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CONTENTS

Original Articles

Review

TJPD Turkish Journal of Pediatric Disease

Original Article

Clinical and laboratory findings of patients with cystic fibrosis: a single center experience from Türkiye

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ABSTRACT

Objective: The objective of this study was to evaluate the clinical characteristics of patients with cystic fibrosis (CF) and to determine whether there is a relationship between nutritional status and pulmonary functions. Additionally, the study aimed to investigate the relationship between the CFTR genotype and the occurrence of cystic fibrosis-related diabetes (CFRD), as well as the impact of CFTR mutations on the severity of CF pulmonary disease.

Material and Methods: The data of 300 CF patients were retrospectively analyzed. Clinical and laboratory characteristics were obtained from unit database. The patients' growth indices and nutritional status were assessed based on age groups.

Results: Among the 300 patients, 69.5% were diagnosed under age one year old. The earliest diagnosed patient was 2 days old, and the latest diagnosed patient was 31 years old. The most common presenting complaints were recurrent lung infections and gastrointestinal symptoms. Genotyping was performed in 241 patients (80.3%), and 16.6% of these were found to be homozygous for F508del. The allelic frequency of F508del was found to be 41.4%. Eighty-three patients (29.7%) were colonized with *Pseudomonas aeruginosa*, and they were found to have more severe lung disease compared to non-colonized patients (p=0.004). We observed that 30% of the patients with CFRD and 12.7% of the non-diabetic patients had severely impaired pulmonary function (p=0.004). The patients who had F508del mutation in at least one allel were found to have a higher risk of developing diabetes compared to those who did not have (p=0.049).

Conclusion: *Pseudomonas aeruginosa* colonisation and development of CFRD are associated with impairment in pulmonary functions in CF patients.

Keywords: Cystic fibrosis, Mutation, Pulmonary function

INTRODUCTION

Cystic fibrosis (CF) which was once considered as a childhood disease, is now recognized as a condition affecting adults as well, due to better understanding the pathophysiology of the disease, development of reliable diagnostic methods, identification of new mutations and advances in the treatment modalities. CF is an autosomal recessive metabolic disorder characterized by the dysfunction of the cystic fibrosis transmembrane conductance regulator (CFTR) protein. This condition primarily affects the sweat glands, pancreas, and the mucous glands of the respiratory, gastrointestinal, and genital tracts. As a result, individuals with CF often experience chronic pulmonary infections and inflammation, pancreatic exocrine insufficiency, and male infertility (1).

In a recent study from our country the incidence of CF was found to be 2.9 per 10000 live births in Central Anatolia (2). However, since the rate of consanguineous marriage is high in our country, the prevalance of the disease is estimated to be much higher than expected.

In this study, we aimed to evaluate the clinical characterictics and laboratory findings at the time of the diagnosis and at the time of the last follow-up in patients with CF. The nutritional classifications were compared with FEV1 and *P. aeruginosa* colonization across different age groups. Additionally, we

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aimed to identify relationship between CFTR genotype and the occurrence of cystic fibrosis-related diabetes (CFRD), and the impact of CFTR mutations on the severity of CF pulmonary disease.

MATERIALS and METHODS

In this retrospective study, data of 300 CF patients were analyzed between January 2011 and June 2012 at pediatric pulmonology unit of Hacettepe University. Among 300 patients, 278 of them had clinical visit during this period because two patients died and 20 did not apply for follow-up visits.

The patients who fulfilled the following diagnostic criteria of CF were included in the study: (a) two sweat tests greater than 60 mmol/L chloride, and (b) one sweat test greater than 60 mmol/L chloride and DNA analysis revealing two identified disease causing CF mutations. If the sweat test result is less than or equal to 60 mmol/L: (a) DNA analysis revealing two identified disease causing CF mutations, and (b) clinical presentation consistent with typical features of CF (3).

We evaluated clinical and laboratory findings of patients from the unit databases including; gender, age, complaints, symptoms, physical examination findings, height and weight measurements, presence of pancreatic insufficiency, results of genotyping, sweat chloride test, pulmonary function test. Also microbiological evaluation of the respiratory cultures, findings of the chest X-rays, abdominal ultrasonography and if available computed tomography were recorded. Patients were evaluated for the development of CF related complications.

Sweat chloride concentration was measured via the quantitative pilocarpine iontophoresis test, as described by Gibson and Cooke method (4). Genetic mutation analysis was performed using a CF gene panel that included 36 CFTR gene mutations. Pancreatic insufficiency was defined as having the classic symptoms and signs including the weight loss, gas, dyspepsia, bloating, foul-smelling oily stools and steatorrhea. Although fecal elastase is the most used test to screen pancreatic exocrine insufficiency, during the time of the study this test could not be analyzed in our hospital (5). Anemia was defined as a reduction of the hemoglobin concentration or red blood cell volume below the range of values according to age and sex (6). Elevation of liver transaminases was categorized depending on age and sex (7).

The nutritional status of the patients were evaluated according to the age groups. Weight, height measurements and body mass index (BMI) were expressed in percentiles by using reference values issued by Centers for Disease Control (8). Weight-for-height (WfH) percentiles were recorded for patients under two years of age, while BMI percentiles were noted for those aged 2 to 18 years, and BMI measurements were taken for patients over 18 years old. For the patients under two years of age, WfH percentiles under 10 was defined as inadequate nutrition, percentile between 10 and 25 as nutrition at risk, percentile between 25 and 75 as normal nutrition, and percentile over 95 was defined as obese. For the ages between 2 and 18 years of age, BMI percentiles under 10 was defined as inadequate nutrition, percentile between 10 and 25 as nutrition at risk, percentile between 25 and 75 as normal nutrition, and percentile over 95 was defined as obese. For the patients over age 18, BMI under 19 was defined as inadequate nutrition for boys and girls, BMI between 19 and 22 for girls and 19 and 23 for boys as nutrition at risk, BMI over 22 for girls and 23 for boys were defined as normal nutrition.

Pulmonary function tests were conducted using spirometry in accordance with the standard guidelines by the American Thoracic Society (ATS) and the European Respiratory Society (ERS). The Forced Expiratory Volume in the first second (FEV1) was measured in liters and expressed as a percentage (%) of the predicted value, based on reference data from healthy individuals of the same age, sex, height, and racial/ethnic background (9).

Chronic colonization by *Pseudomonas aeruginosa* or *Staphylococcus aureus* was defined as the presence of *Pseudomonas aeruginosa* or *Staphylococcus aureus* in respiratory cultures for at least 6 months, based on at least three positive cultures with at least one month intervals between them (10).

Patients were evaluated for the development of CF related complications. Pseudo_Bartter syndrome was defined as acute exacerbation with hyponatremia, hypochloremia, hypokalemia and metabolic alkalosis (11). The occurence of Allergic Bronchopulmonary Aspergillosis (ABPA) was recorded; which is a pulmonary hypersensitivity disease mediated by an allergic response due to *Aspergillus fumigatus* (12). For the diagnosis of diabetes mellitus, patients with a fasting plasma glucose ≥126 mg/dl required either a confirmatory fasting plasma glucose obtained the next day or a casual blood glucose level measured. If the repeated fasting plasma glucose was ≥126 mg/dl or if the casual glucose was ≥200 mg/dl, CFRD was diagnosed. To rule out diabetes mellitus in individuals with fasting plasma glucose <126 mg/dl and with clinical symptoms of diabetes, a standard oral glucose tolerance test (OGTT) was performed. Impaired glucose tolerance was defined as 2-hour glucose levels of 140 to 199 mg/dL on the OGTT (13).

Statistical analysis

Statistical analyses were performed using the SPSS Statistics for Windows, version 15.0 (SPSS Inc., Chicago, Ill., USA). The variables were investigated using visual (histogram) and analytical (Kolmogorov–Smirnov/ Shapiro–Wilk test) methods to determine whether or not they are normally distributed. Descriptive analyses were presented using medians, interquartile range (IQR), minimum, and maximum for the nonnormally distributed and ordinal variables, means and standard deviations for normally distributed variables. The categorical data were analysed as frequency and percentage. Chi-square test was used to compare proportions in different

groups. Mann-Whitney U and Kruskal Wallis tests were used to compare nonnormally distributed parameters. A p value of less than 0.050 was considered to show a statistically significant result.

RESULTS

A total of 300 CF patients were included in the study. Among all patients 50.3% of them were male and the rate of consanguineous marriage was 39.7%.

Clinical characteristics and laboratory findings of the patients at the time of the diagnosis

Among 300 patients, 69.5% of them were diagnosed under age one year. The earliest diagnosed patient was 2 days old, and the latest diagnosed patient was 31 years old. The median age at the diagnosis was 5 (IQR, 3-29) months. Ninety-nine of the patients were diagnosed in our hospital, 201 were referred to us with the suspected diagnosis of CF. Forty-three (14.3%) of our patients had siblings with CF.

The common presenting symptoms were recurrent lung infection, diarrhea, vomiting and Pseudo-Bartter syndrome (Table I). The evaluation of the patients' nutritional status revealed that among those under 2 years of age, 71% had a WfH measurement below the 10th percentile, 2.7% were between the 10th and 24th percentiles, and 26.3% were above the 25th percentile. In patients aged between 2 and 18 years, 45.7% were below the 10th percentile, 11.9% were between the 10th and 24th percentiles, 40.7% were above the 25th percentile, and 1.7% were classified as obese. Among patients over 18 years of age, 5 had a BMI lower than 19, while 2 had a normal BMI.

Most of the patients had anemia (40.9%) and elevated liver transaminases (40.9%). The other abnormal laboratory findings were vitamin A,E,D deficiency, metabolic alkalosis, hypoalbuminemia, electrolyte imbalance, elevated immunglobulin E, elevated hemoglobin a1c, hyperbilirubinemia and hyperglycemia. Pancreatic insufficiency was diagnosed in 94.7% of the patients. Sweat tests were performed in 287 patients. The mean sweat chloride level was 99.5±24.44 mmol/l.

Thirty-seven patients performed spirometry, the median FEV1% was 80 (IQR, 60-88). Thirty-six percent of the patients had normal chest radiograph. The most common pathological findings, in decreasing order, were bilateral hyperinflation of the lungs, infiltration, chronic changes, bronchiectasis, and atelectasis. Within one year of the diagnosis, chest tomograpy was performed to 19.7% of the patients and bronchiectasis and peribronchial thickening were the most common findings. Abdominal ultrasonography was normal in 51.5% of the patients, while hepatomegaly was detected in 18.7%.

At the time of the diagnosis *Pseudomonas aeruginosa* (n=52) and *Staphylococcus aureus* (n=54) were the most frequently isolated microorganisms in the respiratory cultures. No microorganism was isolated in 59% of the patients.

Table I: The presenting symptoms and the findings of the patients

Symptoms	Values*
Recurrent lung infection	117 (39)
Diarrhea /vomiting	95 (31.7)
Pseudo-Bartter syndrome	55 (18.3)
Failure to thrive	51 (17)
Chronic cough	44 (14.7)
Steatorrhea	35 (11.7)
Siblings of patients with CF	21 (7)
Meconium ileus	20 (6.7)
Anemia	15 (5)
Saltytaste on skin	13 (4.3)
Pretibial edema	8 (2.7)
Elevated liver transaminases	7 (2.3)
Constipation	5 (1.7)
Prolonged jaundice	5 (1.7)
Rash	4 (1.3)
Hemoptysis	3 (1)
Rectal prolapse	3 (1)
Atelectasis	2 (0.7)
Acute pancreatitis	1 (0.3)

**: n (%)*

Table II: Allelic frequencies of the 14 most common CFTR mutations

Mutation name	Allelic frequencies*
F508del	133 (41.4)
G85E	33 (10.2)
1677delTA	23 (7.1)
2789+5G-A	22 (6.8)
N1303K	18 (5.6)
2183AA-G	13 (4.0)
G542X	11 (3.4)
R334W	9 (2.8)
W1282X	9 (2.8)
CFTRdele2,3	7 (2.1)
3120+1G-A	6 (1.8)
3849+10kbC-T	6 (1.8)
621+1G-T	4 (1.2)
R347P	4 (1.2)

**: n(%)*

Genotyping was performed in 241 (80.3%) patients. Two mutations were identified in 171 (70.9%) patients and no mutations could be found in 48 (19.9%) patients. The prevalence of the patients who were F508del homozygous was 16.6% and F508del heterozygous was 22%. Among 315 allels where a mutation was detected, the most common mutation

was F508del. The other most common mutations were G85E, 1677delTA,2789+5G-A and N1303K in decreasing frequency (Table II).

Clinical characteristics and laboratory findings at the time of inclusion

Among 300 patients, 278 of them had clinical visit during this period because two patients died and 20 did not apply for follow-up visits. The median age was 7 years and 6 months (min 3 months of age, max 39 years). Sixty-seven percent of the patients had normal physical examination findings, while the most common pathological findings were crepitant rales and rhonchi.

The evaluation of the patients' nutritional status at the time of inclusion revealed that among those under 2 years of age, 9.4% were below the 10th percentile, 9.4% were between the 10th and 24th percentiles, and 81.2% were above the 25th percentile. In patients aged 2 to 18 years, 27.7% were below the 10th percentile, 11% were between the 10th and 24th percentiles, 54.8% were above the 25th percentile, and 6.5% were classified as obese. Among patients over 18 years of age, 19 had a BMI lower than 19, while 24 had a normal BMI. The mean BMI for boys and girls over 18 years was 22.2±4.59 and 21±2.82, respectively.

At the time of inclusion 128 patients performed spirometry, the median FEV1% was 76 (IQR, 59-90). Eight of the patients had FEV1 below 30%. The most common pathologic findings of chest radiograph were bronchiectasis, atelectasis and chronic changes. Chest tomography was performed in 32 patients, the most frequent pathologic findings were similar to those found in chest radiograph. Abdominal ultrasonography was normal in 43.9% of the patients, and hepatomegaly was detected in 17.7%.

P.aeruginosa (n=113) and *S. aureus* (n=136) were the most frequently isolated microorganisms in the respiratory cultures at the time of the inclusion consistent with the results at the time of diagnosis. Eighty three (29.7%) patients were colonised with *P. aeruginosa* and nine (3%) were colonised with *S. aureus*.

We observed that up to 35% of the patients developed gastrointestinal and respiratory system complications. The most common respiratory system complication was bronchiectasis, followed by ABPA, asthma, hemoptysis, atelectasis, and pneumothorax. The most frequent gastrointestinal system complications were hepatomegaly, hepatosteatosis, elevated liver transaminases, biliary disease, cirrhosis, splenomegaly and pancreatic steatosis. Endocrin complications occured in 6.7% of the patients. Ten patients had CFRD and 15 patients had impaired glucose tolerance.

The relationship between nutritional status and pulmonary function was evaluated; no statistically significant difference in FEV1% was found based on the nutritional status of patients under and over 18 years of age (p=0.170 and p=0.810, respectively). Figure 1 illustrates the relationship between nutritional status and FEV1% in patients under 18 years old. Figure 2 shows the relationship between the nutritional status and FEV1% in patients over 18 years old.

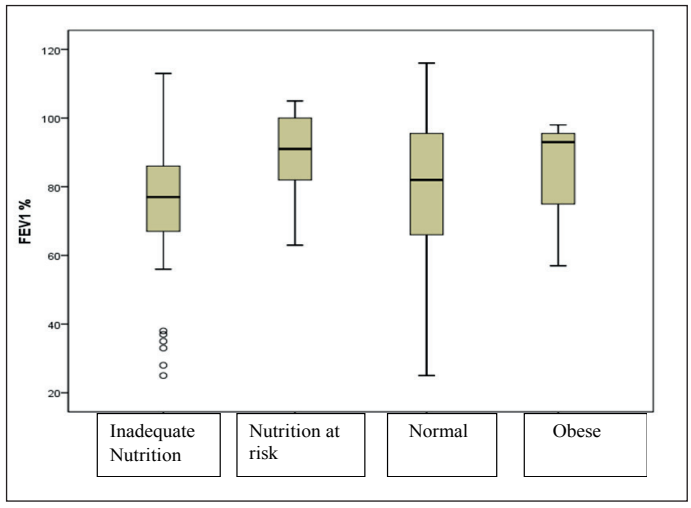

Figure 1: The relationship between the nutritional status and FEV1% of the patients. under age 18 years old.

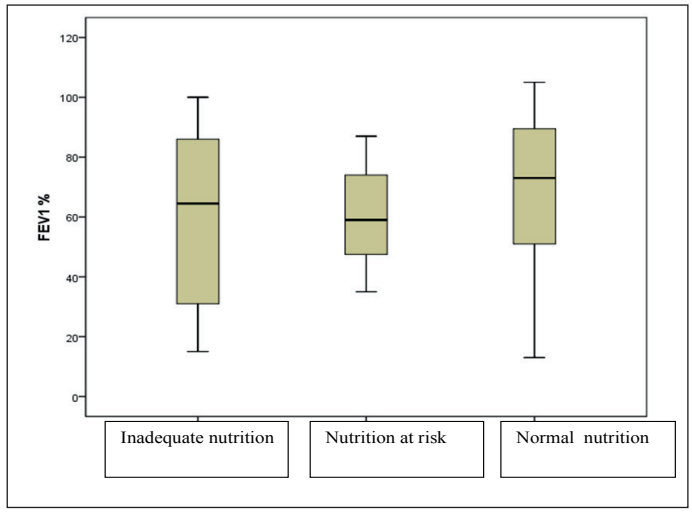

Figure 2: The relationship between the nutritional status and FEV1% of the patients over age 18 years old

The FEV1% of the patients were compared according to CFTR mutations and no statistically significant difference was found (p=0.250) (Figure 3). Twenty percent of the patients who were F508del homozygous had severely impaired pulmonary function. 5.9% of the patients who were F508del heterozygous and 9.1% of patients with F508del and unknown mutation had severely impaired pulmonary function. There was no statistically significant relationship between having F508del mutation and severity of impaired pulmonary function (p=0.360). Patients with *P. aeruginosa* colonization exhibited more severely impaired pulmonary function (p=0.004). The median FEV1% of the *P. aeruginosa* colonized patients was found to be statistically lower than non-colonized group (p=0.001).

