JM, Kamerath CD, et al. Sustained release of epidermal growth factor accelerates wound repair. *Proc Natl Acad Sci USA* 82:40, 1985.
7. Tuncer M, Demirsoy S, Özsoylu Ş, Erdem G. The significance of zinc, copper and magnesium levels of maternal, cord and newborns' sera in hyperbilirubinemia of unknown etiology. *Turk J Pediatr* 24:227, 1982.
8. Erbaş D, Güvendik G, Gönül B. Submandibular bez ekstrelerinin serum çinko düzeylerine etkileri. *Ankara Pharmacology Faculty Journal* 17:11, 1987.
9. Malo C, Ménard D. Influence of epidermal growth factor on the development of suckling mouse intestinal mucosa. *Gastroenterology* 83:28, 1982.

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IgG SUBCLASS DEFICIENCY IN CHILDREN WITH RECURRENT INFECTIONS*

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SUMMARY: Tezcan İ, Ersoy F, Sanal Ö, Göçmen A, Yeniay İ. (Immunology Unit, Hacettepe University Institute of Child Health, Ankara, Turkey). IgG subclass deficiency in children with recurrent infections. Turk J Pediatr 33: 163-166, 1991.

IgG subclass levels were studied in 12 children, aged between 2.5 - 12 years with recurrent respiratory tract infections. They did not have low IgG levels or IgA deficiency. We found combined deficiency of IgG2 - IgG4 in one patient and selective IgG2 deficiency in another (16.6 % of patients). These two patients had bronchiectasis due to recurrent severe pneumonia, however one patient with bronchiectasis had normal IgG subclass concentrations. Our IgG subclass - deficient patients who did not respond to prophylactic antibiotic therapy were given gammaglobulin therapy. IgG subclass deficiency should be considered in children with unexplained recurrent infections even in the presence of normal serum immunoglobulin levels. *Key words: IgG subclass deficiency, recurrent infections, gammaglobulin.*

IgG subclass deficiency has been described in patients with selective IgA deficiency, ataxia telangiectasia, common variable hypogammaglobulinemia, allergic diseases, autoimmune disorders and in individuals with recurrent infections[1-8]. Although the actual incidence of IgG subclass deficiency is unknown, it is probably one of the most common humoral immunodeficiency disorders.

We studied IgG subclasses in 12 children with recurrent respiratory infections who did not have IgA deficiency and whose total IgG levels were not less than two SD below the age-matched control values.

Material and Methods

We examined 12 children, ages 2.5 - 12 years (mean: 5.5 years), who were referred to the Immunology Unit of Hacettepe University Children's Hospital for

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evaluation of recurrent infections. They did not have low total IgG levels or IgA deficiency.

Eleven patients had a history of recurrent episodes of pneumonia, three of whom had bronchiectasis and one recurrent otitis media with purulent drainage.

Serum IgG, IgA and IgM values were measured using the radial immunodiffusion method (Behring - Werke Inst.). IgG subclasses were determined by the ELISA technique (Serotec, England).

The IgG subclass values obtained were compared with the normal ranges reported by Oxelius[9]. A subclass deficiency was established in a patient if the IgG subclass value was two standard deviations below the mean for the age - matched control.

Results

Increased IgG1 levels were found in seven patients including two with IgG2 deficiency (Table 1). Two patients (NB, OS) who had recurrent pneumonia and

TABLE I: Immunoglobulin Levels in Children with Recurrent Infections

Patient	Age (yrs)	Diagnosis	Serum Immunoglobulin Levels (g / l)					
			IgG1	IgG2	IgG3	IgG4	IgM	IgA
1.DD	3	R.P.	7.6	1.45	0.55	0.52	2.08[a]	0.77
2.BB	2.5	R.P.	9.42[a]	0.42	0.17	0.18	1.23	0.70
3.SK	7	R.P.	15.2[a]	1.15	0.30	0.64	1.23	0.77
4.CH	4.5	R.O.M.	7.3	1	0.35	0.08	1.38	0.42
5.SO	4	R.P.	12.4[a]	0.8	0.27	0.13	1.45	0.42
6.SY	4	R.P.	14.8[a]	0.42	0.27	0.07	0.59	0.42
7.NI	3	R.P.	7.9	0.8	1.05[a]	0.04	1.76[a]	1.35[a]
8.MD	7	R.P.	9.1[a]	0.95	1.15	0.08	2.80	1.09
9.NB	12	R.P.+B	16.3[a]	< 0.01[b]	0.85	UD[b]	3.70[a]	0.62
10.BA	5	R.P.	7.6	1.35	1.38[a]	0.17	2.55[a]	0.52
11.OS	9	R.P.+B	8.7[a]	< 0.42[b]	0.65	0.06	9.43[a]	0.66
12 AY	7	R.P.+B	5.9	1.85	2.10[a]	0.06	0.86	0.90

Abbreviations used in Table:

R.P : Recurrent pneumonia.
ROM : Recurrent otitis media.
B : Bronchiectasia.
UD : Undetectable.

a : Two standard deviations above the mean values of the age - matched control.
b : Two standard deviations below the mean value of the age - matched control.

bronchiectasis had very low IgG2 concentrations. Ten patients had normal IgG2 levels and one had bronchiectasis.

Increased IgG3 was found in three patients. IgG4 was undetectable in one patient (NB) with IgG2 deficiency. Thus, of 12 children with recurrent respiratory tract infections, one had combined deficiency of IgG2 - IgG4 and one had selective IgG2 deficiency.

Serum IgM levels were elevated in five patients including two IgG2 - deficient patients.

Discussion

In two of 12 children with a history of recurrent respiratory tract infections, we discovered combined deficiency of IgG2 - IgG4 in one, and selective IgG2 deficiency in the other.

The association of IgG subclass deficiency with infections (particularly sinopulmonary infections) was first reported by Schur et al[5]. IgG2 - deficient patients with recurrent infections tend to show a defective humoral immune response to bacterial polysaccharide antigens[10]. It appears that patients with low IgG2 levels are susceptible to infections caused by encapsulated bacteria (e.g. pneumococci, *H. influenzae*). Since serum IgG4 levels vary widely in the normal population, selective IgG4 deficiency is difficult to interpret.

The real incidence of IgG subclass deficiency is unknown. Shackelford et al[11] reported IgG2 deficiency in seven of 30 children who had recurrent respiratory tract infections, some of whom also had IgA deficiency. Stanley et al[12] studied 100 adults with recurrent or chronic respiratory tract infections and found that seven of them were IgG2 - deficient. In our study, IgG2 deficiency was found in 16.6 percent of patients with recurrent infections and normal IgA levels, which is a higher rate than Stanley's. Aucouturier et al[13] reported that IgG subclass deficiency was detected in one fourth of children and adults studied with abnormally frequent infections, and that isolated IgG2 deficiency and combined deficiency of IgG2 - IgG4 were the most common, which is consistent with our findings.

Shackelford et al[11] emphasized that their IgG2 - deficient patients had more severe infections than the patients prone to infections who had normal immunoglobulin levels. Our two IgG2 - deficient patients had chronic lung disease due to recurrent severe infections. However, in one patient with bronchiectasis and severe recurrent pneumonia, we found the IgG subclass levels to be within the normal range. Patients with normal IgG subclasses and severe recurrent infections may have an inability to produce specific antibodies to several antigens (e.g. IgG2 antibody to polysaccharide antigens) in spite of normal IgG subclass levels[14].

Elevated IgG subclasses and IgM levels were found in some of our patients which might have resulted from chronic antigenic stimulation.

Gammaglobulin replacement therapy is beneficial for IgG subclass deficient individuals[8, 10, 15, 16]. Most authors advise prophylactic antibiotic therapy prior to gammaglobulin treatment. In general, patients who do not respond to prophylactic antibiotic therapy, are treated with gammaglobulin. Our patients who did not respond to antibiotic prophylaxis were administered gammaglobulin therapy.

IgG subclass deficiency should be considered and screened in children with unexplained recurrent respiratory tract infections, even in the presence of normal serum total immunoglobulin levels.

REFERENCES

1. Oxelius VA, Laurell AB, Lindquist B, et al. IgG subclasses in selective IgA deficiency: importance of IgG2 - IgA deficiency. *N Engl J Med* 304:1476, 1981.
2. Ochs HD, Wedgwood RJ. Disorders of the B cell system. In Stiehm ER (ed.). *Immunologic Disorders in Infants and Children.* Philadelphia: WB Saunders, pp. 226-256, 1989.
3. Oxelius VA, Berkel AI, Hanson LÅ. IgG2 deficiency in ataxia-telangiectasia. *N Engl J Med* 306:515, 1982.
4. Björkander J, Bake B, Oxelius VA, Hanson LÅ. Impaired lung function in patients with IgA deficiency and low levels of IgG2 or IgG3. *N Engl J Med* 313:720, 1985.
5. Schur PH, Borel H, Gelfand EW, et al. Selective gamma-G globulin deficiencies in patients with recurrent pyogenic infections. *N Engl J Med* 283:631, 1970.
6. Hanson LÅ, Söderström R, Avanzini A, et al. Immunoglobulin subclass deficiency. *Pediatr Infect Dis J* 7 (5 Suppl): S17, 1988.
7. Plebani A, Duse M, Monafo V. Recurrent infections with IgG2 deficiency. *Arch Dis Child* 60:670, 1985.
8. Jefferis R, Kumararatne DS. Selective IgG subclass deficiency: quantification and clinical relevance. *Clin Exp Immunol* 81:357, 1990.
9. Oxelius VA. IgG subclass levels in infancy and childhood. *Acta Paediatr Scand* 68:23, 1979.
10. Schur PH. IgG subclasses - a review. *Ann Allergy* 58:89, 1987.
11. Shackelford PG, Polmar SH, Mayus JL, et al. Spectrum of IgG2 subclass deficiency in children with recurrent infections: prospective study. *J Pediatr* 108:647, 1986.
12. Stanley PJ, Corbo G, Cole PJ. Serum IgG subclasses in chronic and recurrent respiratory infections. *Clin Exp Immunol* 58:703, 1984.
13. Aucouturier P, Lacombe C, Bremard C, et al. Serum IgG subclass levels in patients with primary immunodeficiency syndromes or abnormal susceptibility to infections. *Clin Immunol Immunopathol* 51:22, 1989.
14. Ambrosino DM, Siber GR, Chilmonczyk BA, et al. An immunodeficiency characterized by impaired antibody responses to polysaccharides. *N Engl J Med* 316:790, 1987.
15. Silk HJ, Ambrosino D, Geha R. Effect of intravenous gammaglobulin therapy in IgG2 deficient and IgG2 sufficient children with recurrent infections and poor response to immunization with Hemophilus influenzae type b capsular polysaccharide antigen. *Ann Allergy* 64:21, 1990.
16. Diaz JD, Nelson RP, Lockey RF. IgG subclass deficiency and recurrent infections. [Clinical Conference]. *Ann Allergy* 61:333, 1988.

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ENCOPRETIC CHILDREN: EXPERIENCE WITH FIFTY CASES*

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SUMMARY: Bulut M, Tekant G. (Department of Pediatric Surgery, Şişli Children's Hospital, Istanbul, Turkey). Encopretic children: experience with fifty cases. Turk J Pediatr 33: 167-172, 1991.

Encopresis is defined as the deposition of stools in a child's underwear despite normal defecation control mechanisms.

Fifty children with encopresis were admitted to the Pediatric Surgery Department of Şişli Children's Hospital between October 1, 1987 and January 1, 1990. Two treatment regimens were used. Thirteen patients received mineral oil and glycerine suppositories and 37 patients received rectal irrigations and Lactulose. Five patients were lost to follow-up and 40 patients have completed their six month treatment schedule.