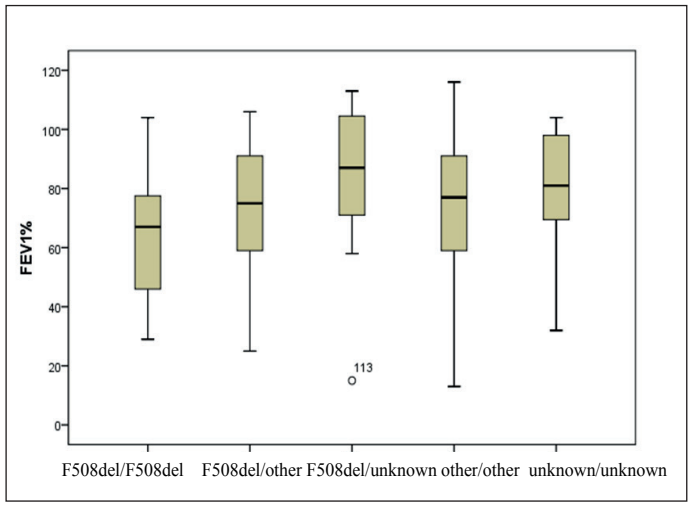

Figure 3: The relationship between FEV1% and type of the CFTR mutations

The relationship between nutritional status and *P. aeruginosa* colonization was evaluated according to the age groups and no statistically significant difference was found between the colonized and non-colonized groups (p=0.670, p=0.720 and p=0.980, respectively). There was no statistically significant difference in *P.aeruginosa* colonization between those with and without F508del mutation (p=0.440). Also no statistically significant difference was found in the severity of the lung disease according to median FEV1% in patients with and without MRSA in the respiratory cultures taken at the time of the inclusion (p=0.300).

We observed that 30% of the patients with CFRD and 12.7% of the non-diabetic patients had severely impaired pulmonary function according to FEV1% values (p=0.004). The patients who had F508del mutation in at least one allel were found to have higher rate of developing diabetes compared to those who did not have (p=0.049).

DISCUSSION

In this study clinical and laboratory findings of 300 patients were examined in our unit which is the largest CF center in Türkiye. Forty-three (14.3%) of our patients had siblings with CF. Our results were consistent with the Cystic Fibrosis Foundation annual report of 2023 that 13.9% of their patients had a family history of CF (14). Recently, a multicenter study from our country which evaluated 1170 CF patients, revealed that the median age at diagnosis was 1.7 years and 240 patients were diagnosed through newborn screening (15). In our study we found that most of our patients (69.5%) were diagnosed within the first year of life and median age at diagnosis was 5 months. According to the data of Cystic Fibrosis Foundation annual report of 2018 and European Cystic Fibrosis Society Patient Registry (ECFSPR) annual report of 2017, median age at the diagnosis was 3 and 4 months, respectively (16,17). It is a pleasure to see that most of the patients in our country are diagnosed at an early age.

Cystic Fibrosis Foundation reported that 54.1% of all new diagnoses and 85.1% of diagnoses among those less than 6 months old were as detected by newborn screening in 2023 (14). Doğru et al. (15) showed that among 293 patients who were under 3 years of age, 81.9% of them were diagnosed via newborn screening. At the time of our study newborn screening for CF was not implemented in our country's national screening programme. Thus our patients were diagnosed upon the presentation of symptoms. We anticipate that the implementation of newborn screening for CF in our country will facilitate earlier diagnoses, leading to improved survival outcomes for patients.

Malnutrition in CF patients is associated with deterioration of pulmonary function; as a result it is essential to assess nutritional status in all CF patients regularly (18). Cystic Fibrosis Foundation recommends that CF patients growth percentile should be within the 50th. percentile according to their age group. When BMI was compared with FEV1%, there was a correlation between improved BMI percentiles and pulmonary function in individuals aged 6 to 20 years and adults (16). Kilinc et al. (18) grouped 143 CF patients according to nutritional status and compared them in terms of pulmonary function test results, lung infections, and the hospitalization rate. Patients in the well-nourished group had significantly higher pulmonary function test results and bacterial lung infections differed significantly between groups.

Ashkenazi et al. (19) investigated the long-term correlations between nutritional status at 10 years of age and pulmonary function as well as the severity of lung disease in adulthood. Their findings indicated that a BMI z-score of less than -0.75 at age 10 was associated with a higher rate of lung transplantation in adults (19). In our study we could not find any association between the nutritional status and pulmonary function. This may be due to factors that may affect pulmonary function tests like age at diagnosis, type of the mutations, sociocultural level of family, treatment compliance and *P. aeruginosa* colonisation.

Genotyping was performed in 80.3% of our patients and 16.6% of them were F508del homozygous and allelic frequency of F508del was 41.4%. Doğru et al. (15), found that in their registry, 8.8% of the patients were F508del homozygous and 12.9% were F508del heterozygous and the allellic frequency of this mutation was 28%. In United States and Europe most of the patients had identified mutations, however, in our study no CFTR mutations were detected nearly in 20% of our patients, in consistence with the results of a recent study in our country (15-17). This can be due to the genetic heterogenity because of the high prevelance of consanguineous marriage in our country and the limited number of the mutations investigated with CF gene panel during the study period. As the CFTR modulators have been developed for specific gene mutations, identification

of the CFTR genotypes is of crucial importance to determine the patients eligibility for these drugs (20).

P. aeruginosa colonization has been shown to be associated with impaired pulmonary function and a major predictor of morbidity and mortality during both children and adults (21). According to the national CF registry in Türkiye, the prevalence of *P. aeruginosa* colonization was found to be 20.9% (15). *P. aeruginosa* was colonised in 29.7% of our patients and they had more severely impaired pulmonary function than non-colonized group. Mésinèle et al. (10), found that the decline in pulmonary function varied with *P. aeruginosa* status. The mean annual decrease in FEV1% was 0.38% per year before the initial acquisition of *P. aeruginosa*, which increased to 0.93% after its initial acquisition, and reached 1.50% per year in patients with chronic *P. aeruginosa* colonization (10). Since chronic inflammation and progressive lung injury are the major causes of morbidity and mortality in these patients, eradicating *P.aeruginosa* from the respiratory tract before the occurence of irreversible lung injury is crucial.

Life expectancy increases in CF patients in the last 60 years. However, this improvement in survival has led to patients experiencing complications in addition to lung disease and impaired nutrition. The most common of these is CFRD, which affects 40–50% of CF adults. Female sex, advanced age, reduced lung function, liver disease, steroid treatment, a family history of type 2 diabetes, and genetic factors, such as mutations in the CFTR gene and other modifier genes are known risk factors for CFRD (22). Genotypes that lead to severe CFTR dysfunction and pancreatic exocrine insufficiency are associated with an increased risk of developing diabetes over time. Individuals with severe CFTR mutations, such as F508del homozygotes, have had a risk of developing diabetes that exceeds 80% by age 50 and approaches 100% by age 60 (23). Consistent with the literature, in our study patients who had F508del mutation in at least one allel were found to have a higher risk of developing diabetes compared to those who did not have (p=0.049). It is shown that in the years preceding therapy for CFRD, reductions in pulmonary functions and BMI are observed (24). In our study FEV1% of patients with CFRD were compared with the patients who had no diabetes and it was found that 30% of the patients with CFRD and 12.7% of the non-diabetic patients had severely impaired pulmonary function (p=0.004). According the data from the adult CF patients from Türkiye, lower BMI and lower FEV1% were observed in CFRD group (21). These results indicate that CFRD adds to difficulties in maintaining nutritional status and pulmonary function in CF patients. Therefore annual screening with an OGTT is recommended for patients starting at age ten (25).

The major limitation of our study is its retrospective nature and the fact that it was conducted in the single center. Although genotyping performed in most of the patients, we were unable to identify the CFTR mutation in approximately one-fifth of the patients due to limited number of mutations screened during the study period.

In conclusion, CF is a significant disease that affects not only children but increasingly impacts adults as well. The study findings highlight the importance of CFRD that is associated with decline in pulmonary functions and minimizing *P. aeruginosa* colonization in CF patients.

Ethics committee approval

This study was conducted in accordance with the Helsinki Declaration Principles. This study was approved by the local ethics committee (Hacettepe University Ethics Board, date: 17.04.2007, reference number: HEK 07/16-21).

Contribution of the authors

Köse Çetinkaya A: Constructing the hypothesis or idea of research and/or article, Planning methodology to reach the conclusions, Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments, Taking responsibility in logical interpretation and conclusion of the results, Taking responsibility in necessary literature review for the study, Taking responsibility in the writing of the whole or important parts of the study, Reviewing the article before submission scientifically besides spelling and grammar. ***Doğru D:*** Constructing the hypothesis or idea of research and/or article, Planning methodology to reach the conclusions, Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments, Taking responsibility in logical interpretation and conclusion of the results, Taking responsibility in the writing of the whole or important parts of the study, Reviewing the article before submission scientifically besides spelling and grammar. ***Cinel G:*** Planning methodology to reach the conclusions, Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments, Taking responsibility in logical interpretation and conclusion of the results, Reviewing the article before submission scientifically besides spelling and grammar. ***Yalçın E:*** Planning methodology to reach the conclusions,Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments, Taking responsibility in logical interpretation and conclusion of the results, Reviewing the article before submission scientifically besides spelling and grammar. ***Özçelik U:*** Planning methodology to reach the conclusions, Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments, Reviewing the article before submission scientifically besides spelling and grammar. ***Kiper N:*** Planning methodology to reach the conclusions, Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments, Reviewing the article before submission scientifically besides spelling and grammar. ***Özen H:*** Planning methodology to reach the conclusions, Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments, Taking responsibility in logical interpretation and conclusion of the results, Reviewing the article before submission scientifically besides spelling and grammar. ***Alikaşifoğlu A:*** Planning methodology to reach the conclusions, Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments, Taking responsibility in

logical interpretation and conclusion of the results, Reviewing the article before submission scientifically besides spelling and grammar. ***Şener B:*** Planning methodology to reach the conclusions, Taking responsibility in logical interpretation and conclusion of the results, Reviewing the article before submission scientifically besides spelling and grammar. ***Dayangaç Erden D:*** Planning methodology to reach the conclusions, Taking responsibility in logical interpretation and conclusion of the results, Reviewing the article before submission scientifically besides spelling and grammar.

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Conflict of interest

The authors declare that there is no conflict of interest.

REFERENCES

1. Shteinberg M, Haq IJ, Polineni D, Davies JC. Cystic fibrosis. Lancet 2021;397:2195-211.https://doi.org/10.1016/S0140-6736(20)32542-3
2. Hangül M, Pekcan S, Köse M, Acıcan D, Şahlar TE, Erdoğan M, et al. The Incidence of Cystic Fibrosis in the Central Region of Anatolia in Türkiye Between 2015 and 2016. Balkan Med J 2019;36:179-83.https://doi.org/10.4274/balkanmedj.galenos.2018.2018.1332
3. ECFS Patient Registry. https://www.ecfs.eu/ecfspr/. Accessed August 14, 2019.
4. Schmidt H, Sharma G. Sweat testing. In: StatPearls. Treasure Island, FL: StatPearls Publishing, 2020.
5. Singh VK, Schwarzenberg SJ. Pancreatic insufficiency in Cystic Fibrosis. J Cyst Fibros 2017;16 Suppl 2:S70-8. https://doi.org/10.1016/j.jcf.2017.06.011
6. Wang M. Iron Deficiency and Other Types of Anemia in Infants and Children. Am Fam Physician 2016;93:270-8.
7. Zhu X, Wang K, Zhou Q, Xu J. Establishment of age- and sex-specific reference intervals for serum liver function tests in pediatric population aged 1-<18 years: A prospective study. J Clin Lab Anal 2021;35:e23708. https://doi.org/10.1002/jcla.23708
8. Centers for Disease Control and Prevention. A SAS program for the 2000 CDC growth charts (ages 0 to <20 years) http://www.cdc.gov/nccdphp/dnpao/growthcharts/resources/sas.htm/
9. Miller MR, Hankinson J, Brusasco V, Burgos F, Casaburi R, Coates A, et al. ATS/ERS Task Force. Standardisation of spirometry. Eur Respir J 2005;26:319-38. https://doi.org/10.1183/09031936.05.00034805
10. Mésinèle J, Ruffin M, Kemgang A, Guillot L, Boëlle PY, Corvol H. Risk factors for Pseudomonas aeruginosa airway infection and lung function decline in children with cystic fibrosis. J Cyst Fibros 2022;21:45-51. https://doi.org/10.1016/j.jcf.2021.09.017
11. Abdul Aziz D, Siddiqui F, Abbasi Q, Iftikhar H, Shahid S, Mir F. Characteristics of electrolyte imbalance and pseudo-bartter syndrome in hospitalized cystic fibrosis children and adolescents. J Cyst Fibros 2022;21:514-8. https://doi.org/10.1016/j.jcf.2021.09.013
12. Francis NZ, Southern KW. Antifungal therapies for allergic bronchopulmonary aspergillosis in people with cystic fibrosis. Cochrane Database Syst Rev 2022;9:CD002204. https://doi.org/10.1002/14651858.CD002204.pub5
13. Moran A, Brunzell C, Cohen RC, Katz M, Marshall BC, Onady G, et al. CFRD Guidelines Committee. Clinical care guidelines for cystic fibrosis-related diabetes: a position statement of the American Diabetes Association and a clinical practice guideline of the Cystic Fibrosis Foundation, endorsed by the Pediatric Endocrine Society. Diabetes Care 2010;33:2697-708. https://doi.org/10.2337/dc10-1768
14. Cystic Fibrosis Foundation Patient Registry 2023 Annual Data Report. https://www.cff.org/medical-professionals/patient-registry
15. Dogru D, Çakır E, Şişmanlar T, Çobanoğlu N, Pekcan S, Cinel G, et al. Cystic fibrosis in Türkiye: First data from the national registry. Pediatr Pulmonol 2020;55:541-8. https://doi.org/10.1002/ppul.24561
16. Cystic Fibrosis Foundation Patient Registry 2018 Annual Data Report. https://www.cff.org/sites/default/files/2021-10/2018-Annual-Report.pdf
17. Zolin A, Orenti A,Naehrlic L, van Rens J, Fox A, Jung A, et al. ECFS Patient Registry Annual Data Report data 2017. Denmark
18. Kilinc AA, Beser OF, Ugur EP, Cokugras FC, Cokugras H. The effects of nutritional status and intervention on pulmonary functions in pediatric cystic fibrosis patients. Pediatr Int 2021;63:316-22. https://doi.org/10.1111/ped.14417
19. Ashkenazi M, Nathan N, Sarouk I, Aluma BEB, Dagan A, Bezalel Y, et al. Nutritional status in childhood as a prognostic factor in patients with cystic fibrosis. Hai 2019;197:371-6. https://doi.org/10.1007/s00408-019-00218-3
20. Çobanoğlu N, Özçelik U, Çakır E, Şişmanlar Eyüboğlu T, Pekcan S, Cinel G, et al. Patients eligible for modulator drugs: Data from cystic fibrosis registry of Türkiye. Pediatr Pulmonol 2020;55:2302-6. https://doi.org/10.1002/ppul.24854
21. Er B, Çelebioğlu E, Yalçin E, Doğru D, Erden Aki Ö, Uzun Ö, et al.Factors associated with severe lung disease in an adult population with cystic fibrosis: a single-center experience. Turk J Med Sci 2020;50:945-52. https://doi.org/10.3906/sag-1912-101
22. Iafusco F, Maione G, Rosanio FM, Mozzillo E, Franzese A, Tinto N. Cystic Fibrosis-Related Diabetes (CFRD): Overview of Associated Genetic Factors. Diagnostics (Basel) 2021;11:572. https://doi.org/10.3390/diagnostics11030572
23. Lewis C, Blackman SM, Nelson A, Oberdorfer E, Wells D, Dunitz J, et al. Diabetes-related mortality in adults with cystic fibrosis. Role of genotype and sex. Am J Respir Crit Care Med 2015;191:194-200. https://doi.org/10.1164/rccm.201403-0576OC
24. Kayani K, Mohammed R, Mohiaddin H. Cystic Fibrosis-Related Diabetes. Front Endocrinol (Lausanne) 2018;9:20. https://doi.org/10.3389/fendo.2018.00020
25. Castellani C, Duff AJA, Bell SC, Heijerman HGM, Munck A, Ratjen F, et al. ECFS best practice guidelines: the 2018 revision. J Cyst Fibros 2018;17:153-78. https://doi.org/10.1016/j.jcf.2018.02.006

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Original Article

Port-catheter complications in children with malignancy

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ABSTRACT

Objective: Central venous access catheters (CVC) are crucial for chronically ill patients, especially in pediatric cancer patients. The aim of this study was to determine subcutaneous implanted port-catheter-related early and late complications and outcomes of catheters in children with malignancy.

Material and Methods: This retrospective study evaluated complications related to subcutaneous implanted port catheters in children with malignancies who were hospitalized in the Pediatric Hematology and Oncology clinics.

Results: The mean age of 69 patients (M/F,37/32) at diagnosis were 6.4±4.85 years (6 months-17 years). During the study period, 89 port catheters were inserted and 141 complications were detected in 54 (60%) of 89 port catheters in a total of 19226 catheter days. Infectious, thrombotic, and mechanical complications were noted in 98 (69.5%), 29 (20.5%), and 14 (10%) port catheters, respectively. Six different complications were identified in one port catheter, while seven complications were found in three different port catheters of a patient with acute myeloid leukemia (AML). The patients who had severe neutropenia (neutrophil count $<0.5x10^9$) on the day of insertion showed more complications than non-neutropenic patients (63.6%, p <0.001). Fifty-seven early (40.4%) and 84 late (59.6%) complications were noted. The most catheterized vein was the right external jugular vein (n=45), with 32 of these cases resulting in complications. The complication rate for the catheters in right external jugular vein was significantly higher than the others (p= 0.024). Infectious complications were most prevalent both in the early and in the late periods (p<0.001). Gram-positive bacteria, gram-negative bacteria and fungi were identified in 61.6%, 34.9%, and 3.5% catheter cultures, respectively. An antibiotic lock therapy with systemic antibiotics was used in 20 infection episodes; and the antibiotic lock failed only in two infection episodes.

Conclusion: Our study highlighted a high rate of complication-related port catheter removal, with skin flora infections. The choice of vein for insertion and the positioning of the port-catheter tip are also key factors contributing to complications. Ensuring proper implantation, usage, maintenance is essential to minimize both early and late complications.

Keywords: Children, Complication, Malignancy, Port catheter

INTRODUCTION

Central venous access catheters (CVCs) are essential for chronically ill patients, particularly pediatric cancer patients, as they provide a long-term route for administering medications, blood products, nutritional supplements, and fluids. These catheters are typically inserted into a large vessel, with the tip extending to the heart or major vessels like the superior vena cava (1). Implantable port catheters are often the most suitable choice for cancer patients due to their lower risk of infection and extravasation (2). However, both the nature of the malignancy and the irritative effects of vesicant antineoplastic drugs can increase the risk of catheter-related complications (3). Early complications, which occur within 30 days of central line insertion, primarily include cardiac, vascular, pulmonary, and catheter placement issues. Late complications are mainly associated with infections and device dysfunction (3, 4).

This study aimed to determine the early and late complications related to subcutaneous implanted port catheters, as well as the outcomes of catheter use, in children with malignancies hospitalized in pediatric hematology and oncology clinics.

MATERIALS and METHODS

This retrospective study evaluated complications related to subcutaneous implanted port catheters in 69 children with

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malignancies who were hospitalized in the Pediatric Hematology and Oncology clinics of Ankara Children's Health and Diseases Hematology and Oncology Training and Research Hospital between January 2014 and June 2015. The patient's age, diagnosis, port-catheter insertion date, the vein in the catheter placed, the position of the tip of the catheter, neutrophil count and presence of fever at the time of port insertion, port catheter, and peripheral blood culture results and if present, the treatment characteristics with antibiotics or antibiotic lock therapy were recorded. Port-catheter complications of patients who underwent hematopoietic stem cell transplantation were not included. If a port catheter was removed and a new one was inserted in a subsequent session, the new port was considered a distinct catheter.

Port catheters were inserted by pediatric surgeons under general anesthesia in the operating room of our hospital, 7-10 days after the diagnosis of leukemia or solid tumors. Different brands and sizes were used based on the patient's age and weight. The catheters were inserted into the vein deemed appropriate by the surgeon, without the use of ultrasound guidance. After obtaining a blood culture from the catheter, heparinized saline (100 international units/ml, 2-3 ml) was administered, and the catheter was used the following day. The catheter tip was verified using chest radiographs and echocardiography for all patients.

Port catheter complications were classified as early or late. Early complications were defined as those occurring within the first 30 days after catheter insertion, while late complications were those that developed after this period. Both early and late complications were further examined in three categories.

Mechanical complications included fractures, migration, catheter kinking, leakage, extravasation, reservoir rotation, catheter failure, pneumothorax, and bleeding. Thrombotic complications were classified as partial catheter lumen occlusion (suspected when there is an inability to aspirate blood but the ability to infuse) or complete venous thrombus (suspected when there is an inability to both aspirate and infuse), confirmed by Doppler ultrasound or echocardiogram.

Infectious complications were defined as catheter-related local infections (indicated by symptoms such as tenderness, erythema, induration, and purulent discharge at the catheter reservoir), microbiologically documented catheter infections (where a bacterium was identified in the port-catheter culture but not in peripheral blood), or catheter-related bloodstream infections (where the same microorganism was detected in both catheter and peripheral blood cultures in cases with clinical signs of suspected infection). Coagulase-negative Staphylococci (CoNS) and other skin contaminants were considered infection-related agents if two blood cultures were positive, or if one culture was positive along with clinical signs of active infection.

Statistical analysis

Statistical analysis was performed using SPSS Statistics for Windows, version 17.0 (SPSS Inc., Chicago, Ill., USA). Descriptive statistics were presented as numbers and percentages for categorical variables, and as mean values with standard deviations for numerical variables. Chi-square and Fisher's Exact tests were employed to compare categorical variables. A significance level of $p<0.050$ was considered statistically significant.

RESULTS

In this study, 89 port catheters of 69 patients were analyzed. The mean age of the patients were 6.4±4.85 years (6 months- 17 years) (Figure 1). Male female ratio was 37/32. Port catheters were mostly used in patients with hematologic malignancies (n=60, 86.9%), followed by (n=9, 13.1%) patients with solid tumors. Forty-six of these patients (66.6%) were diagnosed with acute lymphoblastic leukemia (ALL), 14 (20.2%) had acute myeloid leukemia (AML) and nine patients had different oncologic malignancies (Neuroblastoma (n=4), rhabdomyosarcoma (n=2), Wilms tumor (n=1), Ewing sarcoma (n=1), and desmoplastic small round cell tumor (DSRCT) (n = 1).

A total of 141 complications were detected in 54 (60%) of the 89 port catheters during the study period, which included an analysis of 19 226 catheter days. The time between port insertion and detection of complications was 19 to 546 days. Six different complications were detected in one port catheter, while seven complications were detected in three different port catheters of a patient with AML.

Infectious, thrombotic, and mechanical complications were noted in 98 (69.5%), 29 (20.5%), and 14 (10%) port catheters, respectively. Nine patients (5%) were febrile on the day of port-catheter insertion and seven of them developed infectious complications, however the complication rates weren't different in patients with and without fever (p=0.150). All complications

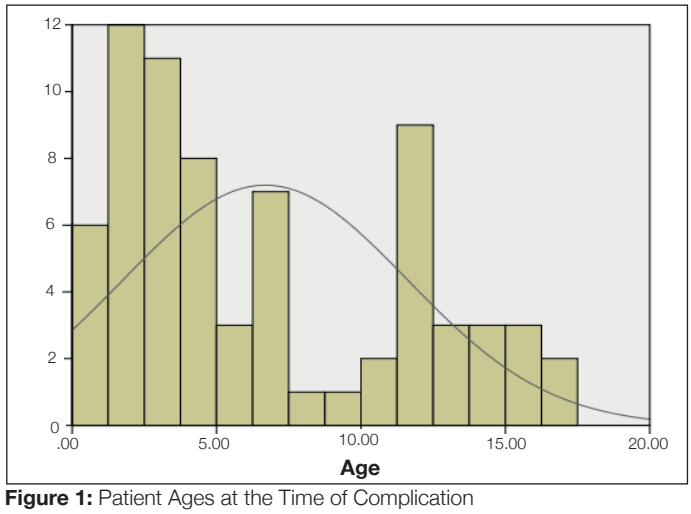

Figure 1: Patient Ages at the Time of Complication

Table I: Types of complications

Complication	n (%)	per 1000 days
Infection (n=98)		
Microbiologically documented catheter infection	70 (71.4)	5
Catheter related blood stream infections	16 (16.4)	
Local infections	12 (12.2)	
Mechanical (n=14)		
Leakage	4 (28.5)	0.8
Reservoir rotation	1 (7.1)	
Extravasation	3 (21.5)	
Catheter fracture	2 (14.3)	
Bleeding in catheter site	1 (7.1)	
Catheter failure	3 (21.5)	
Trombotic (n=29)		
Partial catheter occlusion	25 (86.3)	1.5
Venous thrombosis	4 (13.7)	

Table II: Complications of port catheters based on the time of occurrence

Complication	Mechanical*	Infectious*	Thrombotic*
Early (<30 days) (n=57)	10 (17.5)	42 (73.7)	5 (8.8)
Late (>30 days) (n=84)	4 (4.8)	56 (66.7)	24 (28.5)
Total (n=141)	14 (10)	98 (69.5)	29 (20.5)

***:** *n(%)*

Table III: Microorganisms identified in port catheter cultures

Microorganisms	n (%)
Gram positive bacteria	68 (61.8)
Coagulase negative *staphylococcus*	60 (54.6)
Others	8 (7.2)
Gram negative bacteria	36 (32.7)
Klebsiella	17 (15.4)
E. coli	5 (4.5)
Pseudomonas	4 (3.6)
Enterobacter	4 (3.6)
Others	6 (5.5)
Candida	6 (5.5)

were more frequently observed in the patient group that was neutropenic at the time of port insertion (63.6%, p <0.001), while a platelet count below 50 000/mm³ did not influence the frequency of complications (%53.4 vs 46.6%, p=0.880).

Total complications were noted at 7.3 per 1000 catheter days. Infectious, thrombotic, and mechanic complications were noted at 5, 1.5, and 0.8 per 1000 catheter days, respectively. Fifty-seven early (40.4%) and 84 late (59.6%) complications were noted. Complication types and periods were shown in Table I and Table II. Infectious complications were most prevalent in both the early and the late periods (p<0.001). Although early mechanical and late infectious complications were more common, no statistically significant difference was detected

Table IV: Port catheters after complications and outcome

	Continuation*	Revision*	Removal*
Mechanical (n=14)	8 (57)	4 (28.5)	2 (4.5)
Infectious (n=98)	77 (78.5)	0	21 (21.5)
Thrombotic (n=29)	25 (86)	3 (10.5)	1 (3.5)

***:** *n(%)*

between the type of complications (p=0.090). Most thrombotic complications occurred in the late period (p<0.001).

The vein used for port insertion and the types of complications were evaluated. The most used veins were the right external jugular vein (n=45; 32 of them complicated) and the left external jugular vein (n=25; 12 of them complicated). The complication rates of the external jugular vein and internal jugular vein were similar, however, the complication rate in the right external jugular vein was significantly higher compared to the left external jugular vein (50.6% vs 49.4%, p=0.010). All port-catheter tips were evaluated using echocardiography, and the frequency of complications was significantly higher in patients with catheter tips located in the right atrium (RA) (44.9%, p=0.011).

Infectious complications accounted for 98 (69.5%) of the 141 total complications. Among these, local infections were noted in 12 cases (12.2%). Microbiologically documented catheter infections and catheter-related bloodstream infections were detected in 70 (71.4%) and 16 (16.4%), respectively. Gram-positive bacteria, gram-negative bacteria and fungi were identified in 61.8%, 32.7%, and 5.5% catheter cultures, respectively. The most identified microorganisms were coagulase-negative *Staphylococci* (CoNS) and *Klebsiella species* (Table III). Port catheters were removed in 21 infection episodes due to infectious complications. Antibiotic lock therapy, in conjunction with systemic antibiotics, was used in 20 episodes of infection. Vancomycin was administered in 17 episodes for CoNS and *Enterococci*, while ciprofloxacin was used in two episodes for *E. coli* and one episode for *K. kingae*. Antibiotic lock therapy was ineffective in only two episodes.

Twenty-five (86.3%) of the thrombotic complications were classified as partial catheter occlusions. In cases of catheter occlusion, a heparin lock was attempted first; if recanalization was not achieved, a tissue plasminogen activator (tPA) was administered into the catheter lumen. Venous thrombosis was detected with doppler USG in four of 29 (13.7%) thrombotic complications. The locations of thrombosis were the right external jugular vein (n=2), superior vena cava (n=1), and left external jugular vein (n=1). Two of the four thromboses were acute, and these ports continued to be used with low molecule weight heparin therapy. Port catheters that were not recanalized with either heparin or tPA were revised or removed, respectively.

Mechanical complications were 10% (n=14) of all complications. These were leakage (n=4), reservoir rotation (n=1), extravasation (n=3), catheter fracture (n=2), bleeding in the catheter site (n=1), and catheter failure (n=3). Pneumothorax, arterial injury, air embolism, and arrhythmia did not occur in any patient.

In all complications, 78% of the port catheters continued to be used, 5% were revised, and 17% were removed. The outcome of catheters are shown on Table IV. The most common cause of catheter removal was an infection. Coagulase-negative *Staphylococci*, which is the main element of skin flora, are the most common infectious agent.

DISCUSSION

The use of port catheters provides significant convenience for both patients receiving chemotherapy and healthcare workers (5-7). Reported incidence rates of catheter-related complications in children with malignancies range from 6% to 41% (8-10). During our study period, the incidence of port-catheter-related complications was higher, with complications detected in 75% of port catheters, and early complications accounting for 40.4% of these. Although our study found a higher rate of port catheter-related complications compared to the literature, most of the affected ports remained usable, as most complications were microbiologically documented catheter infections with CoNS. The infection-related catheter removal rate has been reported as 9.3% to 26.8% (11).

The overall complication rate was found to be 7.3 per 1.000 catheter days. In a study including all types of central venous catheters, infection, thrombosis, and mechanical complications were reported at rates of 3.7, 0.2, and 0.4 per 1.000 catheter days, respectively, in patients with hematologic diseases (12). Port catheter-related infection rates were reported as 2.5 and 7.4 per 1.000 catheter days in two different studies from our country, which are consistent with our findings (13,14). In another study, port catheter-associated venous thrombosis, and infections were noted at 0.67 events and 0.84 events per 1000 catheter days, respectively (15). The higher incidence of complications in our study compared to the literature may be attributed to factors such as the variety of underlying diseases and treatments, differing criteria for classifying complications, variations in catheter care protocols, the type of vein used for catheter insertion, and the experience of both the insertion and management teams. Infectious complications were the most prevalent in both the early and late periods, while mechanical complications were more common in the early period. The early period is representative of the induction phases of leukemia treatment in which the patients are not in remission, have neutropenia or thrombocytopenia, and are most susceptible to infection. A high rate of mechanical complications might have been caused due to not using USG or fluoroscopic guidance at the time of catheter insertion.

In our study, thrombotic complications were common in the late period. Catheters are artificial surfaces and lack vascular endothelial function such as inhibition of platelet adhesion or inflammatory response. Thrombotic complications in the late period can be clarified by the duration of use is extended, the risk of both fibrin sheath formation and thrombosis increases due to the precipitation of the medical drugs, blood components, and handling errors (2,16). Neutropenia is another risk factor for leading port catheters to infectious complications (17). In the mechanical, infectious and thrombotic complication groups, early complications were seen statistically significantly in cases with neutrophil count <1500/mm^3 at the time of port insertion.

Efforts are ongoing to identify effective flushing and locking solutions to prevent both infectious and mechanical complications associated with port catheter devices (18). The use of antibiotic prophylaxis before placement is also controversial (19,20). A single dose of antibiotics targeted to skin flora is still given to immunocompromised patients before the insertion of CVC in some centers. Disinfection of the hub, needleless connector, or injection port with alcohol or chlorhexidine is also recommended (21). Consistent with the literature, we did not use routine antibiotic prophylaxis in pediatric cancer patients with port catheters during the study period. Studies have shown that implementing a catheter care bundle can prolong catheter life and reduce treatment costs by decreasing the frequency of infections. The key components of the catheter care bundle identified in these studies include skin cleansing with chlorhexidine, ensuring sterility during catheter insertion, selecting non-femoral veins for insertion, and adhering to strict sterility protocols in daily care (22). Devrim et al. (23) inserted split-septum catheters and utilized pre-filled syringes as part of their catheter care bundle. They demonstrated that the use of central line bundles not only decreased the risk of catheter-related infections but also prolonged the time to develop infections, resulting in cost-effectiveness (24).

In conclusion, port catheter complications remain a significant concern for children with cancer, presenting challenges despite the availability of standardized practice guidelines. Our study highlighted a high rate of complication-related port catheter removal, with infections—especially those caused by CoNS, a common element of skin flora—being the leading cause. The choice of vein for insertion and the positioning of the port-catheter tip are also key factors contributing to complications. Ensuring proper implantation, usage, and maintenance is essential to minimize both early and late complications. Furthermore, comprehensive training for clinicians, patients, and caregivers, including simulations, hand hygiene education, and the use of ultrasound guidance during catheter placement, is essential for improving outcomes and preventing complications.

Ethics committee approval

This study was conducted in accordance with the Helsinki Declaration Principles. The study was approved by Ankara Child Health and Diseases Hematology Oncology Training and Research Hospital (2014-246).

Contribution of the authors

Bağcı MS: Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments, Taking responsibility in logical

interpretation and conclusion of the results, Taking responsibility in necessary literature review for the study, Taking responsibility in the writing of the whole or important parts of the study. ***Yazal Erdem A:*** Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in logical interpretation and conclusion of the results, Taking responsibility in the writing of the whole or important parts of the study. ***Özbek NY:*** Constructing the hypothesis or idea of research and/or article. ***Yaralı N:*** Constructing the hypothesis or idea of research and/or article, Planning methodology to reach the conclusions, Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in logical interpretation and conclusion of the results, Taking responsibility in the writing of the whole or important parts of the study, Reviewing the article before submission scientifically besides spelling and grammar.