The results of our study suggest that encopresis is a complex abnormal motility disorder, requiring a multidiciplinary approach. Best results will be obtained by good parent-child-doctor cooperation and close follow-up. *Key words: encopresis, fecal soiling.*

In 1926, Weissenberg[1] applied the term "encopresis" to describe the deposition of stools in a child's underwear despite normal defecation control mechanisms. Since these children have completely normal anal sphincters and have established normal bowel control for a period of time, this condition has been accepted by many as being psychogenic in origin[2, 3]. Thus for many years these children mostly underwent psychiatric treatment. But motility studies, anorectal manometry, and electromyography evaluations support a physiopathologic basis for encopresis[4-11]. In the light of the data accumulated on the subject, we set up a treatment protocol and began managing these patients in our department. In time, our experience with encopresis increased, and our patient series expanded. Chronic stool retention has been noticed as the major etiologic factor in this disorder. The aim of this report is to describe the findings of study conducted on encopresis, and to stress the important role of effective stool evacuation with close follow-up in the treatment of these unfortunate children.

Material and Methods

Fifty children with fetal incontinence seen in the Department of Pediatric Surgery at Şişli Children's Hospital between October 1, 1987, and January 1, 1990 were studied. Encopresis was defined as fecal incontinence lasting for a period of no

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less than six months, and occurring more than once a day. A detailed history was obtained and a thorough physical examination was performed to differentiate diseases with similar presenting symptoms. Known predisposing factors for encopresis were investigated in each patient.

Two treatment regimes were used. The first group, who were not hospitalized, included 13 patients. They received mineral oil and glycerine suppositories twice a day. Due to the late response to treatment in the first group, a second group, consisting of 37 patients, received rectal irrigation and Lactulose for a period of three days supervised by a surgeon and a psychologist. Rectal irrigations were administered after breakfast and dinner to stimulate the gastrocolic reflex. All patients were placed on a low-fiber diet for a period of two weeks and they were trained to sit on a toilet for a period of 10 minutes following irrigations which were administered twice a day (Table I). Both regimes were adhered to for three months.

TABLE I: Treatment Programmes

Group 1: 13 patients (Home treatment).
- Mineral oil 2 x 15 - 30 cc / day.
- Glycerine suppositories 2 x 1 - 2 / day.

Group 2: 37 patients (Admitted to hospital).
- Lactulose 2 x 15 - 30 cc / day.
- Rectal irrigation 10 cc / kg.

The patients were scheduled for a six month follow - up following the initial therapy. In the first month the visits were arranged weekly, in the second month once every two weeks, and then once every month. At each visit, the patients were seen by the senior author, and the therapy was modified according to the child's response to the regimen. The treatment regimens were extended to more than three months, if necessary.

At the last follow - up, each child was graded according to response to the regimen. An excellent result was defined as the disappearance of fecal soiling and normal bowel control without the use of therapy. A moderate result consisted of minimal fecal staining of less than three times monthly, and complete resolution with appropriate therapy. A poor result was defined as a child who continued to soil despite therapy.

Results

The clinical characteristics of children with encopresis are presented in Table II. The majority of children in our study were males (n: 32; 64 %). The age

TABLE II: Clinical Characteristics of Study Group

	No. of Patients	Frequency (%)
Male	32	64
Female	18	36
Age at initial clinic visit (yrs).		
4 - 8 yrs	27	54
8 - 12 yrs	23	46
12 above	–	–
Enuresis	19	38
Average age at onset (yrs): 4.4 yrs		

distribution at initial visit to the clinic were: 27 patients between 4 to 8 years (54 %), 23 patients between 8 to 12 years (46 %). The average age of onset was 4.4 years and enuresis was an accompanying symptom in 19 (38 %) patients.

The distribution of predisposing factors are presented in Table III. There was a predominance of simple constipation, anal fissure and psychosocial stress with a frequency of 26 %, 8 %, 12 %, respectively. No predisposition was determined in 19 (38 %) patients.

Table III: Predisposing Factors

	No. of Patients	Frequency (%)
Simple constipation	13	26
Anal fissure	4	8
Psychosocial stress	6	12
Severe gastroenteritis	4	8
Rectal prolapse	1	2
Wrong toilet training	2	4
Family history	1	2
No predisposition	19	38

From the 50-patient study group, five patients (10 %) were lost to follow - up. Forty patients completed their six-month schedule, and the remaining five patients completed an average of three months.

The results of the study group at the last follow - up are presented in Table IV. Excellent results were obtained in 40 patients (88.9 %), while in four patients (8.9 %) there were moderate results, and in one patient (2.2 %) the result was poor. But on the average, the response to treatment was 12.9 days in the first group, and 4.8 days in the second group.

TABLE IV: Results

	Excellent	Moderate	Poor	Total
Group 1 Mineral oil + Glycerine supp.	9	2	–	11
Group 2 Lactulose + Rectal irrigations	31	2	1	34

Discussion

Encopresis is a symptom resulting from chronic stool retention leading to complex abnormal motility. The onset of disease is around the age of four. The majority of patients are constipated and have fecal impaction[4-16]. Chronic stool retention is usually unnoticed by the patient and family. In children with encopresis, the critical volume of rectal distention required to produce conscious awareness is increased. This leads to relaxation of IAS (internal anal sphincter) at volumes that do not stimulate conscious awareness leading to soiling[7]. For this reason children with encopresis have a history of not sensing the urge to defecate before soiling occurs[6, 7].

Thus predisposing factors result in stool retention. The following predisposing factors for encopresis have been determined by Levine[6]:

Stage I : Infants and Toddlers
- a) Simple constipation
- b) Early colonic inertia
- c) Congenital anorectal problems
- d) Other anorectal conditions
- e) Parenteral overreaction
- f) Coercive medical interventions

Stage II : Training and Autonomy (2 - 5 years of age)
- a) Psychosocial stress during training period
- b) Coercive or extreme permissive training
- c) Toilet fears
- d) Painful or difficult defecation

Stage III: Extramural Function - Early School Years
- a) Avoidance of school bathrooms
- b) Prolonged or acute gastroenteritis
- c) Psychosocial stress
- d) Food intolerance
- e) Frenetic life style

Predisposing factors were able to be determined in 62 percent of our patients with a dominance of simple constipation, anal fissure and psychosocial stresses, all supporting chronic stool retention.

Differential diagnosis includes structural diseases of the anus, rectum, colon or secondary abnormalities (endocrinologic, neurologic) or side effects of drugs[6, 7]. A detailed history and thorough physical examination is sufficient to differentiate these diseases. Since soiling, the major presenting symptom in these children is not present in Hirschsprung's disease, we believe that barium enemas and rectal biopsies, used routinely at some centers are unnecessary[16].

Today, most authors agree that therapy is successful only if the colon is evacuated, usually by means of enemas[6, 7, 12, 15, 16]. Our findings support this opinion, too. The group receiving mineral oil and suppositories has been more resistant to therapy than the Lactulose and rectal irrigation group. We believe that rectal irrigations are more efficient in evacuating the over-distended rectum and help regain IAS tonus faster. In fact, 70 percent of our patients receiving rectal irrigations began to have bowel control after their first stool evacuation, and this has led to a positive psychological boost, encouraging them to follow the treatment programme.

For many years, these children have been treated by psychiatrists but present data demonstrates that they do not have clinically significant problems and will not benefit from psychotherapy alone[4, 9, 17]. The aim of this study was to combine psychological support with medical therapy to reduce hospitalization and allow for good parent - child - doctor cooperation. Despite all these efforts, five (10 %) patients were lost to follow - up.

The presence of enuresis as an accompanying symptom in 19 (38 %) children also supports a complex motility disorder. It is interesting to note that after fecal continence was regained, enuresis ceased spontaneously in 12 patients.

A low-fiber diet was introduced for two weeks, with the aim of reducing the bulk in the colon and helping the IAS regain its tonus faster.

In cases highly resistant to medical therapy, a search for food intolerance[8], WBI (whole bowel irrigation)[12] and a surgical approach[18, 19] have been suggested. In fact in this group, a patient was resistant to Lactulose and excellent bowel control was achieved after WBI. Some authors have reported surgical therapy as the first mode of treatment[18, 19]. But the high success rate obtained in the presented study suggests that medical therapy must always be the first choice of treatment in these children.

Conclusion

Since knowledge is limited about bowel movements, the etiology of encopresis still remains a mystery. Encopresis is a result of chronic stool retention and the cornerstone of treatment is first the total evacuation of the bowel. In patients normal bowel control should be regained by toilet training. Therapy should be adapted according to the child's needs, rather than the enforcement of strict protocols. The length of therapy may be extended in cases that are resistant to treatment. Good parent-child-doctor cooperation should be assured. Encopresis with its complex physiopathology, requires teamwork, where either a pediatric gastroenterologist or a pediatric surgeon directs the therapy.

REFERENCES

1. Weissenberg S. Über Enkopresis. *Z Kinderheilkd* 40:674, 1926.
2. Garrard SD, Richmond JB. Psychogenic megacolon manifested by fecal soiling. *Pediatrics* 10:474, 1952.
3. Bemporad JR, Kresch RA, Asnes R, Wilson A. Chronic neurotic encopresis as a paradigm of a multifactorial psychiatric disorder. *J Nerv Ment Dis* 166:472, 1978.
4. Abrahamian FP, Lloyd-Still JD. Chronic constipation in childhood: a longitudinal study of 186 patients. *J Pediatr Gastroenterol Nutr* 3:460, 1984.
5. Cucchiara S, Coremans G, Staiano A, et al. Gastrointestinal transit time and anorectal manometry in children with fecal soiling. *J Pediatr Gastroenterol Nutr* 3:545, 1984.
6. Levine MD. Encopresis: its potentiation, evaluation, and alleviation. *Pediatr Clin North Am* 29:315, 1982.
7. Hatch TF. Encopresis and constipation in children. *Pediatr Clin North Am* 35:257, 1988.
8. Milla PJ. Gastrointestinal motility disorders in children. *Pediatr Clin North Am* 35:311, 1988.
9. Loening-Baucke VA, Cruikshank B, Savage C. Defecation dynamics and behavior profiles in encopretic children. *Pediatrics* 80:672, 1987.
10. Loening-Baucke VA. Sensitivity of the sigmoid colon and rectum in children treated for chronic constipation. *J Pediatr Gastroenterol Nutr* 454:1984.
11. Buser WD, Miner PB Jr. Delayed rectal sensation with fecal incontinence. Successful treatment using anorectal manometry. *Gastroenterology* 91:1186, 1986.
12. Sarahan T, Weintraub WH, Coran AG, Wesley JR. The successful management of chronic constipation in infants and children. *J Pediatr Surg* 17:171, 1982.
13. Lowery SP, Srour JW, Whitehead WE, Schuster MM. Habit training as treatment of encopresis secondary to chronic constipation. *J Pediatr Gastroenterol Nutr* 4:397, 1985.
14. Schnaufer L, Mahesh Kumar AP, White JJ. Differentiation and management of incontinence and constipation problems in children. *Surg Clin N Am* 50:895, 1970.
15. Silber DL. Encopresis-discussion of etiology and management. *Clin Pediatr* 8:225, 1969.
16. Katz C, Drongowski RA, Coran AG. Long-term management of chronic constipation in children. *J Pediatr Surg* 22:976, 1987.
17. Friman PC, Mathews JR, Finney JW, et al. Do encopretic children have clinically significant behavior problems? *Pediatrics* 82:407, 1988.
18. Shandling B, Desjardins JG. Anal myomectomy for constipation. *J Pediatr Surg* 4:115, 1969.
19. Hata Y, Sasaki F, Uchino J. Sphincteromyectomy and sphincteroplasty in chronic constipation with megarectum. *J Pediatr Surg* 23:141, 1988.

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CREATION OF RIGHT VENTRICLE - PULMONARY ARTERY CONTINUITY WITHOUT CARDIOPULMONARY BYPASS*

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SUMMARY: Paşaoğlu İ, Doğan R, Özkutlu S. (Departments of Thoracic and Cardiovascular Surgery, and Pediatric Cardiology, Hacettepe University Faculty of Medicine, Ankara, Turkey). Creation of right ventricle - pulmonary artery continuity without cardiopulmonary bypass. Turk J Pediatr 33: 173-179, 1991.

A closed technique was applied successfully in a patient with Fallot's tetralogy in whom total correction had been previously performed, for creation of continuity between the right ventricle and left pulmonary artery without the use of cardiopulmonary bypass. A surgical approach was achieved by an anterolateral thoracotomy incision in the left third intercostal space. A Dacron non - valved tubular graft, 10 mm. in diameter, was anastomosed to the proximal end of the left pulmonary artery. The proximal end of the graft was anastomosed to the pericardial outflow tract patch with a running technique using a monofilament suture. The peripheral arterial oxygen saturation increased postoperatively, and the patient's condition improved dramatically. *Key words: tetralogy of fallot, pulmonary artery stenosis.*

Intracardiac correction of tetralogy of Fallot (TOF), first described by Lillehei[1] in 1955, has become the treatment of choice for almost all variations of this anomaly. Parallel to the improvement of surgical techniques, anesthesia, extra-corporeal circulation, and postoperative intensive care units, the weight and age of patients no longer constitute an obstacle to correction. However, hypoplastic pulmonary arteries are still an impediment to one stage correction in infants with TOF. In such patients, several palliative surgical methods have been used to improve the pulmonary blood flow before correction. One of these procedures is conduit implantation (with or without valve) between the right ventricle and pulmonary artery.

This report describes establishment of right ventricle - pulmonary artery continuity with a conduit to increase pulmonary blood flow, without the use of cardiopulmonary bypass, In a patient with TOF in whom total correction had been previously performed.

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Case Report

A six-year-old female developed cyanosis at the age of one month and a cardiac murmur was noted. The electrocardiogram revealed a normal sinus rhythm, right axis deviation and right ventricular hypertrophy. An anteroposterior telecardiogram showed diminutive pulmonary vessels and an elevated rounded apex consistent with right ventricular dominance, giving the heart a "coeur en sabot" appearance. Two - dimensional echocardiography revealed TOF. Propranolol was given orally to relieve the hypoxemia. Cardiac catheterization was performed at five years of age. The infundibulum of the pulmonary artery was extremely narrow, and a left ventricular angiogram revealed a normal - sized ventricle. The combination of a large ventricular septal defect (VSD) and overriding of the aorta, which resulted in equalization of pressure in both ventricles, verified the diagnosis of TOF.

When the patient was six years of age, total correction was performed which consisted of closure of the VSD with a Dacron patch using a continuous suture. Since the diameter of the pulmonary annulus was less than the confidence limit for the patient's age, the transannular incision was closed with a pericardial patch extending up to the bifurcation of the pulmonary arteries. Both pulmonary artery orifices were observed to have normal diameters. The next day, low cardiac output developed and the oxygen tension and saturation decreased (PO_2 44 mm Hg and 72 percent). The patient's general condition did not improve. A chest roentgenogram showed decreased vascularity on the left side. Repeat cardiac catheterization and angiography demonstrated proximal occulsion of the left pulmonary artery resulting from stenosis and kinking. The left pulmonary artery was not demonstrated with contrast material (Fig. 1). Since the patient was in

Fig. 1: Anteroposterior view of a right ventricular angiogram, left pulmonary artery was not demonstrated.

poor general condition, it was decided that in order to increase pulmonary blood flow, a closed technique should be applied immediately which would allow establishment of right ventricule pulmonary artery continuity. After a few days passed correct diagnosis was made by catheterization and angiocardiography and a second operation was performed. The chest was incised through the left third intercostal space. The pericardium was opened longitudinally, anterior to the phrenic nerve, and extending up to the pericardial reflection. After the pulmonary bifurcation was dissected from the aorta, it was observed that the left pulmonary artery showed severe narrowing and kinking, 1.5 - 2 cm from the bifurcation (Fig. 2a). There was no blood flow within the left pulmonary artery. The left pulmonary

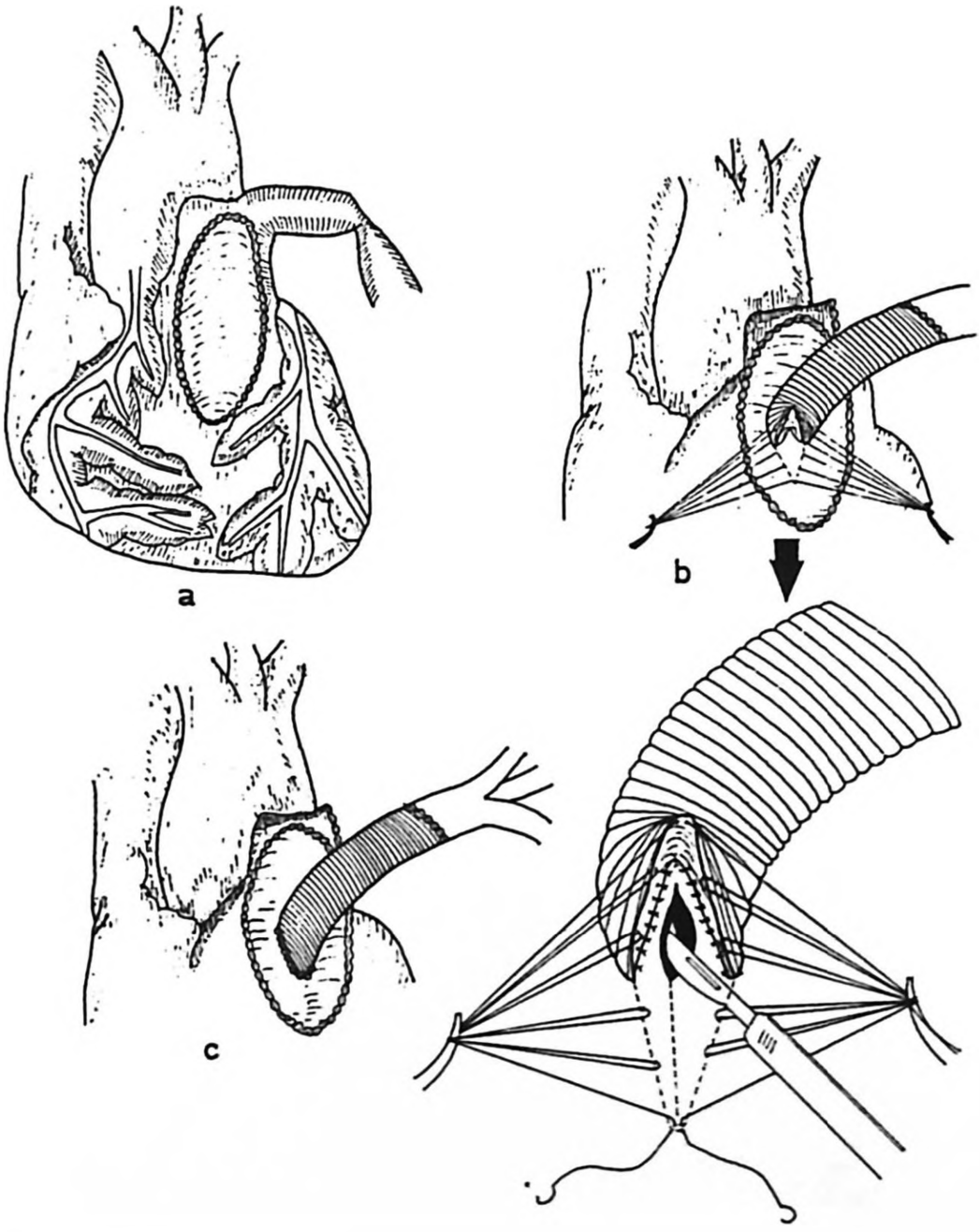

Fig. 2: Schematic presentation of surgical technique.

artery was severed from the bifurcation and the proximal end was closed using a running suture. A vascular clamp was applied to the left pulmonary artery and the narrowed segment was resected. A Dacron non - valved tubular conduit, 10 mm in diameter, was preclotted and end-to-end anastomosis was performed on the left pulmonary artery with a 5/o monofilament continuous suture. The proximal end of the conduit was trimmed to an adequate length and was sutured to the pericardial outflow patch in an elliptical shape using the running suture technique with a 4/o monofilament suture (Fig. 2b). The posterior and lateral suture line was completed, but the last four stitches were allowed to remain loose and retracted. A scalpel was then inserted between these stitches and entered the right ventricular cavity. A pericardial patch was opened along the extent of the anastomosis. To minimize blood loss after the scalpel was removed, the ends of the suture were tied off (Fig. 3). The conduit was stretched and flattened. No pressure gradient was noted between the conduit and the left pulmonary artery.

Fig. 3: Operative view of the implanted Dacron tubular graft between the left pulmonary artery and pericardial right ventricular outflow patch.

Postoperatively, the arterial oxygen saturation increased dramatically (94 %). The next day, scintigraphy demonstrated left lung perfusion to be nearly within normal limits (Fig. 4). The postoperative course was uneventful and the patient was

Fig. 4: Patient's postoperative lung perfusion scintigraphy.

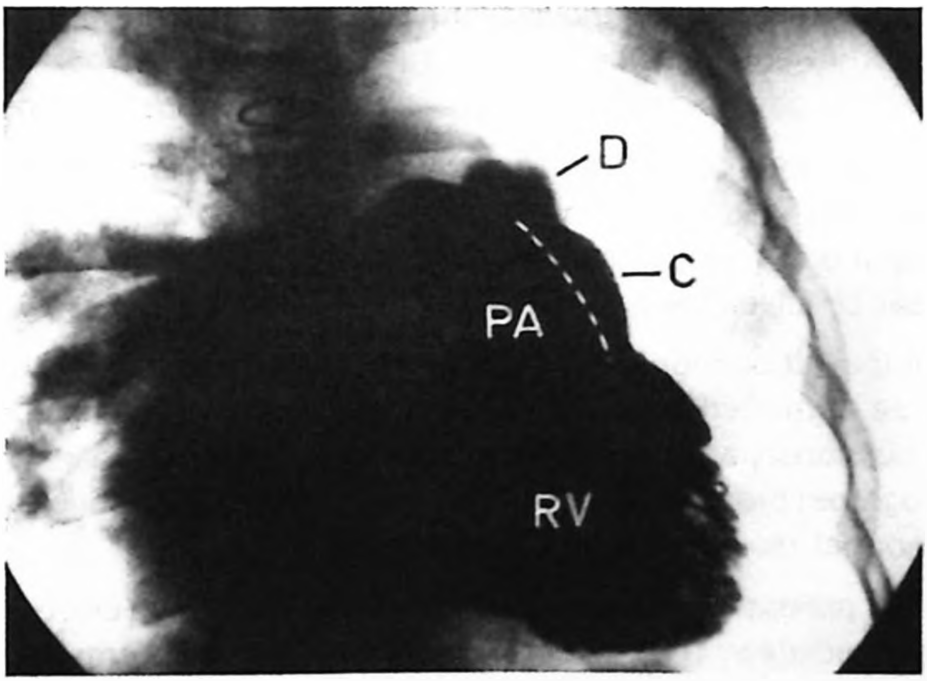

Fig. 5: Right ventricular angiogram, conduit was open but left pulmonary artery was slightly dilated just distal to the anastomosis (C: Conduit, PA: Pulmonary artery, D: Pulmonary artery dilatation).

discharged on the 12th postoperative day. The patient had no complaints and was followed up at six month intervals. At her last follow-up (December 1989), cardiac catheterization was performed which revealed an 8 mm Hg gradient between the left pulmonary artery and the right ventricle (LPA 32/12 mm Hg mean 17, Rt ventricle 40/0-5 mm Hg). An angiogram revealed that the conduit was open, but that the left pulmonary artery was slightly dilated just distal to the conduit.