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Conflict of interest

The authors declare that there is no conflict of interest.

REFERENCES

1. Ullman AJ, Bernstein SJ, Brown E, Aiyagari R, Doellman D, Faustino EVS, et al. The Michigan Appropriateness Guide for Intravenous Catheters in Pediatrics: miniMAGIC. Pediatrics 2020;145:S269-S284. https://doi.org/10.1542/peds.2019-3474I
2. Bawazir O, Banoon E. Efficacy and clinical outcome of the port-a-cath in children: a tertiary care-center experience. World J Surg Oncol 2020;18:134. https://doi.org/10.1186/s12957-020-01912-w
3. Machat S, Eisenhuber E, Pfarl G, Stübler J, Koelblinger C, Zacherl J, Schima W. Complications of central venous port systems: a pictorial review. Insights Imaging 2019;10:86. https://doi.org/10.1186/s13244-019-0770-2
4. Ross AB, Rouanet E, Murphy AJ, Weldon CB, Weil BR. Complications associated with totally implantable access ports in children less than 1 year of age. J Pediatr Surg 2022;57:463-8. https://doi.org/10.1016/j.jpedsurg.2021.12.004
5. Kılıç S, Soyer T, Karnak İ, Çiftçi AÖ, Tanyel FC, Şenocak ME. Evaluation of the removal reasons of totally implantable venous devices in children: a retrospective study. Turk J Pediatr 2016;58:187-94. https://doi.org/10.24953/turkjped.2016.02.010
6. Zhang H, Li Y, Zhu N, Li Y, Fu J, Liu J. Comparison of peripherally inserted central catheters (PICCs) versus totally implantable venous-access ports in pediatric oncology patients, a single center study. Sci Rep 2022;12:3510. https://doi.org/10.1038/s41598-022-07584-8
7. van den Bosch CH, Spijkerman J, Wijnen MHWA, Hovinga ICLK, Meyer-Wentrup FAG, van der Steeg AFW, et al. Central venous catheter-associated complications in pediatric patients diagnosed with Hodgkin lymphoma: implications for catheter choice. Support Care Cancer 2022;30:8069-79. https://doi.org/10.1007/s00520-022-07256-3
8. Jan HC, Chou SJ, Chen TH, Lee CI, Chen TK, Lou MA. Management and prevention of complications of subcutaneous intravenous infusion port. Surg Oncol 2012;21:7-13. https://doi.org/10.1016/j.suronc.2010.07.001
9. Cesaro S, Cavaliere M, Pegoraro A, Gamba P, Zadra N, Tridello G. A comprehensive approach to the prevention of central venous catheter complications: results of 10-year prospective surveillance in pediatric hematology-oncology patients. Ann Hematol 2016;95:817-25. https://doi.org/10.1007/s00277-016-2634-x
10. Sosnowska-Sienkiewicz P, Moryciński S, Januszkiewicz-Lewandowska D, Michalik K, Madziar K, Kukfisz A, et al. Totally implantable venous ports in infants and children: a single-center retrospective study of indications and safety. Front Oncol 2024;14:1351630. https://doi.org/10.3389/fonc.2024.1351630
11. Kugler E, Levi A, Goldberg E, Zaig E, Raanani P, Paul M. The association of central venous catheter placement timing with infection rates in patients with acute leukemia. Leuk Res 2015;39:311-3. https://doi.org/10.1016/j.leukres.2014.12.017
12. Pektaş A, Kara A, Gurgey A. Cohort Study: Central Venous Catheter-Related Complications in Children with Hematologic Diseases at a Single Center. Turk J Haematol 2015;32:144-51.https://doi.org/10.4274/Tjh.2013.0403
13. Sarper N, Zengin E, Corapçioglu F, Tugay M. Totally implantable central venous access devices in children with hemato-oncologic malignancies: evaluation of complications and comparison of incidence of febrile episodes with similar patients without central venous access devices. Pediatr Hematol Oncol 2006;23:459-70. https://doi.org/10.1080/08880010600712514
14. Celebi S, Sezgin ME, Cakır D, Baytan B, Demirkaya M, Sevinir B, et al. Catheter-associated bloodstream infections in pediatric hematology-oncology patients. Pediatr Hematol Oncol 2013;30:187-94. https://doi.org/10.3109/08880018.2013.772683
15. Beckers MM, Ruven HJ, Seldenrijk CA, Prins MH, Biesma DH. Risk of thrombosis and infections of central venous catheters and totally implanted access ports in patients treated for cancer. Thromb Res 2010;125:318-21. https://doi.org/10.1016/j.thromres.2009.06.008
16. Onyeama SN, Hanson SJ, Dasgupta M, Baker K, Simpson PM, Punzalan RC. Central Venous Catheter-associated

Venous Thromboembolism in Children With Hematologic Malignancy. J Pediatr Hematol Oncol 2018;40:e519-e524. https://doi.org/10.1097/MPH.0000000000001229

17. Planas Díaz I, Molina Mata M, Casal Beloy I, Cabello Laureano R. Indwelling-tunneled-central-venous-catheter-related early bacteremia and preoperative prophylaxis: a case and control study. Cir Pediatr 2024;37:99-103. https://doi.org/10.54847/cp.2024.03.10
18. Rickard CM, Ullman AJ. Bloodstream infection and occlusion of central venous catheters in children. Lancet Infect Dis 2018;18:815-7. https://doi.org/10.1016/S1473-3099(18)30267-6
19. Johnson E, Babb J, Sridhar D. Routine Antibiotic Prophylaxis for Totally Implantable Venous Access Device Placement: Meta-Analysis of 2,154 Patients. J Vasc Interv Radiol 2016;27:339-43. https://doi.org/10.1016/j.jvir.2015.11.051
20. Akbar SA, Qudsia ZF, Ashraf MN, Tarar MA, Qazi AQ. Prophylactic Antibiotics for Reducing Central Line-associated Blood Stream Infection in Children with Totally Implantable Venous Access Devices. J Coll Physicians Surg Pak 2020;30:304-8. https://doi.org/10.29271/jcpsp.2020.03.304
21. Chesshyre E, Goff Z, Bowen A, Carapetis J. The prevention, diagnosis and management of central venous line infections in children. J Infect 2015;71:S59-75. https://doi.org/10.1016/j.jinf.2015.04.029
22. Rabelo BS, de Alvarenga KAF, Miranda JWFB, Fagundes TP, Cancela CSP, de Castro Romanelli RM, et al. Risk factors for catheter-related infection in children with cancer: A systematic review and meta-analysis. Am J Infect Control 2023;51:99-106. https://doi.org/10.1016/j.ajic.2022.05.005
23. Devrim İ, Özkul MT, Çağlar İ, Oruç Y, Demiray N, Tahta N, Vergin C. Central line bundle including split-septum device and single-use prefilled flushing syringes to prevent port-associated bloodstream infections: a cost and resource-utilization analysis. BMC Health Serv Res 2020;20:336. https://doi.org/10.1186/s12913-020-05221-6
24. Devrim İ, Sandal OS, Çelebi MY, Hepduman P, Gönüllü A, Atakul G, et al. The impact of central line bundles on the timing of catheter-associated bloodstream infections and their microbiological distribution in critically ill children. Eur J Pediatr 2023;182:4625-32. https://doi.org/10.1007/s00431-023-05141-7

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Original Article

Neurologic toxicity in children with acute lymphoblastic leukemia

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ABSTRACT

Objective: The objective of this study was to evaluate the occurence of neurological complications and long-term neurological sequelae in children with acute lymphoblastic leukemia (ALL). These complications were examined in relation to demographic characteristics, leukemia risk groups, and chemotherapy type.

Material and Methods: A total of 165 patients aged between 1 and 18 years of age who underwent ALL IC-BFM 2009 chemotherapy at the Pediatric Hematology Unit of Ankara Pediatric Hematology and Oncology Research and Training Hospital between June 2013 and December 2018 were retrospectively evaluated.

Results: Forty-two neurological complication episodes (1 to 3 per patient) were observed in 37 (22.4%) patients during chemotherapy. No significant differences between groups with or without neurological complications were detected in terms of age, gender, type of leukemia, risk group assignment, and relapse status (p=0.150, p= 0.170, p= 0.810, p= 0.370, and p=0.340, respectively). Complications were more likely to occur in preB-ALL patients with intermediate to high-risk status, and approximately half of the complications were identified during the early phases of treatment, i.e., induction and early consolidation; also, vincristine, methotrexate, and corticosteroids were more likely to lead to neurotoxicity. The two most common complications included polyneuropathy in 47.6% of the patients and posterior reversible encephalopathy syndrome in 16.7%. Other complications included cranial neuropathy, secondary intracranial hypertension, cortical atrophy, epilepsy, encephalopathy, myopathy, cranial thrombosis, psychotic disorder, and cerebral edema. While none of the neurological complications were associated with mortality, 21.4% of the patients had varying types of sequela, the most common being epilepsy.

Conclusion: Despite increased success rates with intense therapeutic approaches in pediatric ALL patients, 22.4% of this population experienced neurological complications. Long-term follow-up is warranted to evaluate the adverse effects and sequelae of chemotherapy more definitely.

Keywords: Acute lymphoblastic leukemia, Neurologic complications, Polyneuropathy, Posterior reversible encephalopathy syndrome

INTRODUCTION

Acute lymphoblastic leukemia (ALL) comprises one-third of all cancers of childhood and 75% to 80% of all acute leukemias. It occurs more commonly in males, Caucasians, and pediatric patients between 2 and 5 years of age (1). The reported incidence in our country is estimated to be 1.4/100000 (2). Significant variations may exist between ALL patients in terms of clinical and laboratory characteristics, treatment, follow-up, and many disease subtypes have been described. Treatment strategies include different chemotherapy regimens and are based on four basic therapy steps constructed on risk groups and prognostic factors. These treatment steps include remission induction, consolidation for residual leukemia, central nervous system (CNS) eradication, and maintenance. The objectives of the treatment include achievement of remission, prevention of relapses, and eradication of minimal residual disease (MRD). Recently introduced treatments not only have been able to reduce the risk of relapse but also achieved event-free survival rates of up to 85% to 90% (3,4). However, this also led to an increased occurrence of complications caused by early and/or late side effects of such treatments. Early recognition and treatment of these complications are of utmost importance due to associated high morbidity and mortality (5,6). The dosage, route, length of administration, and concomitant use of chemotherapeutic agents are among the important

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determinants of the risk of complications (7,8). While CNS and peripheral nervous system involvement is rare at the time of diagnosis, complications involving both of these nervous systems may occur during ALL treatment. Involvement of the peripheral nervous system is defined as the involvement of any part of the nervous system, including motor neurons, sensory ganglia, nerve roots, plexuses, cranial and peripheral nerves, and neuromuscular connections.

Chemotherapy may cause both peripheral neurotoxicity, consisting mainly of a peripheral neuropathy, and central neurotoxicity, ranging from minor cognitive deficits to encephalopathy. Peripheral neuropathy may be the dose-limiting toxicity, therefore the assessment of peripheral neuropathy and its impact on quality of life should be evaluated during therapy. CNS toxicities such as seizures, drowsiness, cognitive deficits or even coma are observed less frequently. However, these adverse effects should always be taken into account when starting clinical chemotherapeutic trials.

In this study, we aimed to evaluate neurological complications and long-term neurological sequela in a group of pediatric ALL patients receiving ALL IC-BFM 2009.

MATERIALS and METHODS

A total of 165 ALL patients aged between one and 18 years and receiving ALL IC-BFM 2009 protocol at Pediatric Hematology Unit, Ankara Pediatric Hematology and Oncology Research and Training Hospital between June 2013 and December 2018 were retrospectively examined for neurological complications (9).

Risk groups were defined based on BFM ALL IC 2009 protocol; standard risk group (SRG):<1x 10^9 /L blasts on peripheral blood smear on day eight, age between one and six years, initial leukocyte count <20x10^9/L, MRD on day 15<0.1%, and <5% blasts on bone marrow aspirate on day 33; high-risk group (HRG): hypodiploidy or t(9;22) or t(4;11) or more than 1x10^9/L blasts on peripheral blood smear on day 8 or MRD on day 15>10% or more than 5% blasts on bone marrow aspirate on day 33; intermediate-risk group (IRG): patients not stratified as SRG or HRG.

CNS status was defined as CNS-1 (no detectable blast cells in a sample of cerebrospinal fluid), CNS-2 (<5 leukocytes per cubic millimeter with blast cells in a sample with <10 erythrocytes per cubic millimeter), CNS-3 (≥5 leukocytes per cubic millimeter with blast cells in a sample with <10 erythrocytes per cubic millimeter), or traumatic lumbar puncture with blast cells (≥10 erythrocytes per cubic millimeter with blast cells).

Demographic data such as age, gender, age at diagnosis, age at onset of symptoms as well as the type of leukemia, risk group, presence of CNS involvement, relapse/remission status, neurological complications before treatment, concomitant symptoms and signs, phase of chemotherapy at the time of complications, relationship to drugs, comorbid conditions, laboratory results, computed tomography (CT) and magnetic resonance imaging (MRI) findings, and electroencephalography (EEG) and electromyography (EMG) results were retrieved from the electronic database. Exclusion criteria included the presence of leukemias other than acute lymphoblastic leukemia, infant leukemia, follow-up treatments in another center, and bone marrow transplantation.

Statistical analysis

The Statistical Package for the Social Sciences (SPSS) version 22.0 (IBM Corp., Armonk, New York, USA) was used to analyze the data. Descriptive statistics were presented as frequency, percentage, mean, and standard deviation. The normal distribution of the data was tested with Shapiro-Wilk's test. The difference in age and age at onset of symptoms between patients with or without neurological complications was examined using the Mann-Whitney test, while percentages of demographic characteristics were analyzed with the chi-square test. p value <0.050 was considered statistically significant.

RESULTS

Of the 165 ALL patients included, 97 (58.8%) were male and 68 (41.2%) were female. The medan age at diagnosis was 7.5± 4.5 years (min-max, 1.6-17.7 years). Pre B ALL and T-cell ALL were diagnosed in 144 (87.3%) and 21 (12.7%) of the patients, respectively. There were 13 patients (7.9%) in the standard risk group, 83 patients (50.3%) in the intermediate-risk group; and 69 patients (41.8%) in the high-risk group. One-hundred and forty-eight patients (89.7%) were in remission, while 31 (18.8%) had a relapse during follow-up. Central nervous system involvement (CNS-3) was present in 5/165 patients (3%) at the time of diagnosis. During the study, 42 neurological complications were detected in 37 patients (22.4%) on average at the 7th months of the treatment (min-max, 1-26 months). The mean age of these patients was 8.4±4.7 years and 23 (62.2%) were male.

Of the 37 patients, 31 (83.8%) had a diagnosis of pre B-ALL (21.5% of all pre B-ALL patients), and 35 neurological episodes were identified in these cases. On the other hand, six (16.2%) had a diagnosis of T-ALL (28.5% of all T-ALL patients), and neurologic episodes were detected of them. Both IR and HR patients had 20 episodes each (47.6%), while two complications (4.8%) emerged in SRG patients.

Nine of the patients (24.3%) who developed neurological complications were relapsed ALL. In these patients, neurological complications occurred during relapse treatment in seven (77.8%) and maintenance treatment in two. Two of the patients with neurological complications (5.4%) had leukemic involvement of the CNS at the time of diagnosis. Patients with

Table I: Characteristics of patients

Properties		Neurological Complications		
	All patients (n=165)	Yes (n=37)	No (n=128)	p
Age *(year)	7.5±4.5	8.4±4.7	7.2±4.5	0.150‡
Gender†				
Male	97 (58.8)	23 (62.2)	74 (57.8)	0.170§
Female	68 (41.2)	14 (37.8)	54 (42.2)	
Type of leukemia†				
Pre B-ALL	144 (87.3)	31 (83.8)	113 (88.3)	0.810§
T-ALL	21 (12.7)	6 (16.2)	15 (11.7)	
Risk†				
SRG	13 (7.9)	2 (5.4)	11 (8.6)	0.370§
IRG	83 (50.3)	18 (48.6)	65 (50.8)	
HRG	69 (41.8)	17 (45.9)	52 (40.6)	
Relapse†	31 (18.8)	9 (24.3)	22 (17.2)	0.340§
CNS involvement†				
Yes	5 (3)	2 (5.4)	3 (2.3)	
No	160 (97)	35 (94.6)	125 (97.7)	

***:** *mean*±SD, **†:** *n(%)*,**‡:** *Mann-Whitney test*, **§:** Chi-square test, ***ALL:** acute lymphoblastic leukemia*, ***SRG:** standard risk group*, ***IRG:** intermediate risk group*, ***HRG:** high-risk group*, ***CNS:** central nervous system*

Table II: Neurological complications

Diagnosis	Complication-detected* (n=42)	All ALL patients† (n=165)
Polyneuropathy	18 (42.8)	10.9
PRES	7 (16.7)	4.2
Cranial neuropathy	3 (7.1)	1.8
Myopathy	3 (7.1)	1.8
Secondary intracranial hypertension	3 (7.1)	1.8
Cortical atrophy	2 (4.8)	1.2
Epilepsy	2 (4.8)	1.2
Encephalopathy	1 (2.4)	0.6
Cranial thrombosis	1 (2.4)	0.6
Psychotic disorder	1 (2.4)	0.6
Cerebral edema	1 (2.4)	0.6

***:** *n(%)*,**†:** *%*, ***PRES:** posterior reversible encephalopathy syndrome*

or without neurological complications were not significantly different in terms of age, gender, leukemia type, risk groups, and relapse status (p=0.150, p= 0.170, p= 0.810, p= 0.370, and p=0.340, respectively) (Table I).