Discussion

It is possible to reach the ultimate stage of treatment with total correction of TOF. But, in order to avoid right ventricular failure, the use of palliative techniques is preferred in patients with diminutive pulmonary arteries and pulmonary vascular bed as well as in those with a small left ventricular cavity.

However, in infants with severe cyanosis, anoxic spells, high hematocrit levels and low birth weights palliative techniques might be preferred as an immediate measure, until total surgical correction can be performed, because it carries a low mortality. Reconstruction of the right ventricular outflow tract is one of the palliative operations.

Establishment of right ventricle - hypoplastic pulmonary artery continuity without cardiopulmonary bypass was first performed by Puga and colleagues[2] in 1982 on six patients with VSD, hypoplastic pulmonary arteries and total or near total atresia of the pulmonary valve with zero mortality. In the same year, Diaz et al[3] reported using a similar technique which they applied to 11 patients for partial reconstruction of the right ventricular outflow tract without the use of cardiopulmonary bypass. In both studies it was stressed that the oxygen saturation had increased and that good clinical improvement had been observed postoperatively.

In 1985 Maeta and co-workers[4] reported a new technique in which a wire-guided knife was used. The technique had been applied in two patients with TOF and in one with coarctation of the aorta, and neither flow occlusion nor extracorporeal circulation was used.

If stenosis of the left pulmonary artery is recognized preoperatively or intraoperatively, it can be corrected by a patch angioplasty and/or reimplantation into the native main pulmonary artery. However, in our case left pulmonary artery stenosis was not recognized preoperatively, and the pulmonary artery orifice was observed to have a normal diameter intraoperatively.

Because of our patient's poor condition, it was decided that a closed technique be performed immediately. The case presented shows that for creation of continuity between the right ventricle and left pulmonary artery without the use of cardiopulmonary bypass, the closed technique can be applied to a patient with

TOF in whom total correction had been previously performed. We decided to present this case because, as far as we know, this is the first time that such a technique has been used.

REFERENCES

1. Lillehei CW, Cohen M, Warden HE, et al. Direct vision intracardiac surgical correction of tetralogy of Fallot, pentalogy of Fallot, and pulmonary atresia defects: report of first ten cases. *Ann Surg* 142:418, 1955.
2. Puga FJ, Uretzky C. Establishment of right ventricle-hypoplastic pulmonary artery continuity without the use of extracorporeal circulation. *J Thorac Cardiovasc Surg* 83:74, 1982.
3. Diaz FA, Salvador JC, Zurdo GC. Partial reconstruction of right ventricular outflow tract without cardiopulmonary bypass [letters]. *J Thorac Cardiovasc Surg* 83:149, 1982.
4. Maeta H, Sakakibara Y, Koishizawa T, et al. Patch enlargement of the cardiovascular flow tract with neither flow occlusion nor extracorporeal circulation. *J Thorac Cardiovasc Surg* 89:146, 1985.

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HEREDITARY FRUCTOSE INTOLERANCE IN A PATIENT WITH PHENYLKETONURIA*

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SUMMARY: Coşkun T, Özalp I, Tekinalp G. (Department of Pediatrics, Hacettepe University Faculty of Medicine, Ankara, Turkey). Hereditary fructose intolerance in a patient with phenylketonuria. Turk J Pediatr 33: 181-184, 1991.

Classical phenylketonuria (PKU) and hereditary fructose intolerance (HFI) are two inborn errors of metabolism that have an autosomal recessive mode of inheritance. In this paper, we described a 3 - year - old girl with PKU and HFI. The occurrence of these two defects in the same patient is thought to be fortuitous and not genetically related since this is the first reported case and the statistical probability of such an occurrence is very low. *Key words: phenylketonuria, hereditary fructose intolerance.*

Classical phenylketonuria (PKU), resulting from a deficiency of the enzyme phenylalanine hydroxylase (PAH) is transmitted as an autosomal recessive trait[1]. Hereditary fructose intolerance which is caused by a deficiency of the enzyme fructaldolase B (HFI) also has an autosomal recessive mode of inheritance[2]. The probability of these two inborn errors in the same patient must be low since it has not been described before in the literature. This paper describes a 3-year-old girl with PKU and HFI.

Case Report

B.E., a 3-year-old girl, was transferred to the Neonatal Intensive Care Unit of our hospital two hours after birth, because of a history of sibling deaths in the family occurring within the first few months of life. Pregnancy and delivery were uneventful. Birth weight was 3760 g (75^{th} percentile), length 53 cm (95^{th} percentile), and head circumference 36 cm (95^{th} percentile). The parents were first cousins. The first-born child, a 7-day-old boy, had been admitted to our hospital with an abdominal mass, but died shortly after admission. Hamartoma of the liver had been found at autopsy. A urine sample from the second-born child, a 3-month-old boy, had been analyzed in our laboratory. A generalized aminoaciduria associated with glucosuria and proteinuria led to the diagnosis of the Fanconi syndrome. Subsequently, the child died and further investigations could not be carried out to find the etiology of the Fanconi syndrome.

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The initial physical examination in the nursery revealed unremarkable findings. The early neonatal course was complicated by hypoglycemia, and was treated with an intravenous glucose solution. She was breast-fed thereafter.

A complete routine blood count and urinalysis produced normal results. A test for reducing substance in the urine was negative. The blood sugar level fluctuated between 15-120 mg/dl. Transaminase activities were within the normal range. The leukocyte glycogen content was 10 µg / mg protein (normal ≤ 50 µg/mg protein); blood lactate was 34 mg/dl (normal 10-14 mg/dl) and pyruvate 1.33 mg/dl (normal 0.5 - 1 mg/dl). Serum and urinary amino acid patterns were normal. A repeat chromatogram of serum at the end of the first month demonstrated a high level of phenylalanine (23 mg/dl).

The patient was placed on a low phenylalanine diet. Sucrose and vegetable oil were added to her dietary regimen to meet energy requirements. She vomited the first two or three feedings and developed severe acidosis (CO_2 content 5.8 mEq/L). She was comatose, and the liver was palpable 5 cm below the right costal margin. SGOT was 200 IU, SGPT 310 IU, blood sugar 9 mg/dl, phosphorus 2.5 mg/dl, uric acid 9.5 mg/dl, potassium 3.5 mEq/L, lactate 209 mg/dl, and pyruvate 5 mg/dl. There was a generalized amino aciduria.

Intravenous fluid and bicarbonate therapy were initiated. The acidosis did not respond to bicarbonate therapy and peritoneal dialysis was performed. Her general condition improved gradually and she began to tolerate oral feedings. Sucrose was eliminated from the low phenylalanine diet.

When she was 15-months-old, she experienced a second attack, after the ingestion of fruit, which was similar to the first one noted at the age of 1.5 months. The second attack responded to the infusion of bicarbonate and glucose.

She received a low phenylalanine and sucrose/fructose free formula. She did not experience another such episode. The blood phenylalanine level was maintained within the normal range, and the renal tubular functions returned to normal. Her physical growth and mental-motor development were normal.

Discussion

The association of PKU with scleroderma[3], Duchenne muscular dystrophy[4], Charcot - Marie - Tooth disease[5], Down's syndrome[6], cystinuria[7], homozygous hypobetalipoproteinemia[8], and bilateral iris coloboma and optic atrophy[9] have been reported previously. Also, there have been cases reported in the literature describing PKU and galactosemia[10] and PKU and glycogenosis[11] within the same family. A review of the literature revealed neither the occurrence of PKU and HFI in the same patient nor within the same family.

A phenylalanine level of 23 mg/dl with a normal tyrosine level and a positive ferric chloride test of the urine led to the diagnosis of PKU in our patient. A subsequent BH_4 loading test showed no BH_4 deficiency.

Although fructaldolase B activity was not measured, the development of vomiting, lethargy, hepatomegaly following the ingestion of fructose from sucrose which was added to the low phenylalanine diet and from the fruit, coupled with a positive family history in which one sibling had died having similar clinical signs and symptoms led to the diagnosis of HFI. Biochemical evidence of hepatic malfunction, generalized hyperaminoaciduria due to defective renal tubular reabsorption, hypoglycemia, hyperuricemia, hypophosphatemia, a low serum potassium level and metabolic acidosis supported the diagnosis of HFI[2].

In Turkey PKU has a frequency of 1/3954 and is not uncommon[12]. The gene controlling the production of PAH is located on chromosome 12[13]. Although, the actual incidence of the deficiency of fructaldolase B, an enzyme that is controlled by a gene locus on chromosome 9, is not known, it is presumed to be around 1:20,000 as reported from Switzerland[2, 14]. If these figures are taken into consideration, the statistical probability of the two defects co-existing is 1:79,080,000.

Since this is the first case reported in which the two defects were associated, we believe that the combination of these two hereditary diseases, with different chromosomal locations, are not genetically related and that their occurrence was purely coincidental. The consanguinity of the parents should be considered the most important factor regarding this coincidence.

REFERENCES

1. Tourian A, Sidbury JB. Phenylketonuria and hyperphenylalaninemia. In Stanbury JB, Wyngaarden JB, Fredrickson DS, et al (eds). *The Metabolic Basis of Inherited Disease* (5th ed). New York: McGraw Hill, 1983, p. 270.
2. Gitzelmann R, Steinmann B, Van den Berghe G. Essential fructosuria, hereditary fructose intolerance and fructose-1, 6-diphosphatase deficiency. In Stanbury JB, Wyngaarden JB, Fredrickson DS, et al (eds). *The Metabolic Basis of Inherited Disease* (5th ed). New York: McGraw Hill, 1983, p. 118.
3. Kornreich HK, Shaw KN, Koch R, Hanson V. Phenylketonuria and scleroderma. *J Pediatr* 73:571, 1968.
4. Roth KS, Cohn RM, Berman P, Segal S. Phenylketonuria and Duchenne muscular dystrophy: a case report. *J Pediatr* 88:689, 1976.
5. Meier C, Lütschg J, Vassella F, et al. Progressive neural muscular atrophy in a case of phenylketonuria. *Dev Med Child Neurol* 17:625, 1975.
6. Fisch RO, Horrobin JM. Down's syndrome with phenylketonuria. *Clin Pediatr* (Phila) 7:226, 1968.
7. Minami R, Olek K, Wardenbach P. Phenylketonuria in a patient with cystinuria. *Hum Genet* 28:319, 1975.

8. Leititis JU, Stahl M, Tackmann W, et al. Homozygous hypobetalipoproteinaemia and phenylketonuria. *Eur J Pediatr* 144:174, 1985.
9. Tanzer F, Özalp İ, Yakut A, Bilgiç S. Bir fenilketonüri vakasında bilateral iris kolobomu ve optik atrofi. *Çocuk Sağlığı ve Hastalıkları Dergisi* 24:263, 1981.
10. Fisch RO, Goosens KA, Tsai MY, et al. The occurrence of phenylketonuria and galactosemia within the same family. *Clin Pediatr (Phila)* 24:456, 1985.
11. Coşkun T, Özalp İ, Koçak N, Tekinalp G. Aynı ailede fenilketonüri ve glikojenozis. *Çocuk Sağlığı ve Hastalıkları Dergisi* 29:233, 1986.
12. Özalp İ. Coşkun T, Tokol S, et al. Incidence of PKU and other HPA in a sample of Turkish newborn population. *Abstracts of 19th International Congress of Pediatrics*, Paris, July 23-28, 1989, p. 354.
13. McKusick VA. *Inheritance in Man: Catalogs of Autosomal Dominant, Autosomal Recessive and X-Linked Phenotypes* (7th ed). Baltimore: The Johns Hopkins University Press, 1986, p. 1193.
14. McKusick VA. *Inheritance in Man: Catalogs of Autosomal Dominant, Autosomal Recessive and X-Linked Phenotypes* (7th ed). Baltimore: The Johns Hopkins University Press, 1986, p. 976.

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MULTIPLE PRIMARY MALIGNANCY: A REPORT ON LANGERHANS CELL HISTIOCYTOSIS ASSOCIATED WITH HODGKIN'S DISEASE*

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SUMMARY: Karadeniz C, Sarıalioğlu F, Göğüş S, Akyüz C, Küçükali T, Kutluk T, Büyükpamukçu M. (Hacettepe University Institute of Oncology, Ankara, Turkey). Multiple primary malignancy: a report on Langerhans cell histiocytosis associated with Hodgkin's disease Turk J Pediatr 33: 185-190, 1991.

Since multiple primary malignant tumors are rare in children, their presence can be a diagnostic and therapeutic problem. In this report, we present a six - year - old boy with Langerhans cell histiocytosis and Hodgkin's disease. On admission, the patient had lytic lesions and a periosteal reaction on the left trochanter major from which an open bone marrow biopsy was performed. The biopsy revealed Langerhans cell histiocytosis. Eight months later, the child presented with enlarged left cervical lymph nodes and the biopsy demonstrated Hodgkin's disease. Although there was an eight - month interval between the two histopathological diagnoses, the diffuse pulmonary parenchymal infiltration observed on the first admission, was later confirmed by an open - lung biopsy as Hodgkin's disease. The patient was said to have two concurrent lymphoreticular malignancies. To our knowledge, this is the youngest case reported with this association in the English language literature. *Key words: Langerhans cell histiocytosis, Hodgkin's disease, multiple primary malignancy, childhood tumors.*

Histiocytosis X denotes a poorly understood group of disorders affecting both children and young adults. Recently, The Writing Committee for the Histiocyte Society used the term, "Langerhans cell histiocytosis" (LCH) in order to identify this disorder among childhood histiocytoses[1].

Few cases have been reported of the association of Hodgkin's disease and Langerhans cell histiocytosis occurring in the same patient[2-4]. Most of the reported cases describe lesions involved in Langerhans cell histiocytosis as occurring in the lymph nodes of patients with malignant lymphoma.

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We recently had a patient with Langerhans cell histiocytosis who had been diagnosed eight months previously, and who had later developed cervical lymphadenopathy and progressive pulmonary infiltration. Histologically, the lesions in both sites proved to be typical of Hodgkin's disease.

Case Report

A six - year - old boy with an eight - month - history of left hip pain was seen in another hospital in July 1987. An open - bone biopsy was obtained from the left trochanter major and a pathological report showed LCH. The patient was referred to our hospital for further evaluation and therapy. Physical examination revealed normal findings except for a left trochanteric incision. A two-dimensional chest X - ray showed hilar lymphadenopathy and bilateral parenchymal infiltration. Roentgen studies of all bones did not disclose any abnormalities except for some lytic lesions and a periosteal reaction at the left trochanter major. Other laboratory studies including complete blood count, urinalyses, liver and renal function tests, and bone marrow aspiration were normal.

We did not perform pulmonary function tests because the patient had no respiratory symptoms. Thus the patient was assigned to stage IB LCH according to the classification devised by Raney et al[5]. He was put on chemotherapy which consisted of vinblastine (0.2 mg/kg, i.v. weekly) and prednisolone (2 mg/kg, p.o, daily).

After two months, there was no response to the therapy, as indicated by no changes in the pulmonary and skeletal lesions. The chemotherapy was changed to a regimen of methotrexate (30 mg/m^2, p.o. weekly) and 6 - mercaptopurine (75 mg/m^2, p.o. daily).

In December 1987, after five months of chemotherapy, there was progressive enlargement of the cervical lymph nodes. A lymph node biopsy was performed on March 4, 1988 and a diagnosis of Hodgkin's disease was established.

The routine staging procedure including complete blood count, liver and kidney function tests, two dimensional chest X - rays, abdominal ultrasonography and bone marrow biopsy was performed.

On the chest X - rays, hilar lymphadenopathies were visible and the entire lung parenchyma had a diffuse reticulo - granular appearance. Abdominal ultrasonography revealed multiple lymphadenopathies which were one cm in diameter in the paraaortic and parailiac regions. Since pulmonary involvement in Langerhans cell histiocytosis occurring without any parenchymal organ infiltration is rare in a six - year - old child, the pulmonary lesions observed were thought to be a manifestation of Hodgkin's disease. Moreover, the chemotherapy given to treat Langerhans cell disease had not been effective. The patient was assigned to stage IV Hodgkin's disease and chemotherapy with C - MOPP was initiated[6].

Following three courses of chemotherapy, there was no improvement seen in the patient's disease status. A bone survey showed multiple lytic lesions and periosteal reactions in different bones. The chest X - ray revealed a small area of consolidation in addition to apparent progression in the pulmonary lesions. An open left lung biopsy was performed to clarify the lung pathology (July 29, 1988). The histopathological examination again revealed Hodgkin's disease. Despite different types of combined therapeutic intervention (chemotherapy and radiotherapy), the patient died of progressive pulmonary disease two years after his first admission.

Histopathologic Findings:

First biopsy (July 23, 1987): The histopathological evaluation of the left trochanter major biopsy showed granulomas of various diameters which were composed of mixed inflammatory cells with large numbers of eosinophils and histiocytes. Some of the histiocytic cells had large cytoplasms and large lobulated nuclei. There were no Reed - Sternberg cells (Fig. 1).

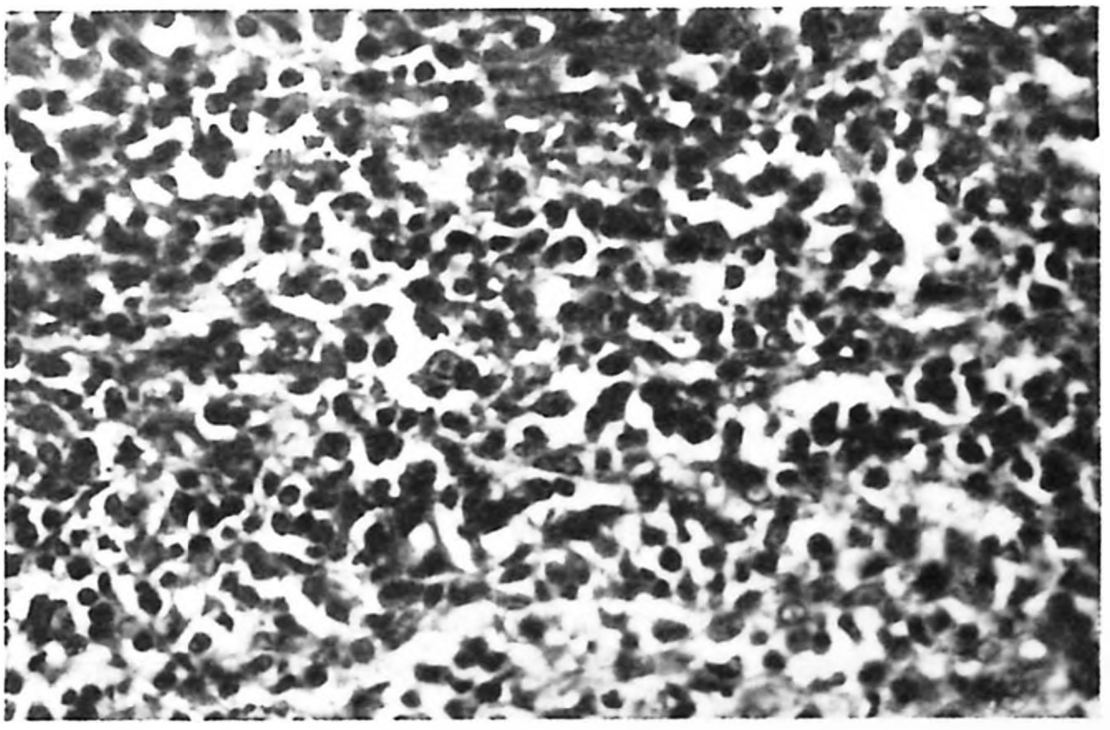

Fig. 1: Biopsy obtained from the left trochanter of the femur.
Dense infiltration of mixed inflammatory cells and some histiocytic cells. (HE x 400).

Second biopsy (March 4, 1988): The histopathological findings of the left cervical lymph node biopsy showed a cellular pattern consisting of lymphocytes, histiocytes, plasma cells and few eosinophils with some areas of nodularity and sclerosis. The typical Reed - Sternberg cells and variants were present (Fig. 2).

Third biopsy (July 29, 1988): The typical Reed - Sternberg cells and variants were

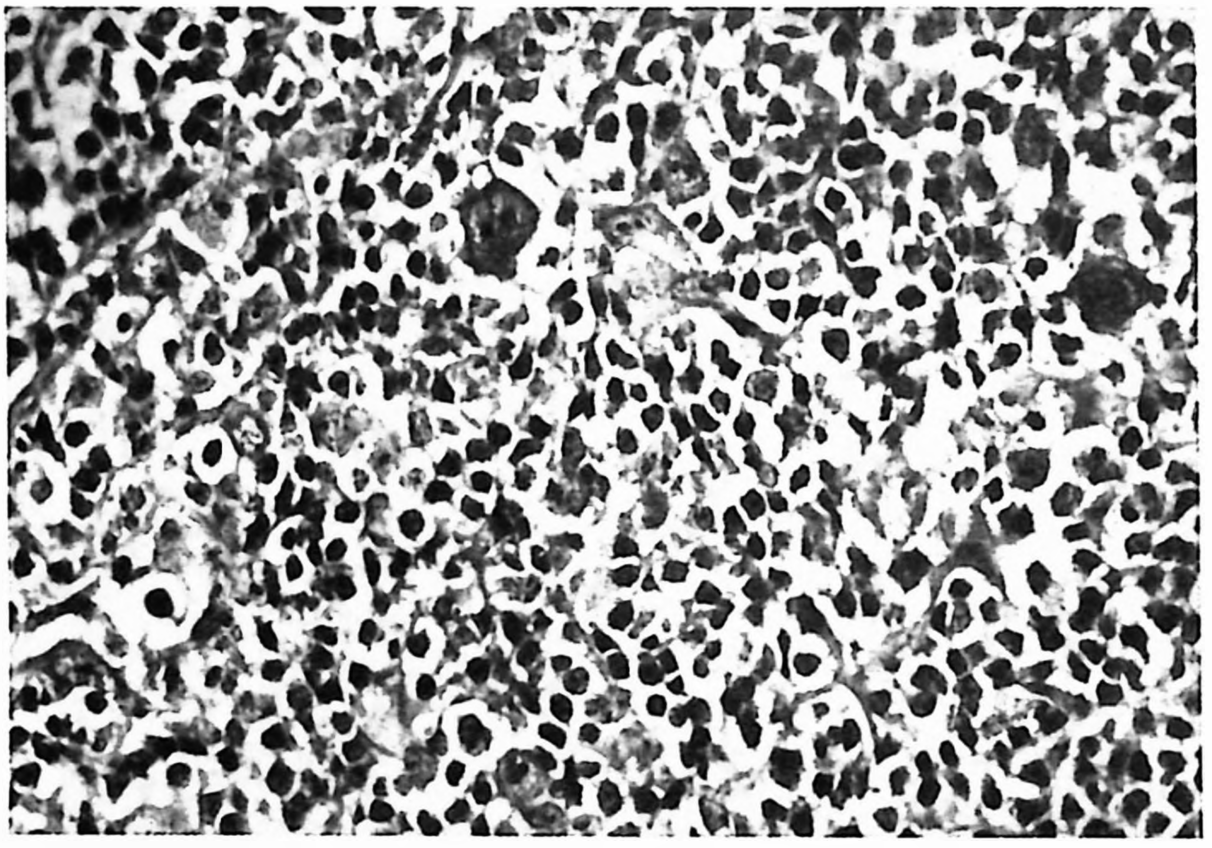

Fig. 2: Cervical lymph node biopsy showing mixed cellularity. Typical Reed - Sternberg cells and Hodgkin's cells (HE x 460).

seen in the open left lung biopsy. Hypocellularity and lymphocyte depletion were also noted.