There was a total of 22 patients (13.3%) who had muscle weakness, gait impairment, leg pain, and loss of deep tendon reflexes. Motor and/or sensory axonal polyneuropathy (PNP) was diagnosed in 14 patients based on EMG assessment, and in 4 patients based on clinical findings (n=18; 10.9%); in all of these cases, the event was considered associated with vincristine (VCR) treatment. Three patients have one or more of neuropathic symptoms and signs, three patients were diagnosed steroid-related myopathy and one patient was diagnosed intrathecal MTX-related cytotoxic cerebral edema.

Of the patients with polyneuropathy, 83.3% had pre B-ALL with 50% diagnosed in induction, 16.7% in reinduction, and 5.5% in HR consolidation. All of these protocol phases included use of Vincristine and Prednisone/dexamethasone.

Following the emergence of the complications, treatment consisting of B-vitamin complex and gabapentin was given. Three patients (1.8%) developed neuropathic pain and one (0.6%) had dropped leg as a sequela. In three patients (1.8%) cranial neuropathy was present. Autonomic neuropathy symptoms such as constipation could not be retrieved from patients' records due to missing data. There were 11 patients (6.7%) who had convulsions, in association with headache, visual and speech impairment, altered consciousness, and loss of muscle strength. In seven patients (4.2%), a cranial MRI was performed, and posterior reversible encephalopathy syndrome (PRES) was diagnosed. In four other patients with convulsions, sinus venous thrombosis, encephalopathy, and epileptic activity were detected. Six (85.7%) of the patients with PRES had pre B-ALL, and five (71.4%) had HR status and received induction, reinduction, HR block, and relapse treatments. Six (85.7%) had hypertension, and two (28.6%) had hyponatremia at the time of the episode. Following the occurrence of the complications, appropriate antiepileptics, antihypertensives, and sodium replacement were given to symptoms and comorbidities. Of the patients with PRES, three (42.8%) had epileptic sequela. Table II shows all neurological complications.

In a patient (0.6%) in the IR group aged 16.6 years and diagnosed with pre B-ALL, a cranial MRI was performed due to headache, convulsion, and loss of motor strength during induction treatment revealed hemorrhage; also, venography showed sinus venous thrombosis which was thought to be associated with steroids and L-asparaginase given during induction. Low molecular weight heparin at a dose of 100 U/kg BID was administered for one month, followed by once-daily administration for six months. Thrombosis was completely

Table III: Summary of clinical, radiographic and electrophysiological findings

Leukemia Type	Risk/Patient number	Age (year)	Protocol	Symptoms	Signs	Eye Exam	EEG	EMG	CT	MRI	LP	MR Venography	Diagnosis	Treatment	Sequela
PreB-ALL	SRG 1	4.5	PIA	Walking disorder	DTR loss			None					PNP	B-vit complex	
	2	3.6	PIB	Walking disorder	DTR loss- Loss of strength			None		N			Myopati	B-vit complex	
	IRG 3	3.1	PIA	Walking disorder	Loss of strength			None					PNP	B-vit complex	
	4	10.2	PIA	Seizure	Confusion		Encephalopathy		Cortical atrophy	PRES			PRES	Antihypertensive + Antiepileptic	Epilepsy
	5	4.5	PIA	Seizure	Confusion		Cerebral disfunction			PRES			PRES	Antihypertensive + Antiepileptic	Epilepsy
	6	4.2	PIA	Weakness	DTR loss			Motor axonal PNP					PNP	B-vit complex + Gabapentin	
	7	14.5	PIA	Weakness- Walking disorder	DTR loss			Motor axonal PNP					PNP	B-vit complex + Gabapentin	
	8	16.6	PIA	Seizure- Headache	Loss of strength	N				Subdural hemorrhage	Sagittal sinüs thrombosis		Cranial thrombosis	Antiepileptic + LMWH	
		17	PII-faz1	Aggression	None	N				N			Psychotic disorder	Antipsychotic	
	9	3.3	PM	Weakness- Walking disorder	Loss of strength			Motor axonal PNP		N			PNP	B-vit complex + Gabapentin	
	10	12.2	PIB	Headache	None					N	Increased CSF pressure		Secondary intracranial hypertension	Acetazolamide	
	11	6.5	PIB	Weakness- Walking disorder	DTR loss			Motor axonal PNP		Cortical atrophy			PNP	B-vit complex + Gabapentin	
	12	5.8	PIB	Walking disorder	None			None					Myopati	B-vit complex	
	13	3.9	PIB	Weakness- Walking disorder	DTR loss			None					Myopati	B-vit complex	
		6.9	Relaps	Ptosis	Myosis	Myosis, Ptosis				N			Cranial neuropathy	Gabapentin	
PreB-ALL	IRG 14	16.7	PII-faz1	Weakness- leg pain	DTR loss- Loss of strength			Motor axonal PNP		N			PNP	B-vit complex + Gabapentin	Neuropathic pain
	15	4.9	PII-faz1	Weakness- Walking disorder	DTR loss			Motor axonal PNP					PNP	B-vit complex + Gabapentin	
	16	11.3	PII-faz2	Weakness	Loss of strength				N	Cytotoxic edema		N	Cerebral edema		
	17	10.4	Relapse	Headache- Vomting	None	N				N	Increased CSF pressure		Secondary intracranial hypertension	Acetazolamide	
	18	15.9	Relapse	Drooping of the mouth	Cranial nerve paralysis- hemiplegia					Restricted diffusion		N	Cranial neuropathy		
	HRG 19	6.6	PIA	Weakness- leg pain- Walking disorder	DTR loss- Loss of strength			Motor axonal PNP					PNP	B-vit complex + Gabapentin	

Leukemia Type	Risk/Patient number	Age (year)	Protocol	Symptoms	Signs	Eye Exam	EEG	EMG	CT	MRI	LP	MR Venography	Diagnosis	Treatment	Sequela
PreB-ALL	20	11.9	PIA	Weakness- Walking disorder	DTR loss- Loss of strength			Motor axonal PNP					PNP	B-vit complex + Gabapentin	Drop foot
	21	2	PIA	Walking disorder	None			Motor axonal PNP					PNP	B-vit complex + Gabapentin	
	22	3.1	PIA	Ptosis	Cranial nerve paralysis	Left eye ptosis				N			Cranial neuropathy		
	23	6.6	PIA	Weakness- leg pain	DTR loss			Motor axonal PNP		N			PNP	B-vit complex + Gabapentin	
	24	6.8	PIB	Weakness- Walking disorder	Loss of strength			Motor axonal PNP		N			PNP	B-vit complex + Gabapentin	
	25	12.6	PII-faz2	Seizure	Confusion				N	PRES		N	PRES	Antiepileptic	Epilepsy
	26	15.1	1.HR1	Seizure- Headache	Confusion		N		N	PRES			PRES	Antihypertensive + Antiepileptic	
	27	5.9	2.HR1	Seizure	Confusion		N			PRES			PRES	Antihypertensive + Antiepileptic	
PreB-ALL	HRG 28	17.1	2.HR2	Seizure- Dysarthria	Confusion	N	Cerebral disfunction- Epileptic activity			Cerebral-cerebellar atrophy			Atrophy	Antiepileptic	
		17.1	2.HR2	Weakness- Tremor	DTR loss-dismetri			Bilateral sensorimotor PNP		Cerebral-cerebellar atrophy			PNP	B-vit complex + Gabapentin	Neuropathic pain
		17.2	2.HR3	Seizure	None	N	Epileptic activity		N	Cerebral-cerebellar atrophy			Epilepsy	Antiepileptic	Epilepsy
	29	9.6	Relapse	Headache- Vomiting- Seizure	Confusion					PRES			PRES	Antihypertensive + Antiepileptic	
	30	6.1	Maintenance	Seizure	Confusion		N		Cortical atrophy	N		N	Atrophy		
	31	15.3	Meintenance	Weakness- leg pain	None			None					PNP	B-vit complex + Gabapentin	Neuropathic pain
T-ALL	IRG 1	3.2	PIA	Weakness- Walking disorder	DTR loss			None					PNP	B-vit complex + Gabapentin	
	2	16.2	Relapse	Headache- Speech disorder	None		Epileptic activity			N			Epilepsy	Antiepileptic	Epilepsy
	HRG 3	9.8	PIA	Seizure – Vision problem	Confusion		Abnormal background activity			N			Encephalopathy	Antihypertensive + Antiepileptic	
	4	14.3	PII-faz1	Weakness- leg pain- Walking disorder	DTR loss- Loss of strength- Numbness			Bilateral sensorimotor PNP					PNP	B-vit complex + Gabapentin	
		15.1	Relapse	Seizure	Confusion	N			N	PRES			PRES	Antihypertensive + Antiepileptic	
	5	14.6	PII-faz1	Weakness- Walking disorder	Gowers's sign			Motor axonal PNP					PNP	B-vit complex + Gabapentin	
	6	4	Relapse	Vision Loss	None	Papil edema			N	N	Increased CSF pressure		Secondary intracranial hypertension	Acetazolamide	

EEG: *electroencephalography,* ***EMG:*** *electromyography,* ***CT:*** *computed tomography,* ***MRI:*** *magnetic resonance imaging,* ***LP:*** *lumbar puncture,* ***PreB-ALL:*** *precursor B cell acut lymphoblastic leukemia,* ***T-ALL:****T cell acut lymphoblastic leukemia,* ***IRG:*** *intermediate risk group,* ***HRG:*** *high risk group,* ***PIA:*** *protocol IA,* ***DTR:****deep tendon reflex,* ***PNP:*** *polyneuropathy,* ***PRES:*** *posterior reversible encephalopathy syndrome,* ***CSF:*** *cerebrospinal fluid,* ***B-vit:*** *B-vitamin,* ***N:*** *normal*

resolved with no sequela. The same patient exhibited aggressive behavior during reinduction treatment, although his cranial imaging studies were unremarkable. In consultation with pediatric psychiatry, a diagnosis of steroid-associated psychotic disorder was made. Anti-psychotic treatment was given and the patient recovered without sequela.

Sudden visual loss was seen in three patients (1.8%) (2 of them were pre B-ALL in the IR group and the other patient was T-ALL in the HR group). These patients' cranial imaging showed normal findings, although a lumber puncture indicated increased cerebrospinal fluid pressure. Thus, these patients were diagnosed with secondary intracranial hypertension. These patients recovered fully with acetazolamide treatment. Chemotherapy protocol was modified in seven patients after neurological complications. In three patients (one patient with sinus venous thrombosis, and the other two with PNP), chemotherapy was suspended for one week; in two patients, (one with convulsions and PRES, another with PNP) chemotherapy was maintained with dose reduction; in two patients diagnosed with PNP, vincristine dose was skipped until the next chemotherapy course. Clinical, radiographic and electrophysiological findings of patients are summarized in Table III.

DISCUSSION

Neurological side effects in pediatric ALL patients were detected as 22.4%. Of these, 11.5% and 10.9% affected the CNS and peripheral nervous system, respectively. In several previous studies, the respective figures range between six to 11% and 18 to 50% (10,11). Ethnicity, genetic factors, age, gender, and obesity may account for some of these differences in the reported rates of incidence (16).

In the current study, most of the patients with neurological complications had HR or IR status, and side effects mostly occurred during induction and early consolidation. Baytan et al. (10), administering a very similar chemotherapy protocol to ours (i.e. ALL-BFM 2000), observed neurological complications in 65.2% of the patients in the IR group, and 56.5% of the patients receiving induction treatmet. On the other hand, Aytaç et al. (13) found a neurological complication rate of 60% in HR patients receiving St. Jude's total XI and XIII protocol. In our study, 18 patients (10.9%) were diagnosed with motor and/or sensory axonal polyneuropathy. Of the cases with polyneuropathy, 66.7% were associated with VCR used in induction and early consolidation phases. It appears that incidence may vary according to age, gender, ethnicity, obesity, malnutrition, and genetic polymorphism. The TT (rs924607) genotype of CEP72, a gene involved in microtubule formation; C > T (rs3784867) variation of ABCC1 gene; ABCC2 GG (rs3740066) and GG (rs12826) genotypes; and all homozygous mutant alleles of SLC5A7 gene are associated with reduced CYP3A4 and CYP3A5 enzyme functions, with a consequent increase in the risk of vincristine related neuropathy (14,15). We did not examine any of our patients' polymorphisms associated with VCR neuropathy. The mean age of patients with polyneuropathy was comparable with the overall group of patients with neurological complications, and there were no significant gender differences (55% male). Three patients (1.8%) had cranial neuropathy. No information could be collected from patient files in terms of the development of autonomic neuropathy. As opposed to peripheral or cranial neuropathy, the common occurrence of more general symptoms such as constipation and abdominal pain in patients with autonomic neuropathy may lead to the underdiagnosis of this condition. Clinicians should have a higher index of suspicion for autonomic symptoms when assessing patients with possible neuropathy.

Peripheral neuropathy may be confused with pathological signs of myopathy. Muscle weakness and/or gait disorder could not be related to vincristine use in three of our patients (1.8%), and a diagnosis of myopathy associated with steroid use was made. A routine physical exercise program may have a protective role against the future development of muscular atrophy or joint problems (16).

Seven patients (4.2%) with seizures were diagnosed with PRES, and 5 of these (71.4%) had a high-risk status. Patients were receiving induction, consolidation, re-induction, and relapse treatments, and seizures were probably related to intrathecal MTX or corticosteroids. Six patients (85.7%) had hypertension, and two (28.6) had hyponatremia at the time of the seizure. In the Baytan study, the incidence of PRES was 4.8%, with a higher frequency in HR patients (17). In a multi-center study by Bilir et al. (18), 84.4% of the 58 patients with PRES also had concomitant hypertension. In studies by Tang et al. (19) hypertension was reported to occur in 36.4% of the patients, respectively. Furthermore, it has also been proposed that intravascular hypotonicity due to hyponatremia may lead to extravasation, which then may enter astrocytes through water channels such as aquaporin-4 and may result in cranial edema, as a part of the pathogenesis of PRES (20). In our study, epilepsy as a sequela of PRES was observed in three of the patients (42.4%) diagnosed with PRES. This is consistent with previous reports suggesting that PRES developing during leukemia treatment may be irreversible or may result in epileptic sequela (19).

One of our patients (0.6%) had bleeding and sinus venous thrombosis. The reported incidence of thrombosis developing during treatment of pediatric ALL varies between 1% and 37%, depending on the populations and therapeutic regimens examined. Sinus venous thrombosis represents one of the highest-risk areas of involvement, as it is associated with long term neurologic effects and a high mortality rate of 8% to 13% (21). Risk factors for thrombosis included T cell phenotype, older age, and high-risk status. In other studies, intensive chemotherapy regimens involving long-term exposure to

L-asparaginase and steroids appeared to be associated with the highest risk of thrombosis (22). In our patient, thrombosis was observed during induction treatment and was thought to be associated with L-asparaginase administered during that treatment. L-asparaginase increases the risk of thrombosis and bleeding via decreased hepatic synthesis of fibrinogen and anti-thrombin. Particular attention to L-asparaginase treatment and thrombosis should be paid in pediatric patients since these are directly linked with the course of leukemia and survival (23). Our patient who had a thrombotic event developed the steroid-associated psychotic disorder in the subsequent course of chemotherapy. Treatment with an anti-psychotic agent resulted in full recovery.

Three patients (1.8%) in our study were diagnosed with secondary intracranial hypertension, which is a rare condition in the general pediatric population, with a reported incidence of 0.32/100.000 in the US (24). Several studies involving patients with hematological malignancies have found that certain medications including methotrexate, cytarabine, and all trans-retinoic acid may be associated with symptoms of increased intracranial pressure (25). Similarly, in our patients, increased intracranial pressure developed during early consolidation and relapse treatments, which included methotrexate, and cytosine-arabinoside. In Garcia et al. (26) study comparing increased intracranial pressure syndromes among pediatric leukemia patients, headache and papilledema were present in 90% and 25% of the patients at the time of diagnosis, respectively. Similarly, two of our patients had a headache, and one had a sudden loss of vision. Treatment with acetazolamide resulted in full recovery in all cases.

In conclusion, during the treatment of ALL, a total of 22.4% of neurological complications were detected, of which 11.5% were CNS and the most common was PNP. In 21.4% of these cases, long-term sequela such as epilepsy, neuropathic pain, and foot drop was observed. Potential limitations of our study include its retrospective nature, missing data in patient files, absence of accurate data on the relationship between complications and chemotherapy agents, and lack of routine use of EMG in all patients. Larger and prospective studies may allow better identification of side effects of a wide range of chemotherapeutic agents, in addition to providing further insights into pathophysiologic mechanisms and/or genetic factors for such effects. This may allow the prevention of complications with the use of non-invasive tests or relevant genetic techniques; also, appropriate management strategies against such complications may be developed to achieve sequela-free survival.

Ethics committee approval

This study was conducted in accordance with the Helsinki Declaration Principles. The study was approved by the institutional ethics committee of the Ankara Child Health and Disease Hematology-Oncology Training and Research Hospital (date: 08.04.2019, number: 2019/080).