Discussion

Generally, the pulmonary involvement of LCH is seen in the disseminated form in children[7]. Radiologic findings include micronodular and reticular opacities which may form large nodules with a honeycomb appearance, which may sometimes lead to pneumothorax[8]. In our case, the pulmonary findings on admittance was due to the original disease, LCH. However, the lung biopsy obtained during the progression of the disease clarified the etiology as Hodgkin's disease.

The most frequent intrathoracic findings in Hodgkin's disease are tracheobronchial and mediastinal lymph node involvement[9]. Pulmonary parenchymal involvement is found to be around 58 percent in necropsy series[9], and it is almost always associated with mediastinal lymph node involvement[10]. On the first admission, our patient presented with both mediastinal lymph node and pulmonary involvement. Primary skeletal involvement is rarely seen in Hodgkin's disease, but in our patient both the progression of the disease and the bone biopsy were typical of LCH[11]. According to reports in the literature, Hodgkin's disease is rarely associated with eosinophilic granulomas[2-4].

The etiologies of both Hodgkin's disease and LCH are unknown. Histologically they bear some similarities. In both diseases, especially mixed cell Hodgkin's disease, histiocytes and eosinophils are seen. In addition, infectious agents and immunologic factors have been suggested in their pathogenesis[12-14].

According to the literature, when these two diseases occur in association there is usually involvement of the same lymph node. In the six cases reported by Kjeldberg and Kim[2], eosinophilic granuloma and lymphoma were present in the same lymph node (three Hodgkin's disease cases and three non - Hodgkin's lymphoma cases). Transformation from benign to malignant lesions in the same lymph node has not been demonstrated. Kjeldberg and Kim[2] proposed that the histiocytic proliferation in eosinophilic granuloma could be a reaction to etiologic factors of malignant lymphoma or to malignant lymphoma itself or to an aberrant host response. Since the association of Hodgkin's disease with LCH is a rare occurrence, it is considered to be a coincidence. In a case reported by Frederiksen and Thommesen[3], LCH was diagnosed by biopsy of the gingival mucosa and mandibula. A year later, a cervical lymph node biopsy taken from the same patient revealed Hodgkin's disease. The patient died the following year.

In our case, biopsy of the femur showed LCH while the cervical lymph node biopsy taken eight months later revealed mixed cellular Hodgkin's disease. Frederiksen and Thommesen[3] suggested that a benign LCH lesion could have developed into Hodgkin's disease. However, the Birbek granules seen in the histiocytes of LCH have not been demonstrated in Hodgkin's disease. Since there are no histological similarities between Hodgkin's disease and aggressive histiocytosis, and the fact that LCH not only occurs in combination with Hodgkin's disease but also with non - Hodgkin's lymphoma, contradicts this proposal. Besides, Robert J. L'Hoste and colleagues[15] published a case with massive cervical lymphadenopathy in which an initial diagnosis of Hodgkin's disease was established and later histopathological sections also revealed LCH findings.

In conclusion, the rare association of LCH with Hodgkin's disease in the same patient suggests that it is most probably coincidental, as proposed by Kjeldberg and Kim[2], and that the occurrence of both diseases in the same patient may lead to difficulties in diagnosis and treatment. The diagnosis of Hodgkin's disease in a patient with LCH or LCH in a patient with Hodgkin's disease may therefore be missed.

REFERENCES

1. Chu T, D'Angio GJ, Favara BE, et al. Histiocytosis syndromes in children [letter]. *Lancet* 2:41, 1987.
2. Kjeldsberg CR, Kim H. Eosinophilic granuloma as an incidental finding in malignant lymhoma. *Arch Pathol Lab Med* 104:137, 1980.

3. Frederiksen P, Thommesen P. Histiocytosis X. III. Clinical value of serial biopsies. *Acta Radiol Oncol Radiat Phys Biol* 17:362, 1978.
4. Lyon JM, Pezzimenti J, Kranwinkel RN. The development of histiocytosis X in patient with Hodgkin's lymphoma. *Conn Med* 49:354, 1985.
5. Matus-Ridley M, Raney RB Jr, Thawerani H, Meadows AT. Histiocytosis X in children: patterns of disease and results of treatment. *Med Pediatr Oncol* 11:99, 1983.
6. Leventhal BG, Donaldson SS. Hodgkin's disease. In Pizzo PA, Poplack DG (eds), *Principles and Practice of Pediatric Oncology* (1st ed). Philadelphia: Lippincott, 1989, p. 458.
7. Carlson RA, Hattery RR, O'Connell EJ, Fontana RS. Pulmonary involvement by histiocytosis X in the pediatric age group. *Mayo Clin Proc* 51:542, 1976.
8. Stanford W, Spivey C, Lindberg EF, et al. Eosinophilic granuloma of the lung. *Ann Thorac Surg* 11:299, 1971.
9. Colby TV, Hoppe RT, Warnke RA. Hodgkin's disease at autopsy: 1972-1977. *Cancer* 47:1852, 1981.
10. Strickland B. Intra-thoracic Hodgkin's disease. II. Peripheral manifestations of Hodgkin's disease in the chest. *Br J Radiol* 40:930, 1967.
11. Newcomer LN, Silverstein MB, Cadman EC, et al. Bone involvement in Hodgkin's disease. *Cancer* 49:338, 1982.
12. Order SE, Hellman S. Pathogenesis of Hodgkin's disease. *Lancet* 1:571, 1972.
13. Long JC. The immunopathology of Hodgkin's disease. *Clin Haematol* 8:531, 1979.
14. Nesbit ME Jr, O'Leary M, Dehner LP, Ramsay NK. The immune system and the histiocytosis syndromes. *Am J Pediatr Hematol Oncol* 3:141, 1981.
15. L'Hoste RJ Jr, Arrowsmith WR, Leonard GL, McGaw H. Eosinophilic granuloma occurring in a patient with Hodgkin disease. *Hum Pathol* 13:592, 1982.

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URETHROCUTANEOUS FISTULA DUE TO A COIL OF HAIR*

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SUMMARY: Gülmez I, Tatlışen A, Karacagil M, İpekcan C. (Department of Urology, Erciyes University Faculty of Medicine, Kayseri, Turkey). Urethrocutaneous fistula due to a coil of hair. Turk J Pediatr 33: 191-193, 1991.

Strangulation of the penis by a coil of hair is often an unrecognized clinical entity with severe potential complications. In this report, a five - year - old boy with urethrocutaneous fistula caused by the wrapping of a single strand of hair around the penis is presented. *Key words: penile tourniquet syndrome, urethrocutaneous fistula.*

Strangulation of the penis by an encircling object is an uncommon clinical presentation[1], and has been reported following the application of numerous foreign objects including rings, bottles, rubber bands, pipes, threads, ribbons, metal and plastic washers and metal nuts[2, 3]. These objects are placed on the shaft of the penis sometimes to enhance sexual performance[4]. A frequently unrecognized and potentially devastating form of penile strangulation is that caused by human hair[3]. In this report, a case of urethrocutaneous fistula due to a coil of human hair is presented.

Case Report

A five - year - old boy (M.Ö.) had been urinating through a fistula beneath the corona glandis and external meatus for two months. He had been circumcised two years ago. Physical examination of the penis revealed a moderately swollen glans with marked annular constriction at the level of the coronal sulcus. This constricting ring was covered with necrotic debris (Fig. 1) which was removed meticulously. A long strand of hair was found wrapped around the penis several times. The coil of hair was removed by cutting (Fig. 2). The urethra had been transsectioned completely by the hair and the coronal groove was markedly manifest (Fig. 3). The father informed us that he had removed a coil of hair, which had been wrapped around the child's penis, two months after the patient had been circumcised and that there was no evidence of fistula formation at that time.

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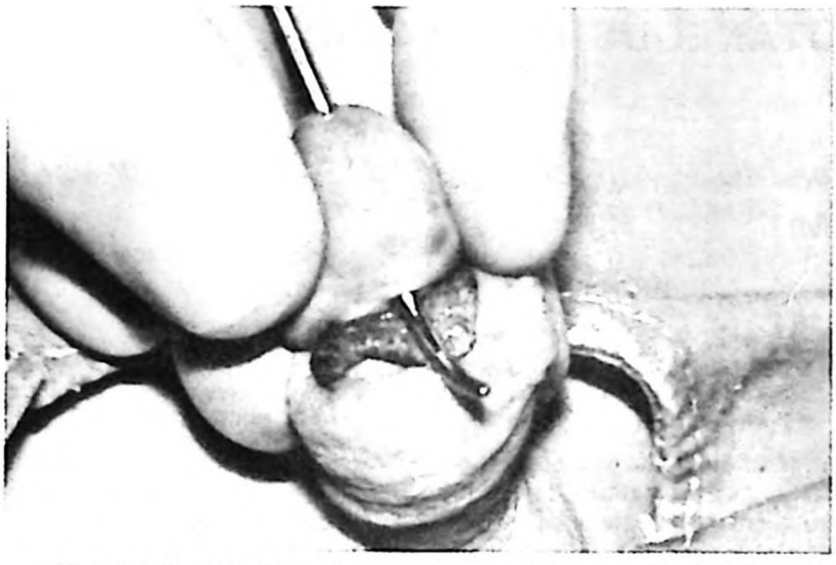

Fig. 1: Constricting ring and urethrocutaneous fistula.

Fig. 2: Removed hair and debris.

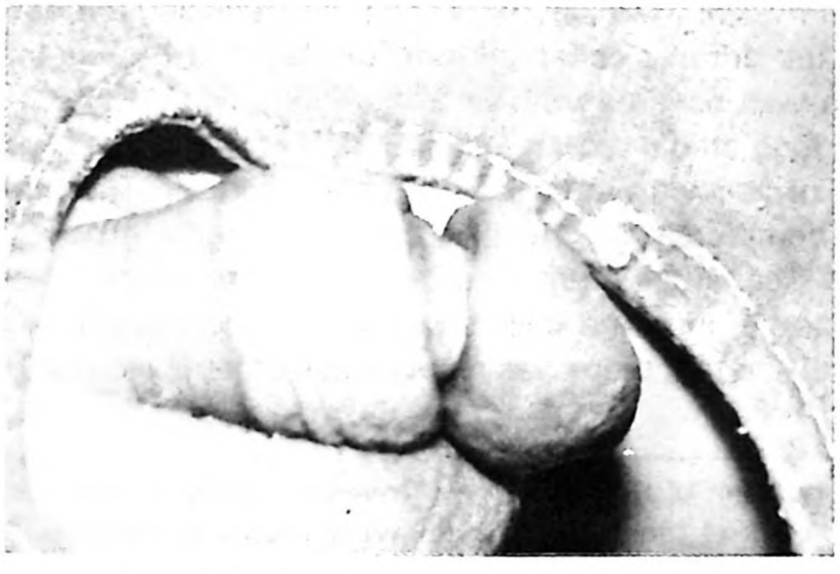

Fig. 3: Following removal of necrotic debris and the hair, a markedly deep coronal groove is seen.

The patient was hospitalized. The penis was cleansed with a solution of povidone iodine twice daily and ampicillin (250 mg qid) therapy was instituted. Seven days later, the patient was discharged. The parents were advised to have the fistula repaired, but the patient was not brought back to the hospital.

Discussion

Penile strangulation due to human hair was first reported in 1888, and since then, a number of cases have been reported. This type of strangulation can cause ulceration, fistula, necrosis and partial or complete amputation of the glans. The patients are always children, with a range in age from 21 days to eight years, and circumcision is an important predisposing factor[3]. In Turkey, only one case of urethrocutaneous fistula secondary to a human hair has been reported[5].

Since human hair is quite stretchable when wet and contracts when dry, it is not difficult to understand how hairs can tighten as they dry and become embedded in the skin. Because human hair is extremely fine, its presence as a foreign body may not be noticed, and the diagnosis may be missed unless the clinician is alert to the possibility of this unusual cause of penile strangulation[6].

How the hair gets wrapped around the penis is a matter of speculation. Application may be accidental or intentional by either the patient, siblings, parents or other adults. Some parents wrap hair around the penis to control enuresis[3]. In our case, the coil was not wrapped around the penis for any specific purpose. We postponed reconstructive surgery until complete healing of the tissues.

REFERENCES

1. Sinha BB. Penile incarceration by a metallic object. *Br J Surg* 75:33, 1988.
2. Fiumara NJ. Annular constriction of the penis-the tourniquet syndrome [letter]. *JAMA* 240:2153, 1978.
3. Sheinfeld J, Cos LR, Ertürk E, Cockett AT. Penile tourniquet injury due to a coil of hair. *J Urol* 133:1042, 1985.
4. McLaughlin T, Coyner W. Removal of a strangulating metal bearing from the penis. *J Urol* 141:617, 1989.
5. Yalçın V. Enteresan bir üretral fistül olgusu. *Türk Üroloji Dergisi* 14:357, 1988.
6. Alpert JJ, Filler R, Glaser HH. Strangulation of an appendage by hair wrapping. *N Engl J Med* 273:866, 1965.

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THE HISTORY OF BETA-THALASSEMIA IN TURKEY*

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SUMMARY: Aksoy M. (Department of Internal Medicine, Istanbul University Faculty of Medicine at Çapa, Istanbul, Turkey, and Department of Biology, Turkish Technical Research Council (TUBITAK), Gebze, Kocaeli, Turkey) The history of beta-thalassemia in Turkey. Turk J Pediatr 33: 195-197, 1991.

The first two patients with beta-thalassemia major in Turkey, were reported in 1941. However, the importance of beta-thalassemia as a health problem was brought to the attention of physicians only after 1950. In Turkey, the clinical and hematological pictures of beta-thalassemia were found to be mostly heterogenous. Hematological data of patients with mild disease (beta-thalassemia intermedia) severe beta-thalassemia (beta-thalassemia major) and heterozygotes indicate that patients can be grouped according to the content of Hb F, Hb A_2 and red cell indices. *Key words: thalassemia, hemoglobinopathies.*

The thalassemias and the sickle cell syndromes are the most frequent genetic disorders occurring throughout the world. Curiously, enough, the first cases of thalassemia major were diagnosed in 1925 by Cooley and Lee[1] in the United States. As rightfully pointed out by Wintrobe[2] it is difficult to comprehend why there were no cases of thalassemia major reported in Italy and Greece during those years. A possible explanation for this might have been the fact that, at that time, all unexplained hepatosplenomegalies were diagnosed as Banti's syndrome[2]. Even today, although quite uncommon, there are some patients with splenomegaly in which a diagnosis of thalassemia has not been established.

The first two cases of Cooley's anemia in Turkey were simultaneously reported by Prof. S. Tavat[3] and Prof. E. Frank[4] in 1940. The parents of Frank's patient were immigrants from Bulgaria, while the father of Tavat's patient was an immigrant from Yugoslavia. In the period that followed, numerous cases of Cooley's anemia were reported[5-9]. The first "International Symposium on Abnormal Hemoglobins and Thalassemia" sponsored by the Council for the "International Organization of Medical Sciences" and organized by Prof. T.H.J. Jonxis was held in Istanbul. At the symposium, the author of this paper presented a report entitled. "Abnormal Hemoglobins and Thalassemia in Turkey".[10] It was proposed in this presentation that considering the wide variation of clinical and hematological pictures observed in some cases of sickle cell-thalassemia disease, there must be more than one thalassemia determinant that interacts with the sickle cell gene. A short while

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later, Ingram and Stratton[11] reported that the genes for thalassemia were responsible for alpha and beta polypeptide syntheses.

In 1963, the author published the first case of homozygous Hb S and alpha-thalassemia seen in a Turkish family[12].

Incidence in Turkey: The first study to determine the incidence of beta-thalassemia in Turkey was performed by Aksoy and Lehmann[13] in which the paper electrophoresis method was used on blood samples taken from 240 Turks, mostly inhabiting the southern Mediterranean coast of Turkey. The incidence rate was found to be 0.4 percent. This figure is considered today as being too low. The most accurate data was reported by Çavdar and Arcasoy[14], who estimated it as around two percent. The incidence of beta-thalassemia, varying widely in different regions of Turkey, ranges between 0.6 and 10.8 percent. The lowest incidence was found by Kürkçüoğlu and workers[15], in the eastern province of Erzurum, and the highest, 10.8 percent, was found by the author and workers[16] to be amongst Turks who originated from Western Thrace.

Silent beta-thalassemia genes: Even though rarely, some genes for beta-thalassemia in the heterozygous state may not cause hematologic abnormalities such as the characteristic microcytosis. The author has been in favor of this assumption since 1959. In 1968, Schwartz[17] showed that some genes responsible for beta-thalassemia may not cause hematologic abnormalities, which he termed "silent genes" for beta-thalassemia. For a long period in the past, it had been generally thought that there was a large amount of fetal hemoglobin in individuals with Cooley's anemia. But, contrary to this, in 1965 the author reported four cases of Cooley's anemia with low levels of fetal hemoglobin ranging between 2 to 10 percent[18].

In addition, in 1978, the author classified 20 patients with beta-thalassemia intermedia according to the results of genetic studies[19] as follows:

1– Beta-thalassemia intermedia homozygous for beta-thalassemia with large amounts of hemoglobins A_2 and F.

2– Beta-thalassemia intermedia homozygous for beta-thalassemia with normal levels of hemoglobins A_2 and F.

3– Beta-thalassemia ihtermedia heterozygous for both beta-thalassemia with large amounts of hemoglobins A_2 and F and beta-thalassemia with normal Hb A_2 and F.

4– Beta-thalassemia intermedia heterozygous for both beta-thalassemia with a large amount of hemoglobin A_2 and "silent" beta-thalassemia.

There are two types of beta-thalassemia intermedia: One is associated with large amounts of hemoglobin F, while in the other, hemoglobin F is only slightly increased.

In recent years, further investigations using new methods have revealed the presence of "silent" abnormal hemoglobins such as "City of Hope" or "Hb Knossos" or the presence of rare haplotypes in some patients with beta-thalassemia intermedia[20, 21].

REFERENCES

1. Cooley TB, Lee P. Series of cases of splenomegaly in children with anemia and peculiar bone changes. *Trans Am Pediatr Soc* 37:29, 1925.
2. Wintrobe MM. *Hematology A Story of Inspiration and Effort.* Philadelphia: Lea & Febiger, 1985, pp. 105-107, 272, 372, 377, 380.
3. Tavat S. Sur les erythroblastoses a proposes d'une cas de Cooley. *Bull Fac Med Istanbul* 4:2411, 1941.
4. Frank E. Erythroblastose. *Bull Fac Med Istanbul* 4:2174, 1941.
5. Saraçoğlu K. Cooleysche Anämie in der Türkei. *Wien Med Wochenschr* 93:217, 1943.
6. Fakaçelli NM. Erythroblastic anemia of Cooley type. *Türk Tıp Cem Mec* 13:238, 1947.
7. Aksoy M. Zwei Cooley-anämische Geschwister und ihre Eltern mit Thalassemia minor-Symptomen. *Wien Klin Wochenschr* 64:397, 1952.
8. Frank E, Aksüğür H, Başsipani E, et al. De la maladie thalassemia minor. *Istanbul Contrib Clin Sc* 1:332, 1951.
9. Aksoy M. Thalassemia minor with large amount of fetal haemoglobin; report of four cases. *Acta Haematol* (Basel) 22:188, 1959.
10. Aksoy M. Abnormal hemoglobin and thalassemia in Turkey. In Jonxis TH, Delefrasnaye JF (eds). *A Symposium in Abnormal Hemoglobins,* September 8, 1957, Istanbul. Oxford: Blackwell, 1959.
11. Ingram VM, Stretton AO. Genetic basis of thalassemia diseases. *Nature* (London) 184:1903, 1959.
12. Aksoy M. The first observation of homozygous hemoglobin S-alpha thalassemia disease and two types of sickle cell thalassemia disease: (a) sickle cell-alpha thalassemia disease, (b) sickle cell-beta thalassemia disease. *Blood* 22:757, 1963.
13. Aksoy M, Lehmann H. (unpublished data).
14. Çavdar AO, Arcasoy A. The incidence of beta-thalassemia and abnormal hemoglobins in Turkey. *Acta Haematol* (Basel) 45:312, 1971.
15. Kürkçüoğlu M, Dağcı A, Gençelli Y, Arcasoy A. Doğu Anadolu bölgesinde beta-thalassemia ve anormal hemoglobin taraması. *Doğa* 10:3, 1986.
16. Aksoy M, Kutlar A, Kutlar F, et al. Survey on haemoglobin variants. Beta-thalassemia, glucose-6-phosphate dehydrogenase deficiency, and haptoglobin types in Turks from Western Thrace. *J Med Genet* 22:288, 1965.
17. Schwartz E. The silent carrier of beta-thalassemia. *N Engl J Med* 281:1327, 1969.
18. Aksoy M, Erdem Ş. The Thalassemia syndromes. V. Cooley's anaemia with low level of fetal haemoglobin. A genetic study in four families. *Acta Haematol* (Basel) 34:291, 1965.
19. Aksoy M, Dinçol G, Erdem Ş. Different types of beta-thalassemia intermedia. A genetic study in 20 patients. *Acta Haematol* (Basel) 59:178, 1978.
20. Kutlar A, Kutlar F, Aksoy M, et al. Beta-thalassemia intermedia in two Turkish families is caused by the interaction of Hb Knossos [beta 27 (B9) Ala-Ser] and of Hb City of Hope [beta 69 (E13) Gly-ser] with beta degree-thalassemia. *Hemoglobin* 13:7, 1989.
21. Gonzales-Redondo JM, Stoming TA, Kutlar A, et al. A CT substitution of nt-101 in a conserved DNA sequence of the promoter region of the beta-globin gene is associated with "silent" beta-thalassemia. *Blood* 73:1705, 1989.