Contribution of the authors

Özcan AS, Yaralı N: Concept, ***Özcan As, Yaralı Ny:*** Design, ***Özcan AS, Yaralı NY, Akçabelen Ym:*** Supervision, ***Özcan AS, Işık M:*** Data Collection, ***Özcan As, Koca Yozgat A, Akçabelen YM***: Analysis And/Or Interpretation, ***Özcan AS, Koca Yozgat A, Yaralı N:*** Literature Review, ***Özcan AS, Yaralı N:*** Writing, ***Özbek NY Yaralı N:*** Critical Review, ***Özcan AS, Yaralı N:*** References And Fundings, ***Özcan AS, Koca Yozgat A:*** Materials

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Conflict of interest

The authors declare that there is no conflict of interest.

REFERENCES

1. Hunger SP, Mullighan CG. Acute lymphoblastic leukemia in children. N Engl J Med 2015;373:1541-52. https://doi.org/10.1056/NEJMra1400972
2. Kutluk T. Çocukluk çağı kanserlerinin epidemiyolojisi. Klin Gelişim 2006: p. 11-5.
3. Tekgunduz E, Demir M, Akpinar S. Prognostik faktörler ışığında akut lenfoblastik lösemi. Int J Hematol Oncol 2010;20:57-66.
4. Carroll WL, Bhatla T. Acute lymphoblastic leukemia. in: Lanzkowsky P, Lipton JM, Fish JD, editors. Lanzkowsky's Manual of Pediatric Hematology and Oncology. 6th ed. Academic Press 2016: 367-89. https://doi.org/10.1016/B978-0-12-801368-7.00018-1
5. Ward E, DeSantis C, Robbins A, Kohler B, Jemal A. Childhood and adolescent cancer statistics, 2014. CA Cancer J Clin 2014;64:83-103.https://doi.org/10.3322/caac.21219
6. Haddy TB, Mosher RB, Reaman GH. Late effects in long-term survivors after treatment for childhood acute leukemia. Clin Pediatr (Phila) 2009;48:601-8. https://doi.org/10.1177/0009922809332680
7. Akyay A, Olcay L. Çocuklarda akut lenfoblastik lösemi tedavisinin erken ve geç yan etkileri. Turkish J Pediatr Dis 2014;1:46-54. https://doi.org/10.12956/tjpd.2014.43
8. Howard SC, Ribeiro RC, Pui C-H. Acute complications. in: Pui C-H, editor. Childhood Leukemias. 3rd ed. Cambridge University Press 2016: 660-700. https://doi.org/10.1017/CBO9780511977633.030
9. ALL IC-BFM 2009. A Randomized Trial of the I-BFM-SG for the Management of Childhood non-B Acute Lymphoblastic Leukemia Final Version of Therapy Protocol from August-14-2009.
10. Baytan B, Evim MS, Güler S, Güneş AM, Okan M. Acute central nervous system complications in pediatric acute lymphoblastic leukemia. Pediatr Neurol 2015;312-8. https://doi.org/10.1016/j.pediatrneurol.2015.03.006
11. Nazir HF, AlFutaisi A, Zacharia M, Elshinawy M, Mevada ST, Alrawas A, et al. Vincristine-induced neuropathy in pediatric patients with acute lymphoblastic leukemia in Oman: Frequent autonomic and more severe cranial nerve involvement. Pediatr Blood Cancer 2017;64:1-5. https://doi.org/10.1002/pbc.26677

12. Hershman DL, Till C, Wright JD, Awad D, Ramsey SD, Barlow W E, et al. Comorbidities and Risk of Chemotherapy-Induced Peripheral Neuropathy Among Participants 65 Years or Older in Southwest Oncology Group Clinical Trials. J Clin Oncol 2016;34:3014-22. https://doi.org/10.1200/JCO.2015.66.2346

13. Aytaç S, Yetgin S, Tavil B. Acute and long-term neurologic complications in children with acute lymphoblastic leukemia. Turk J Pediatr 2006;48:1-7.

14. Diouf B, Crews KR, Lew G, Pei D, Cheng C, Bao J, et al. Association of an inherited genetic variant with vincristine-related peripheral neuropathy in children with acute lymphoblastic leukemia. JAMA 2015;313:815-23. https://doi.org/10.1001/jama.2015.0894

15. Wright GE, Amstutz U, Drögemöller BI, Shih J, Rassekh SR, Hayden MR, et al. Pharmacogenomics of vincristine-induced peripheral neuropathy implicates pharmacokinetic and inherited neuropathy genes. Clin Pharmacol Ther 2019;105:402-10.https://doi.org/10.1002/cpt.1179

16. Winocur G, Wojtowicz JM, Huang J, Tannock Ian F. Physical exercise prevents suppression of hippocampal neurogenesis and reduces cognitive impairment in chemotherapy-treated rats. Psychopharmacology 2014;231:2311-20. https://doi.org/10.1007/s00213-013-3394-0

17. Baytan B, Özdemir Ö, Demirkaya M, Evim MS, Gunes AM. Reversible posterior leukoencephalopathy induced by cancer chemotherapy. Pediatr Neurol 2010;43:197-201. https://doi.org/10.1016/j.pediatrneurol.2010.05.005

18. Bilir ÖA, Dikme G, Malbora B, Evim MS, Siviş ZÖ, Tüfekçi Ö, et al. Posterior reversible encephalopathy syndrome in childhood hematological/oncological diseases: Multicenter results. J Pediatr Hematol Oncol 2020;43:462-5. https://doi.org/10.1097/MPH.0000000000001965

19. Tang J-H, Tian J-M, Sheng M, Hu S-Y, Li Y, Zhang L-Y, et al. Study of posterior reversible encephalopathy syndrome in children with acute lymphoblastic leukemia after induction chemotherapy. J Child Neurol 2016;31:279-84. https://doi.org/10.1177/0883073815589758

20. Jeon J-S, Park S-P, Seo J-G. Posterior reversible encephalopathy syndrome due to hyponatremia. J Epilepsy Res 2014;4:31-3. https://doi.org/10.14581/jer.14008

21. Ranta S, Tuckuviene R, Makipernaa A, Albertsen BK, Frisk T, Tedgard U, et al. Cerebral sinus venous thromboses in children with acute lymphoblastic leukaemia - a multicentre study from the Nordic Society of Paediatric Haematology and Oncology. Br J Haematol 2015;168:547-52. https://doi.org/10.1111/bjh.13162

22. Ghanem KM, Dhayni RM, Al-Aridi C, Tarek N, Tamim H, Chan AKC, et al. Cerebral sinus venous thrombosis during childhood acute lymphoblastic leukemia therapy: Risk factors and management. Pediatr Blood Cancer 2017;64:1-6. https://doi.org/10.1002/pbc.26694

23. dos Santos, AC, dos Santos, JMB, da Costa Lima, E, Land, MGP. L-asparaginase doses number as a prognostic factor in childhood acute lymphoblastic leukemia: A survival analysis study. Cancer Reports 2022; 5:e1533. https://doi.org/10.1002/cnr2.1533

24. Gillson N, Jones C, Reem RE, Rogers DL, Zumberge N, Aylward SC. Incidence and demographics of pediatric intracranial hypertension. Pediatr Neurol 2017;73:42-7. https://doi.org/10.1016/j.pediatrneurol.2017.04.021

25. Peddi PF, Peddi S, Santos ES, Morgensztern D. Central nervous system toxicities of chemotherapeutic agents. Expert Rev Anticancer Ther 2014;14:857-63. https://doi.org/10.1586/14737140.2014.911089

26. Fernández-García MÁ, Cantarin-Extremera V, Andion-Catalan M, Duat-Rodriguez A, Jimenez-Echevarria S, Bermejo-Arnedo I et al. Secondary intracranial hypertension in pediatric patients with leukemia. Pediatr Neurol 2017;77:48-53. https://doi.org/10.1016/j.pediatrneurol.2017.08.013

Turkish Journal of Pediatric Disease

Original Article

Environmental tobacco exposure linked to lower oxygen saturation in infants with community-acquired lower respiratory tract infections

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ABSTRACT

Objective: Tobacco smoke is a pervasive environmental hazard, particularly detrimental to the developing respiratory systems of infants. Exposure to environmental tobacco smoke (ETS) has been consistently implicated in the etiology of a spectrum of lower respiratory tract infections (LRTIs), which are a leading cause of morbidity in children under two years of age. The pathophysiological impact of ETS extends beyond exposure; it actively exacerbates the severity of respiratory conditions, often resulting in increased hospital admissions and prolonged medical care for the youngest and most vulnerable.

This study aimed to explore the relationship between exposure to ETS and the severity of clinical manifestations, laboratory findings, and hospitalization duration in infants with community-acquired LRTIs.

Material and Methods: A cohort of 115 infants aged 1–24 months, hospitalized due to community-acquired LRTIs and without prematurity or chronic diseases, was evaluated. Data on household tobacco use were collected, and infant cotinine levels were measured to assess the impact of ETS on the severity of LRTIs.

Results: Findings revealed that the frequency of urinary cotinine positivity is significantly higher in infants from households with smokers (p=0.001). Among patients with household smoking, the proximity of tobacco consumption to the child did not affect the frequency of cotinine positivity (p=0.501). Notably, the cotinine-positive group had significantly lower oxygen saturation at admission (p=0.038). In the RSV-positive subgroup, this association remained significant (p=0.015), providing stronger evidence that ETS independently exacerbates respiratory distress.

Conclusion: This study demonstrated that tobacco exposure is associated with increased respiratory distress in infants with lower respiratory tract infections. Emphasizing the importance of smoke-free environments during infancy, it also proved the negative effects of not only secondhand smoke but also thirdhand "surface" smoke exposure on infants' respiratory health.

Keywords: Environmental tobacco smoke pollution, Infant health, Oxygen saturation, Respiratory tract infections, Secondhand smoke

INTRODUCTION

Childhood pneumonia remains a critical public health issue, particularly in developing nations, where it accounts for 14% of deaths among children under five years of age (1). Exposure to environmental tobacco smoke (ETS) is a recognized risk factor exacerbating respiratory conditions, particularly in infants under two years (2, 3).

ETS comprises two key components: secondhand smoke (SHS) and thirdhand smoke (THS). SHS refers to the inhalation of airborne tobacco pollutants. Due to their smaller lung capacity and higher respiratory rates, children are more rapidly and significantly affected by these airborne pollutants. In contrast, THS represents residual pollutants left on surfaces and fabrics, such as clothing, furniture, and toys, after tobacco consumption. Children are particularly vulnerable to THS because of behaviors like playing on surfaces, touching objects, and exploring their environment by putting their hands and objects into their mouths (4, 5).

ETS exposure, encompassing both SHS and THS, has been associated with increased hospitalizations for respiratory illnesses, recurrent wheezing, and chronic cough (6-8). The global burden of SHS remains significant, with nearly one-third of the world's population exposed and approximately

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600.000 premature deaths annually attributable to SHS (3, 9). The inclusion of THS in the concept of ETS underscores the pervasive nature of exposure, revealing that the risk extends far beyond direct inhalation and includes contact with contaminated environments (10, 11).

The mechanisms underlying these adverse effects involve complex interactions. SHS exposure in children has been linked to oxidative stress, increased airway inflammation, and disruption of pulmonary defense mechanisms, compounding their susceptibility to respiratory infections (12). Cotinine, a stable nicotine metabolite, serves as a reliable biomarker to quantify ETS exposure. Elevated cotinine levels correlate strongly with respiratory morbidities, including reduced oxygen saturation, heightened airway reactivity, and increased susceptibility to infections (13).

While numerous studies have examined the relationship between ETS exposure and the incidence of acute lower respiratory tract infections (LRTIs), there is a notable gap in understanding how such exposure influences disease severity. This study aimed to address this gap by investigating the impact of ETS exposure on clinical outcomes, including oxygen saturation, hospitalization duration, and laboratory findings, in infants with community-acquired LRTIs.

MATERIALS and METHODS

Study population and design

This prospective observational study included patients aged 1–24 months admitted with community-acquired lower respiratory tract infections to Dr. Sami Ulus Pediatric Training and Research Hospital between December 2012 and February 2013. Exclusion criteria comprised a history of prematurity and chronic diseases such as congenital heart disease, chronic renal or pulmonary conditions, neurologic developmental delay, significant malnutrition, obesity, or immunodeficiency.

Data collection

Informed consent was obtained from the caregivers upon admission, followed by a structured interview using a standardized questionnaire. This questionnaire captured demographic data (age, gender), environmental factors (smoking exposure within the home, relationship of the smoker to the patient, and the number of cigarettes smoked daily in the home), and clinical parameters. A urine specimen was collected on admission day for cotinine analysis, which were stored at -20°C until analysis.

Laboratory analysis

Thawed urine samples were centrifuged at 3000 rpm for 5 minutes. Cotinine levels were quantitatively measured using the solid-phase competitive chemiluminescence immunoassay on an Immulite 2000 Analyzer Nicotine Metabolite device by Siemens, USA, as detailed by Florescu et al. (4). The cotinine threshold to differentiate between exposure and non-exposure was set at ≥10 ng/ml, based on manufacturer recommendations and corroborated by previous literature (5).

Statistical analysis

Statistical procedures were conducted using IBM SPSS Statistics for Windows, version 22 (IBM Corp., Armonk, N.Y., USA). Shapiro-Wilk test assessed the normality of data distribution. The Mann-Whitney U test analyzed non-normally distributed variables, and Pearson's chi-square test assessed categorical variables. Descriptive statistics are presented as mean, standard deviation, minimum and maximum for continuous variables and as frequencies and percentages for categorical variables. A p-value < 0.050 denoted statistical significance.

RESULTS

Among the initial 141 cases, 115 completed the questionnaire and provided urine samples. The average age of the 115 patients participating in the study was 6.13±5.75 months (1–23 months). Of these, 80 were male (69.6%). The questionnaire responses identified 86 patients (74.8%) as having exposure to environmental tobacco.

Urinary cotinine was positive in 40% of cases (n=46), and undetectable in 60% (n=69). When considering the daily number of cigarettes to which subjects were exposed, the frequency of urinary cotinine positivity increased with the number of cigarettes smoked daily. In particular, 53.4% of infants from households that smoked more than 20 cigarettes a day had positive cotinine levels. On the other hand, 33.3% of infants in households with 1-10 smokers per day and 10.3% of infants

Table I: The relationship between presence of continine in urine and the number of cigarettes consumed around infants

Cigarette consumption per day	Cotinine* (-)	Cotinine* (+)	p†
Total	69 (60)	46 (40)	
No consumption	26 (89.7)	3 (10.3)	
1-10	6 (66.7)	3 (33.3)	0.001
11-20	10 (52.6)	9 (47.4)	
>20	27 (46.6)	31 (53.4)	

**: n(%), †: Pearson Chi-Square test*

Table II: The relationship between smoking area and the presence of cotinine in urine in cases with cigarette exposure in the study questionnaire

Consumption area	Cotinine* (-)	Cotinine* (+)	p†
Total	43 (50)	43 (50)	
Next to	4 (40)	6 (60)	0.501
Other room / Balcony / Outside	39 (51.3)	37 (48.7)	

**: n(%), †: Pearson Chi-Square test*

Table III: The relationship between clinical parameters and presence of continine in urine

	Cotinine* (-)	Cotinine* (+)	p†
Oxygen saturation	94 (80-100)	93 (65-98)	0.038
Respiratory rate	60 (20-80)	60 (40-84)	0.719
CRP (mg/L)	9.7 (1-144)	15 (1-172)	0.214
Leucocyte count/mm^3	12100 (4200-25400)	10800 (10000-23800)	0.219
Hospitalization, days	6 (3-14)	6 (4-15)	0.819

**: median (min-max), †: Mann-Whitney U test*

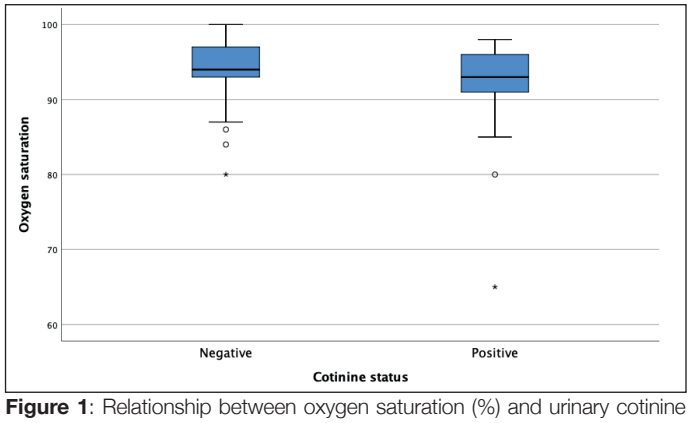

Figure 1: Relationship between oxygen saturation (%) and urinary cotinine in infants hospitalized due to community acquired lower respiratory tract infection (n = 115, p = 0.038, "○" signs for outliers, "*" signs for extreme values)

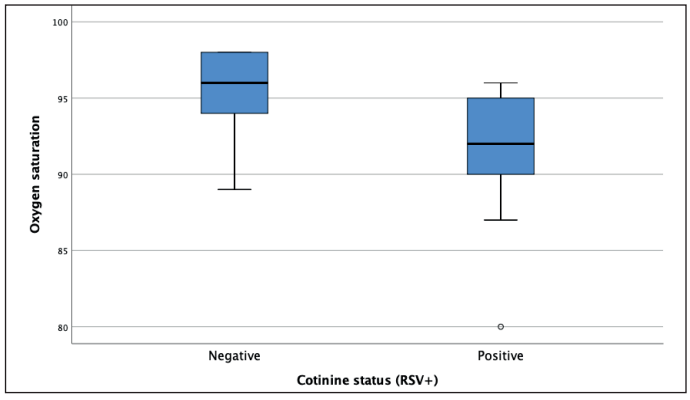

Figure 2: Relationship between oxygen saturation (%) and urinary cotinine in infants hospitalized due to RSV positive community acquired lower respiratory tract infection (n = 24, p = 0.015, "○" signs for an outlier)

in non-smoking households were positive for cotinine in urine (n=115, p=0.001; Table I).