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NEURAL CELL ADHESION MOLECULES*

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SUMMARY: Banu Anlar. (Department of Pediatrics, Hacettepe University Faculty of Medicine, Ankara, Turkey, and Department of Neurology, University of Chicago, School of Medicine, Chicago, U.S.A). Neural cell adhesion molecules. Turk J Pediatr 33: 199-203, 1991.

Adhesion molecules are expressed on the surface of various cells and establish cell - cell interaction, playing important roles in development, inflammatory reaction, immune response, and tissue regeneration. Neural adhesion molecules, found on neurons or glial surfaces, are involved in the migration of neurons, neurite formation, myelination, and denervation - reinnervation. The roles of cell adhesion molecules in malignancies, normal and abnormal development and as receptors in viral infections, constitute major fields of research and may have important diagnostic and therapeutic implications. *Key words: adhesion molecule, integrin, neural cell adhesion molecule, immunoglobulin superfamily.*

Cell - cell interaction is necessary for the development of multicellular organisms. Embryogenetic and histogenetic events, i.e., the migration of embryonic cells, their localization, and the transfer of information between them is made possible by cellular adhesion and recognition. The discovery of cellular adhesion molecules on the surface of various cells and of extracellular matrix proteins in the tissue matrix, while opening a vast field of research, has allowed scientists to better understand the basic principles of development, the mechanisms of immune response, inflammatory reaction, and tissue regeneration[1].

According to its structure, an adhesion molecule may be considered in one of two major groups: 1) the immunoglobulin superfamily, and, 2) the integrin family[2]. Neural adhesion molecule (NAM)'s, which are found on neurons, glial cells and sometimes in other tissues, generally belong to the first group.

Different NAM's are expressed on the cell surface at different stages of development. Moreover, one cell type may have more than one NAM at one time[1]. While two adhesion molecules may cooperate in some locations, they can antagonize each other in some other tissues. The result (i.e., embryogenesis, inflammatory reaction) depends on the type and the quantity of the molecules that are expressed on each group of cells.

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Most of the studies concerning NAM's are done on animals, especially chicken and rat embryos. The structure of these molecules is evolutionally conserved. The early postnatal cerebellum has been the tissue preferred in the study of NAM's; the main reason is the well known morphology and easy accessibility of this organ, and, particularly, the cell migration that occurs during its development and involves the adhesion molecules. Histologically, proliferating premigratory granule cells are first observed in the outer part of the cerebellar granular layer; then these cells, now postmitotic, associate with the Bergmann glial cell processes, forming parallel fibers in the molecular layer. The granule cell body then migrates to the internal layer along these processes where it takes its final position. The important function adhesion molecules have in this process has provided scientists a good model to examine them.

Before their role in normal and pathological situations, the main characteristics of the NAM's will be considered below.

Neural cell adhesion molecule (N - CAM) : This molecule, expressed on early embryonic cells, has been purified from embryonic and adult chicken brains[3]. It is also present on adult brain neurons, glia, muscle, heart, intestine, kidney and lens. N - CAM establishes cell - cell adhesion by a homophilic mechanism, i.e., the N - CAM molecule on one cell binds to the N - CAM of the other[4]. It is a complicated molecule; it has embryonic and adult forms, the latter differing from the former by the lack of polysialic acid, therefore, by molecular weight, and also, by an increased attaching capacity. The two forms occur at different rates in different brain regions and have a role in neuron - neuron, neuron - glia and glia - glia relations. In the peripheral nervous system, N - CAM is involved in Schwann-axon and Schwann - Schwann interaction.

Neuronal - glial adhesion molecule (Ng - CAM) : This molecule, although structurally and immunologically related to N - CAM (with which it shares at least one antigenic determinant), has distinctive properties[5]. It is called a secondary cellular adhesion molecule because it is expressed on more mature (postmitotic) neurons, especially axons, and Schwann cells. It is involved in neuron - neuron and neuron - glia interaction, particularly in neuronal migration, and has been shown on cerebellar granule cells just before their migration. It is not expressed on glial cells, therefore, the mode of action of Ng - CAM is homophilic for neurons, but heterophilic for neuron - glia interaction. The molecule it binds on, astroglia, is still to be identified. The probable mammalian equivalent of Ng - CAM is nerve growth factor - inducible large external glycoprotein (NILE) which has been shown on the PC12 pheochromocytoma cell line[6].

L1 : This molecule is present on unmyelinated fasciculating axons and Schwann cells and is involved in interaction between these elements. It disappears after the completion of myelination. Antibodies to L1 prevent the neurite formation of

Schwann cells[7]. L1 also has a role in the migration of cerebellar granule cells to the internal granular layer. L1, Ng - CAM and NILE have similar immunological and structural properties[8].

Myelin - associated glycoprotein (MAG) : MAG is found around the axons. It is first detected in the oligodendrocytes, then on their surface during the ensheathing of axons by these cells[9]. It may be involved in the pathogenesis of some polineuropathies. In 40 percent of patients with peripheral neuropathy associated with monoclonal IgM, the IgM reacts with MAG[10].

Adhesion molecule on glia (AMOG) : This is a recently discovered molecule that is involved in neuron - astrocyte adhesion and cerebellar granule cell migration[11].

Some other adhesion molecules in the immunoglobulin superfamily are Po (on peripheral nerves), contactin, carcinoembryonic antigen (CEA), intercellular adhesion molecule - 1 and 2 (ICAM - 1 and 2), CD4 (human immunodeficiency virus receptor), and poliovirus receptor. It is probable that new ones will be added as research in this field is pursued.

Liver - CAM (L - CAM) is the major adhesion molecule of the integrin family. This group differs from the first one in terms of structure and binding mechanism (calcium - dependent vs. calcium - independent). Several other molecules resembling L - CAM have been identified: N - cadherin, A - CAM, P - cadherin, T - cadherin, cell CAM 120/80, ARC - 1, etc. They have common points, such as 50 - 80 % identical aminoacid sequences, are all synthetized as large precursor molecules, have 3 - 4 internal repeats and distinct distribution in adult tissues. As shown in animal studies, N - cadherin is known to play a role in the migration of neural cells.

The role of NAM's in development : Because of the interesting events occurring during its development, namely the migration of granular cells, the cerebellum has been an organ preferred by scientists in order to examine the roles of NAM's. The administration of antibodies to different NAM's in experimental conditions constitutes a valuable tool to prove the role of these molecules during development.

N - CAM is present in the developing cerebellum at all times. Ng - CAM is expressed in granule cells just before their migration and disappears later. Anti - N - CAM given to embryonic chicken does not cause significant impairment of cerebellar development. On the other hand, anti - Ng - CAM arrests the migration of granular cells at the external layer. Antibodies to cytoactin, an extracellular matrix protein, stop these cells at the molecular layer, implicating a role for cytoactin at a later developmental stage[12]. Anti - N - CAM, injected into the retina of chick embryos, inhibits the development of visual pathways. The density and the structure of NAM's change during development. In mice and

chicks the concentration of N - CAM is highest in the perinatal period. The conversion of the embryonic to adult form, which involves a change in molecular weight and structure, occurs at different times in different areas of the brain, implicating a role for morphogenesis[13].

Pathological conditions and NAM's : It is now well-established that N - CAM changes in amount and location in relation to denervation and reinnervation. In normal muscle, N - CAM is found in the interstitium. In peripheral nerve injury, it increases in areas which were synapses before the denervation. When reinnervation is complete, N - CAM returns to the baseline amount. In cases of chronic denervation, it remains at high levels[14]. Similar findings were shown in mixed cultures of neurons and Schwann cells. When neurons are taken off the culture medium, L1 and N - CAM increase on the surface of Schwann cells. This event may be mediated by neural growth factor (NGF) which is known to increase the synthesis of N - CAM in culture. These observations show that while N - CAM's affect the neurons, their location and their interactions with other cells, they also are influenced by them[15].

Failure of the normal conversion of embryonic N - CAM to the adult form has been associated with cerebellar histological abnormalities and clinical ataxia in the staggerer mutants of mice[1].

Another interesting and highly significant observation is the changes of adhesion molecules in malignancies or virally infected cells. Brackenbury et al[16] have shown a decrease in N - CAM expression and increase in motility in retinal cells transformed by rous - sarcoma virus. Some adhesion molecules are found on tumor cells, for instance, human mammary tumor cells express cell CAM 120/80.

One of the important discoveries about adhesion molecules has been their role as viral receptors. It has been established that poliovirus and human immunodeficiency virus use adhesion molecules in order to attach to cells. The most recent report on this subject involves the rhinovirus receptor, ICAM - 1[17].

REFERENCES

1. Edelman GM. Modulation of cell adhesion during induction, histogenesis, and perinatal development of the nervous system. *Annu Rev Neurosci* 7:339, 1984.
2. Kishimoto TK, Larson RS, Corbi AL, et al. The leukocyte integrins. *Adv Immunol* 46:149, 1989.
3. Hoffman S, Sorkin BC, White PC, et al. Chemical characterization of a neural cell adhesion molecule purified from embryonic brain membranes. *J Biol Chem* 257:7720, 1982.
4. Grumet M, Hoffman S, Edelman GM. Two antigenically related neuronal cell adhesion molecules of different specificities mediate neuron-neuron and neuron-glia adhesion. *Proc Natl Acad Sci USA* 81:267, 1984.
5. Grumet M, Hoffman S, Chuong CM, Edelman GM. Polypeptide components and binding functions of neuron-glia cell adhesion molecules. *Proc Natl Acad Sci USA* 81:7989, 1984.

6. Friedlander DR, Grumet M, Edelman GM. Neural growth factor enhances expression of neuron-glia cell adhesion molecule in PC12 cells. *J Cell Biol* 102:413, 1986.
7. Seilheimer B, Persohn E, Schachner M. Antibodies to the Li adhesion molecule inhibit Schwann cell ensheathement of neurons in vitro. *J Cell Biol* 109:3095, 1989.
8. Sajovic P, Ennulat DJ, Shelanski ML, Greene LA. Isolation of NILE glycoprotein-related cDNA probes. *J Neurochem* 49:756, 1987.
9. Bartsch U, Kirchhoff F, Schachner M. Immunohistological localization of the adhesion molecules Li, N-CAM, and MAG in the developing and adult optic nerve of mice. *J Comp Neurol* 284:451, 1989.
10. Tucker GC, Dellagi K, Schmitt C, et al. Heterogenity of human anti-MAG IgM as revealed by their reactivity on avian embryonic tissues. *Clin Exp Immunol* 67:352, 1987.
11. Antonicek H, Persohn E, Schachner M. Biochemical and functional characterization of a novel neuron-glia adhesion molecule that is involved in neuronal migration. *J Cell Biol* 104:1587, 1987.
12. Chuong CM, Crossin KL, Edelman GM. Sequential expression and differential function of multiple adhesion molecules during the formation of cerebellar cortical layers. *J Cell Biol* 104:331, 1987.
13. Chuong CM, Edelman GM. Alterations in neural cell adhesion molecules during development of different regions of the nervous system. *J Neurosci* 4:2354, 1984.
14. Daniloff JK, Levi G, Grumet M, et al. Altered expression of neuronal cell adhesion molecules induced by nerve injury and repair. *J Cell Biol* 103:929, 1986.
15. Seilheimer B, Persohn E, Schachner M. Neural cell adhesion molecule expression is regulated by Schwann cell-neuron interactions in culture. *J Cell Biol* 108:1909, 1989.
16. Brackenbury R, Greenberg ME, Edelman SM. Phenotypic changes and loss of N-CAM-mediated adhesion in transformed embryonic chicken retinal celles. *J Cell Biol* 99:1944, 1984.
17. White JM, Littman DR. Viral receptors of the immunoglobulin superfamily. *Cell* 56:725, 1989.

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