Smoking sites were reported as follows: 11.6% (n=10) in the same room as the baby, 60.4% (n=52) on the balcony, or outside, and 28% (n=24) in another room. The frequency of cotinine positivity did not vary significantly based on the site of cigarette consumption. For example, 60% of infants from households where smoking occurred in the same room tested positive for cotinine, compared to 48.7% in households where smoking occurred outside, on the balcony or in another room (n=86, p=0.501; Table II).

Among all participants, infants with positive urinary cotinine levels had a lower oxygen saturation at admission (p=0.038; Table III, Figure 1). In contrast, no significant differences were found in respiratory rate (p=0.719), C-reactive protein (CRP) level (p=0.214), leukocyte count (p=0.219), or length of hospital stay (p=0.819) between cotinine-positive and cotinine-negative groups (Table III).

Forty-seven participants underwent viral PCR analysis of nasal swab samples, with 24 testing positive for Respiratory Syncytial Virus (RSV). In this RSV-positive subgroup, oxygen saturation levels at admission were significantly lower in the cotinine-positive group compared to the cotinine-negative group (p=0.015; Figure 2). No significant differences were observed in respiratory rate, CRP level, leukocyte count, or length of hospital stay between these groups.

DISCUSSION

Our findings reveal ETS exposure is associated with higher frequency of urinary cotinine positivity. Infants with positive cotinine levels had significantly lower oxygen saturation upon admission for community-acquired LRTIs. This association persisted even in the RSV-positive subgroup, where cotinine-positive group again exhibited lower oxygen saturation at admission. The confirmation of this result in the RSV-positive subgroup underscores the impact of tobacco smoke exposure on respiratory outcomes, independent of the infecting agent. These findings further strengthen the evidence that ETS exposure exacerbates respiratory distress in vulnerable populations.

The high prevalence of ETS exposure (74.8%) in our cohort mirrors findings from other studies in Turkey, highlighting a cultural pattern of tobacco use that significantly exceeds reported exposure rates in Europe and North America (9, 14). Such widespread exposure underscores the urgent need for tailored public health interventions to reduce tobacco use and mitigate its impacts on children. Moreover, the observed relationship between household tobacco consumption and positive cotinine levels reinforces the utility of cotinine as a biomarker for ETS exposure and its role in assessing associated health risks (13, 15). The discrepancy between self-reported exposure rates and cotinine levels may be explained by the interval of hospitalization during which the subjects spent time in a relatively smoke-free environment.

SHS exposure has been consistently associated with respiratory morbidities, including increased emergency department visits, recurrent wheezing, and chronic cough in children (7, 16). In our study, the frequency of cotinine positivity did not significantly differ based on the proximity of smoking to the child, indicating that even indirect exposure can lead to measurable biological effects. This finding highlights the role of a relatively new

concept, 'thirdhand smoke exposure,' which refers to the persistence of tobacco smoke residues on surfaces and fabrics. Children's natural behaviors, such as frequent hand-to-mouth activities and close contact with contaminated surfaces, make them particularly vulnerable to THS (4, 5). These findings align with emerging evidence that residual tobacco pollutants pose a significant risk not only through inhalation but also through contact with contaminated surfaces, even when smoking occurs away from children (4, 17). Additionally, non-airborne pollutants can persist long after active smoking has ceased, making it nearly impossible to avoid exposure even with ventilation or air conditioning (10). Public health campaigns should expand their focus to include education about THS and advocate for stricter smoke-free policies to protect children from both SHS and THS.

The finding that the cotinine-positive group had lower oxygen saturation at hospital admission suggests that ETS exposure exacerbates respiratory dysfunction in infants. Components of tobacco smoke are known to impair mucociliary clearance, increase oxidative stress, and trigger inflammatory responses in the respiratory tract, all of which can compound the severity of infections like RSV (12, 18, 19).

Our study contributes to the growing body of evidence linking ETS exposure to worsened clinical outcomes in infants with LRTIs. By utilizing cotinine as a biomarker, we were able to objectively measure ETS exposure, reducing reliance on potentially biased self-reported data. However, the discrepancy between self-reported exposure rates and cotinine levels highlights challenges in accurately quantifying ETS. Future research should adopt standardized questionnaires and explore additional environmental and genetic factors that may mediate the impact of ETS (20).

CONCLUSION

This study highlights the profound impact of ETS exposure on infant respiratory health, emphasizing the critical need for smoke-free environments. The link between positive cotinine levels and reduced oxygen saturation highlights the need for public health efforts to prevent tobacco exposure, especially in homes with infants. By addressing the risks of both SHS and THS, comprehensive strategies can help safeguard children from the short- and long-term health consequences of tobacco smoke exposure.

Ethics committee approval

This study was conducted in accordance with the Helsinki Declaration Principles. The study protocol was approved by the Ethics Committee of T.C. Ministry of Health Zekai Tahir Burak Hospital Gynecology and Obstetrics (11.12.2012/84).

Contribution of the authors

Altuntaş C: Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments, Taking responsibility in logical interpretation and conclusion of the results, Taking responsibility in necessary literature review for the study, Taking responsibility in the writing of the whole or important parts of the study, Reviewing the article before submission scientifically besides spelling and grammar. ***Şenel S:*** Constructing the hypothesis or idea of research and/or article, Planning methodology to reach the conclusions, Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in logical interpretation and conclusion of the results, Reviewing the article before submission scientifically besides spelling and grammar ***Zorlu P:*** Constructing the hypothesis or idea of research and/or article, Planning methodology to reach the conclusions, Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in logical interpretation and conclusion of the results, Reviewing the article before submission scientifically besides spelling and grammar.

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Conflict of interest

The authors declare that there is no conflict of interest.

REFERENCES

1. WHO. Pneumonia 2020. Available from: https://www.who.int/news-room/fact-sheets/detail/pneumonia.
2. Dybing E, Sanner T. Passive smoking, sudden infant death syndrome (SIDS) and childhood infections. H um Exp Toxicol 1999;18:202-5. https://doi.org/10.1191/096032799678839914
3. Kaur J, Upendra S, Barde S. "Inhaling hazards, exhaling insights: a systematic review unveiling the silent health impacts of secondhand smoke pollution on children and adolescents. Int J Environ Health Res 2024;34:4059-73. https://doi.org/10.1080/09603123.2024.2337837
4. Lee J, Kim HB, Jung HJ, Chung M, Park SE, Lee KH, et al. Protecting our future: environmental hazards and children's health in the face of environmental threats: a comprehensive overview. Clin Exp Pediatr 2024;67:589-98. https://doi.org/10.3345/cep.2023.01578
5. Johns Hopkins Medicine. The Impact of Thirdhand Smoke on Kids. Available from: https://www.hopkinsmedicine.org/health/wellness-and-prevention/the-impact-of-thirdhand-smoke-on-kids.
6. Asharam K, Mitku AAA, Ramsay L, Jeena PM, Naidoo RN. Environmental exposures associated with early childhood recurrent wheezing in the mother and child in the environment birth cohort: a time-to-event study. Thorax 2024;79:953-60. https://doi.org/10.1136/thorax-2023-221150
7. Snodgrass AM, Tan PT, Soh SE, Goh A, Shek LP, van Bever HP, et al. Tobacco smoke exposure and respiratory morbidity in young children. Tob Control 2016;25:e75-e82. https://doi.org/10.1136/tobaccocontrol-2015-052383
8. Zheng K, Tang L, Wang X, Chen L, Zhao Y, Chen X. The risk factors for chronic cough in children: A meta-analysis covering five continents. Respir Med 2024;232:107752. https://doi.org/10.1016/j.rmed.2024.107752
9. National Center for Chronic Disease Prevention and Health Promotion Office on Smoking and Health. The health consequences

of smoking-50 years of progress: a report of the surgeon general. Atlanta (GA): Centers for Disease Control and Prevention (US); 2014.

10. Leung LT, Ho SY, Wang MP, Lam TH. Secondhand Smoke From Multiple Sources, Thirdhand Smoke and Respiratory Symptoms in Hong Kong Adolescents. Nicotine Tob Res 2018;20:192-8.
11. Sureda X, Fernandez E, Lopez MJ, Nebot M. Secondhand tobacco smoke exposure in open and semi-open settings: a systematic review. Environ Health Perspect 2013;121:766-73. https://doi.org/10.1289/ehp.1205806
12. Torun E, Kahraman FU, Goksu AZ, Vahapoglu A, Cakin ZE. Serum catalase, thiol and myeloperoxidase levels in children passively exposed to cigarette smoke. Ital J Pediatr 2019;45:59. https://doi.org/10.1186/s13052-019-0652-8
13. Florescu A, Ferrence R, Einarson T, Selby P, Soldin O, Koren G. Methods for quantification of exposure to cigarette smoking and environmental tobacco smoke: focus on developmental toxicology. TTher Drug Monit 2009;31:14-30. https://doi.org/10.1097/FTD.0b013e3181957a3b
14. Inci G, Baysal SU, Şişman AR. Exposure to environmental tobacco smoke by healthy children aged below five (Preliminary study). Turk Pediatri Ars 2018;5:37-44. https://doi.org/10.5152/TurkPediatriArs.2018.5963
15. Hassanzad M, Khalilzadeh S, Eslampanah Nobari S, Bloursaz M, Sharifi H, Mohajerani SA, et al. Cotinine level is associated with asthma severity in passive smoker children. Iran J Allergy Asthma Immunol 2015;14:67-73. https://doi.org/10.1183/13993003.congress-2015.PA1874
16. Rice JL, Collaco JM, Tracy MC, Sheils CA, Rhein LM, Popova AP, et al. Parental Report of Indoor Air Pollution Is Associated with Respiratory Morbidities in Bronchopulmonary Dysplasia. J Pediatr 2024;275:114241. https://doi.org/10.1016/j.jpeds.2024.114241
17. Vevon B, Temples HS. Secondhand Smoke Exposure in Pediatric Patients: What Is the Nurse Practitioner's Role? J Pediatr Health Care 2024;38:936-42. https://doi.org/10.1016/j.pedhc.2024.01.003
18. Maedel C, Kainz K, Frischer T, Reinweber M, Zacharasiewicz A. Increased severity of respiratory syncytial virus airway infection due to passive smoke exposure. Pediatr Pulmonol 2018;53:1299-306. https://doi.org/10.1002/ppul.24137
19. Jing W, Wang W, Liu Q. Passive smoking induces pediatric asthma by affecting the balance of Treg/Th17 cells. Pediatr Res 2019;85:469-76. https://doi.org/10.1038/s41390-019-0276-0
20. Blanco-Ferreiro A, Teijeiro A, Varela-Lema L, Rey-Brandariz J, Candal-Pedreira C, Martin-Gisbert L, et al. Assessment of exposure to secondhand tobacco smoke in Spain: A scoping review. Tob Induc Dis 2024;22. doi: 10.18332/tid/192118. eCollection 2024.

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Original Article

Attitude and barriers towards epilepsy surgery: a survey among pediatricians and pediatric residents in Türkiye

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ABSTRACT

Objective: The aim of this study was to assess knowledge and attitude towards epilepsy surgery among pediatricians and identify the barriers to referral for epilepsy surgery in Türkiye.

Material and Methods: There were 21 statements which included the following: (A) knowledge (B) attitude and (C) barriers towards epilepsy surgery. The survey was mailed to 368 pediatricians.

Results: Among responders (n=240); 56.6% (n=136) were pediatricians and 44.4% (n=104) were pediatric residents. Three quarters (76 %) of them had experiences in neurology department and 60.1 % of them encountered ≥30 epilepsy patients per month. Most of participants who had no idea whether epilepsy surgery is one of the treatment options were residents (p=0.046). Almost all responders (97.5%) agreed to consult a DRE (Drug Resistant Epilepsy) patient to a pediatric neurologist for medical options. Nearly half of them (43.2%) had no idea about long-term positive cognitive effects. Whereas one-third of participants stated that it is not a safe process; more than half (57%) reported not knowing where and when to refer these patients.

Conclusion: An important finding was the apparent lack of inadequate knowledge of long-term benefits and the specificity of epilepsy surgery although most of them had previous experience in neurology department. Besides the lack of epilepsy surgery centers, lack of communication is also a problem in planning the referral of patients.

Keywords: Drug Resistant Epilepsy, Pediatrics, Survey

INTRODUCTION

Epilepsy is one of the most common chronic neurological diseases worldwide and approximately one-third of these patients have seizures that do not respond to antiseizure medication therapy (1). Neurobiological aspects of epilepsy are unique to children and therefore epilepsy surgery is also an alternative safe and effective treatment option in children (2,3). Timely intervention can reduce seizures and epileptiform discharges allowing reduction in antiseizure medication (4). Additionally, successful surgical interventions have shown significant improvements in cognitive development, behavior, and overall quality of life for affected children (5).

In our country, pediatric neurologists are primarily responsible for epilepsy patients, but pediatricians encounter with epilepsy patients more frequently due to vaccination and other health issues. In addition, since epileptic patients frequently present to pediatric outpatient clinics and emergency departments due to seizures in daily practice, pediatricians should be aware of alternative non-drug approaches in epilepsy. As far as we know, few published papers have examined these issues among neurologists or primary care doctors in the literature, and to our current knowledge there is no study from Türkiye. The aim of this study was to collect information about the domains of attitude and barriers of Turkish pediatricians and pediatric residents regarding epilepsy surgery.

MATERIALS and METHODS

The survey was prepared through the questions/statements used in previously published similar studies (6,7). A final modification was done incorporating suggestions of the experts in this field. The statements were designed by the authors considering the expected level of knowledge required for pediatricians for timely treatment and referral of epilepsy patients. After arriving on

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consensus, relevance of the statements was judged independently by 3 experts (pediatric neurologists). The distribution of statements are as follows: four of them about demographic variables, nine of them (Statement 5,6,7,8,9,10,18,19,20) were about knowledge, seven of them (Statement 11,12,13,14,15,16,17) were about attitudes and the last question was about barriers towards epilepsy surgery. Out of the 21 statements, 5 to 20 were codified as agree, disagree or no opinion. and the last statement regarding barriers in practice was open ended. To ensure that all statements were answered, the participants were not allowed to move to the next one before answering the current one.

The study was a cross-sectional, descriptive, questionnaire- based study which was conducted among pediatricians and pediatric residents from Türkiye between October 2, 2022, and November 2, 2022, with the approval of the Institutional Ethics Committee from the Turkish Ministry of Health, Ankara Bilkent City Hospital Ethics Committee-2 (E2-22-1221/02.02.2022). A total of 368 pediatricians were requested to participate in the study by sending questionnaires through e-mails and 240 responded (response rate 65%). Informed consent was obtained online by adding the informed consent form to the survey prepared via Google forms in accordance with the Helsinki guidelines.

Statistical analysis:

All data were tabulated in Microsoft Excel® format and analyzed through the statistical software IBM SPSS Statistics for Windows, version 25 (IBM Corp., Armonk, N.Y., USA). Frequency and percentage were utilized as descriptive statistics. In addition, Chi square tests were done to assess whether demographics (specialty, years in practice, number of patients with epilepsy encountered per month) influenced responders' knowledge and attitude towards epilepsy surgery. p value <0.050 was accepted as significant.

RESULTS

In total, 240 of 306 pediatricians returned the questionnaire (response rate 78%). Of the participants 56.6% (n=136) were pediatricians and 44.4% (n=104) were pediatric residents. Half of the responders (49.2%) had experience of more than five years with pediatric patients. Three quarters (76%) of them had previous experience in neurology department during pediatric residency and 60.1% of them encountered more than 30 epilepsy patients on average per month. The results are summarized in Table I.

Most of participants (59.4%) who had no idea whether epilepsy surgery is one of the treatment options were residents (p=0.046). Almost 90% of responders agreed with the definition of drug resistant epilepsy (DRE) whereas the majority of them thought that only a few DRE patients can be treated with epilepsy surgery. Moreover, when we evaluated the statement about long-term outcomes, almost half of them had no idea about the positive effects of epilepsy surgery on cognitive status. This result was similar between groups and was not associated with the duration of pediatric practice (p=0.716). Approximately 92% of pediatricians

Table I: Demographic characteristics of responders

Characteristics	Values*
Total	240
Pediatricians	136 (56.6)
Pediatric residents	104 (44.4)
Years in pediatric practice	
<2 year	45 (18.8)
2-5 year	77 (32)
≥5 year	118 (49.2)
Number of children with epilepsy attended per month	
<10	36 (15)
10-30	59 (24.9)
≥30	145 (60.1)
Previous experience in neurology department	
Yes	183 (76)
No	57 (24)

*: *n(%)*

stated that DRE patients could remain seizure-free after epilepsy surgery, while this rate was 61.5% among pediatric residents (p=0.022). The participants with pediatric neurology experience (82%) were more likely to agree with this statement (p=0.005). One third of the participants (32.9%) had no idea about how to manage antiseizure medication after epilepsy surgery. Table II lists the statements and responses related to knowledge about epilepsy and epilepsy surgery.

Almost all (97.5%) responders recommended that DRE patients should be consulted to a pediatric neurologist for medical options. In response to the statement regarding the use of medications before referral for epilepsy surgery, 42% of the participants agreed to trying more than two antiseizure medications before referral for surgery. Interestingly, most of participants stated that only a few DRE patients can be treated with epilepsy surgery. There was no significant difference between pediatricians and pediatric residents in their response to this statement (p=0.058). Those with experience in pediatric neurology department agreed with the statement that DRE patients could remain seizure free after epilepsy surgery. Most pediatricians (92 %) considered that DRE patients could remain seizure free after epilepsy surgery. Pediatric residents had less information about this subject (p=0.022). Another notable finding was that half of our participants considered it was a long-lasting procedure and one-third stated that it was not a safe treatment option and there was no significant difference between pediatricians according to duration in pediatric practice. Figure 1 shows the responses related to attitude towards epilepsy surgery.

More than half of the participants (57%) had no idea where and when to refer patients with DRE. Almost 29% of the participants stated that there is no epilepsy surgery center to refer these patients, however just two percent of responders thought it was not cost-effective. There was no difference between the two groups in regards of this question (p=0.165). Statements concerning the barriers towards epilepsy surgery are summarized in Figure 2.

Table II: Statements and responses related to knowledge about epilepsy and epilepsy surgery

Statement	Pediatricians*	Pediatric residents*	p†
5. Epilepsy and seizure are synonymous			
Agree	13 (9)	9 (8.7)	0.939
Disagree	123 (91)	95 (91.3)	
No opinion	0	0	
6. Epilepsy surgery is one of the treatment options for epilepsy patients			
Agree	117 (86)	65 (62.5)	0.046
Disagree	15 (10)	22 (21.2)	
No opinion	4 (3)	17 (16.3)	
7. Failure of adequate trials of two appropriately chosen antiseizure drugs can be considered as DRE.			
Agree	122 (90.2)	89 (85.6)	0.459
Disagree	10 (6.8)	12 (11.5)	
No opinion	4 (3)	3 (2.9)	
8. A child with epilepsy can die during a seizure.			
Agree	131 (96.3)	99 (95.1)	0.432
Disagree	5 (3.6)	5 (4.8)	
No opinion	0	0	
9. Epilepsy surgery is not indicated in all children with DRE.			
Agree	127 (94)	97 (94.2)	0.951
Disagree	0	1 (0.01)	
No opinion	9 (6.0)	6 (5.8)	
10. Only few DRE patients can be treated with epilepsy surgery			
Agree	128 (97)	94 (91.3)	0.058
Disagree	4 (3)	1 (0.01)	
No opinion	4 (3)	9 (8.7)	
18. After epilepsy surgery, anti-seizure medication may be discontinued in patients.			
Agree	81 (59.5)	55 (52)	0.107
Disagree	18 (13.3)	7 (6.7)	
No opinion	37 (27.2)	42 (40.3)	
19. Epilepsy surgery is associated with improvement in long-term cognitive outcomes			
Agree	75 (55)	43 (41.3)	0.716
Disagree	10 (7.5)	12 (11.5)	
No opinion	51 (37.5)	49 (47.2)	
20. DRE patients could remain seizure free after epilepsy surgery			
Agree	125 (92)	64 (61.5)	0.022
Disagree	5 (3.6)	13 (12.5)	
No opinion	6 (4.4)	27 (26)	

: n(%), †: Pearson Chi-square test, **DRE: Drug resistant epilepsy*

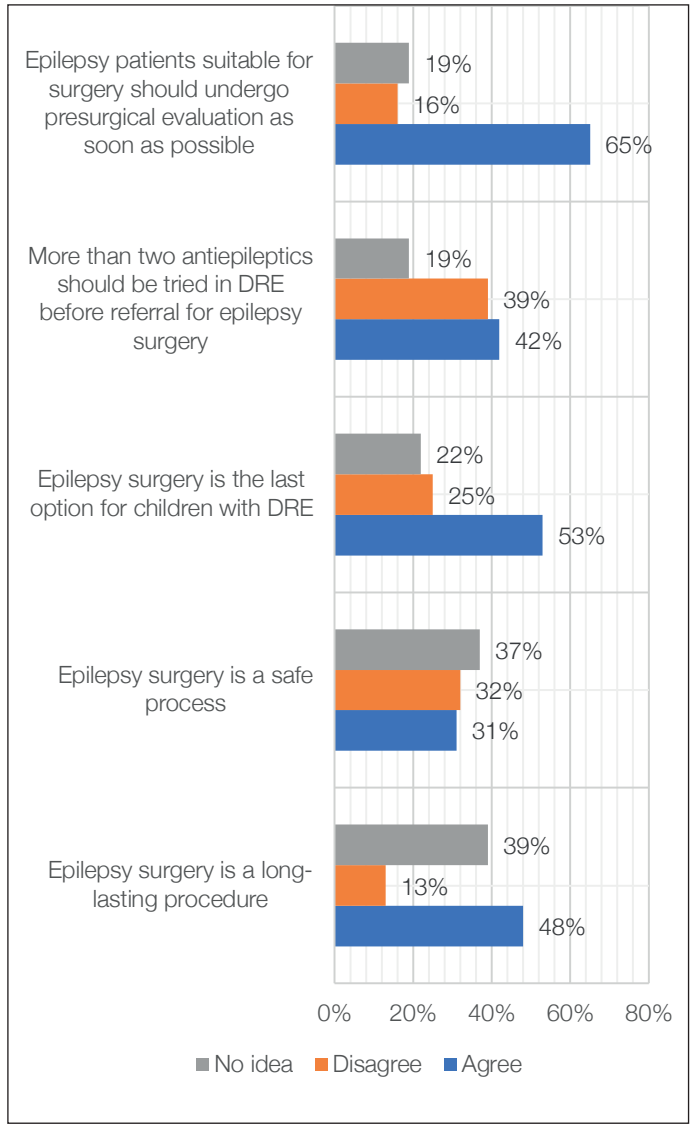

Figure 1: Responses related to attitudes towards epilepsy surgery.

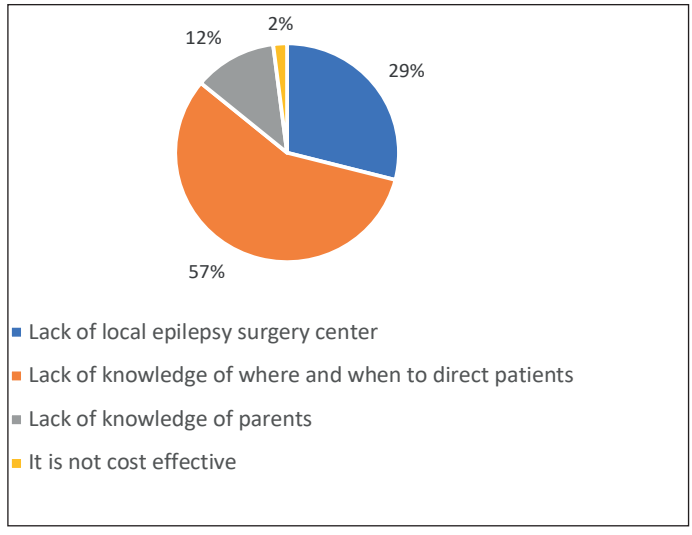

Figure 2: Statements concerning the barriers towards epilepsy surgery.

DISCUSSION

Epilepsy surgery remains one of the least preferred evidence-based treatments, despite evidence supporting better cognitive outcomes and higher cost-effectiveness compared to medical treatment of epilepsy (8,9). Referral for epilepsy surgery was demonstrated more likely to occur through familiarity with epilepsy surgery during training or clinical practice rather than number of years in clinical practice. Additionally, uncertainties about definition of DRE, poor communication and loss of follow up after surgery, expensive preoperative examinations contributed to underutilization of epilepsy surgery in developing countries (10). While analyzing the statements, we noticed that the level of knowledge of pediatricians about epilepsy surgery was inadequate, despite the fact that most of the participants practiced in the pediatric neurology department. Pediatric residents also had insufficient information about the statement of epilepsy surgery is one of the treatment options for epilepsy patients. The majority of participants correctly identified DRE, however more than half of them stated that epilepsy surgery was the last treatment option for DRE. There was also no significant association with the duration in pediatric practice in terms of the remaining seizure-free after epilepsy surgery.

Previous epilepsy treatment studies have shown that there is a need to evaluate surgical options for ongoing dysfunctional activity in drug resistant epilepsy (11-13). Researchers have also concluded that the lack of knowledge about surgical risks and benefits may be a significant barrier towards epilepsy surgery (14). According to an international survey of epilepsy surgery centers, it was stated that one third of children were operated on within two years of epilepsy onset, despite the fact that the epilepsy surgery process started in 60% of patients as young as two years of age (15). Besides advances in structural and functional neuroimaging, neurosurgery has significantly improved surgical outcomes in children in recent years. Furthermore, it was discussed that it causes fear of surgery among pediatricians due to intraoperative complications with high mortality (16). Disagreement with the idea of the safety of procedure in one thirds of participants may be a result of these approaches in our survey. Another notable finding was that pediatric residents were less aware of whether DRE patients could remain seizure-free after epilepsy surgery. This may be due to pediatricians not following up on antiseizure medication in epilepsy patients and not have sufficient knowledge about the side effects.

Healthcare providers identify inadequate healthcare access (e.g. long waiting times, limited resources, referral delays, distance) as the biggest barrier to epilepsy surgery (17,18). Also, in a recent review, barriers including lack of clinical expertise and communication are reported (19,20). There are many pediatric neurology centers in Türkiye however there are just four (two in Ankara and two in İstanbul) epilepsy surgery centers. In addition, not all neurodiagnostic methods commonly used in adult general hospitals are available in some pediatric hospitals in our country, and therefore children are referred to adult hospitals for specialized examinations such as functional MRI, positron emission tomography, magnetoencephalography, etc. Epilepsy surgery conferences to discuss the treatment options could increase communication between specialists and pediatricians and thus increase the number of patients referred for consideration of epilepsy surgery.

There are some limitations of the current study. Firstly, the generalizability of these results to other countries is limited as we reported on Turkish pediatricians and pediatric residents. Hence, the knowledge, attitude and practice of the pediatricians cannot be fully determined. Secondly, approximately half of participants had <5 years of experience in pediatric practice which may have influenced the level of knowledge about treatment options of epilepsy patients.

CONCLUSION

This is the first study based on the perspectives and decision-making processes of Turkish pediatricians regarding epilepsy surgery. Barriers towards epilepsy surgery can be explained by many factors, particularly knowledge gaps. Referral criterias should be established in collaboration of pediatric neurologists and epileptologists. Pediatricians need to periodically update their knowledge and strategies for timely identification for surgical evaluation.

Ethics committee approval

This study was conducted in accordance with the Helsinki Declaration Principles. The study was approved by Ankara Bilkent City Hospital Ethics Committee-2 (E2-22-1221).

Contribution of the authors

Dedeoğlu Ö: Contributed to conception and design, acquisition, data collection and analysis drafted manuscript and agrees to be accountable for all aspects of work ensuring integrity and accuracy and there is no any potential conflict of interest. ***Yılmaz D:*** Contributed to conception and design, acquisition and analysis, drafted manuscript and agrees to be accountable for all aspects of work ensuring integrity and accuracy and there is no any potential conflict of interest. ***Ardıçlı D:*** Contributed to preparation manuscript, critical review manuscript and agrees to be accountable for all aspects of work ensuring integrity and accuracy and there is no any potential conflict of interest. ***Gürkaş E:*** Contributed to conception and design, acquisition and analysis, and agrees to be accountable for all aspects of work ensuring integrity and accuracy and there is no any potential conflict of interest. ***Çıtak Kurt AN:*** Contributed to conception and design, acquisition and analysis, critical review manuscript and agrees to be accountable for all aspects of work ensuring integrity and accuracy and there is no any potential conflict of interest. ***Kartal A:*** Contributed to conception and design, acquisition,data collection and analysis, critical review manuscript and agrees to be accountable for all aspects of work ensuring integrity and accuracy and there is no any potential conflict of interest

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Conflict of interest

The authors declare that there is no conflict of interest.

REFERENCES

1. Dabrowski A, Armstrong C. A pediatrician's guide to epilepsy surgery. Curr Probl Pediatr Adolesc Health Care 2024;54:101578. https://doi.org/10.1016/j.cppeds.2024.101578
2. Dwivedi R, Ramanujam B, Chandra PS, Sapra S, Gulati S, Kalaivani M, et al. Surgery for Drug-Resistant Epilepsy in Children. N Engl J Med 2017;377:1639-47.https://doi.org/10.1056/NEJMoa1615335
3. Nordli Iii DR, Taha M, Freund B, Nordli DR Jr, Galan F. Minimally Invasive Epilepsy Surgery. Neuropediatrics 2024;55:279-88. https://doi.org/10.1055/s-0044-1788061
4. Ramantani G, Wirrell E. Epilepsy surgery in developmental and epileptic encephalopathies. Epilepsy Behav 2024; 59:109985. https://doi.org/10.1016/j.yebeh.2024.109985
5. Jeno M, Zimmerman MB, Shandley S, Wong-Kisiel L, Singh RK, McNamara N, et al. Pediatric Palliative Epilepsy Surgery: A Report From the Pediatric Epilepsy Research Consortium (PERC) Surgery Database. Pediatr Neurol 2024;157:70-8. https://doi.org/10.1016/j.pediatrneurol.2024.04.028
6. Zupan G, Lorber B. Knowledge and Awareness of Epilepsy Surgery among Medical Students. J Epilepsy Res 2017;7:50-3. https://doi.org/10.14581/jer.17009
7. Roberts JI, Hrazdil C, Wiebe S, Sauro K, Vautour M, Wiebe N, et al. Neurologists' knowledge of and attitudes toward epilepsy surgery: a national survey. Neurology 2015;84:159-66. https://doi.org/10.1212/WNL.0000000000001127
8. Berg AT, Loddenkemper T, Baca CB. Diagnostic delays in children with early onset epilepsy: impact, reasons, and opportunities to improve care. Epilepsia 2014;55:123-32. https://doi.org/10.1111/epi.12479
9. Aljafen B, Alomar M, Abohamra N, Alanazy M, Al-Hussain F, Alhumayyd Z, et al. Knowledge of and attitudes toward epilepsy surgery among neurologists in Saudi Arabia. Neurosciences (Riyadh) 2020;25:43-9. https://doi.org/10.17712/nsj.2020.1.20190051
10. Hale AT, Chari A, Scott RC, Helen Cross J, Rozzelle CJ, Blount JP, et al. Expedited epilepsy surgery prior to drug resistance in children: a frontier worth crossing? Brain 2022;145:3755-62. https://doi.org/10.1093/brain/awac275
11. Ryvlin P, Cross JH, Rheims S. Epilepsy surgery in children and adults. Lancet Neurol 2014;13:1114-26. https://doi.org/10.1016/S1474-4422(14)70156-5
12. Skirrow C, Cross JH, Cormack F, Harkness W, Vargha-Khadem F, Baldeweg T. Long-term intellectual outcome after temporal lobe surgery in childhood. Neurology 2011;76:1330-7. https://doi.org/10.1212/WNL.0b013e31821527f0
13. Prideaux L, Barton S, Maixner W, Harvey AS. Potential delays in referral and assessment for epilepsy surgery in children with drug-resistant, early-onset epilepsy. Epilepsy Res 2018; 143:20-6. https://doi.org/10.1016/j.eplepsyres.2018.04.001
14. Cothros N, Burneo JG, Steven DA. Knowledge and Attitudes About Epilepsy Surgery Among Family Doctors in Ontario. Can J Neurol Sci 2016;43:672-7. https://doi.org/10.1017/cjn.2016.277
15. Harvey AS, Cross JH, Shinnar S, Mathern GW. ILAE Pediatric Epilepsy Surgery Survey Taskforce. Defining the spectrum of international practice in pediatric epilepsy surgery patients. Epilepsia 2008;49:46-55. https://doi.org/10.1111/j.1528-1167.2007.01421.x
16. Pindrik J, Hoang N, Smith L, Halverson M, Wojnaroski M, McNally K, et al. Preoperative evaluation and surgical management of infants and toddlers with drug-resistant epilepsy. Neurosurg Focus 2018;45: E3. https://doi.org/10.3171/2018.7.FOCUS18220
17. de Flon P, Kumlien E, Reuterwall C, Mattsson P. Empirical evidence of underutilization of referrals for epilepsy surgery evaluation. Eur J Neurol 2010;17:619-25. https://doi.org/10.1111/j.1468-1331.2009.02891.x
18. Haneef Z, Stern J, Dewar S, Engel J Jr. Referral pattern for epilepsy surgery after evidence-based recommendations: a retrospective study. Neurology 2010;75:699-704. https://doi.org/10.1212/WNL.0b013e3181eee457
19. Hatoum R, Nathoo-Khedri N, Shlobin NA, Wang A, Weil AG, Fallah A. Barriers to epilepsy surgery in pediatric patients: A scoping review. Seizure 2022;102:83-95. https://doi.org/10.1016/j.seizure.2022.08.013
20. Drees C, Sillau S, Brown MG, Abosch A. Preoperative evaluation for epilepsy surgery: Process improvement. Neurol Clin Pract 2017;7:205-213. https://doi.org/10.1212/CPJ.0000000000000357

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Original Article

Clinical and demographic characteristics of Becker muscular dystrophy cases

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ABSTRACT

Objective: Becker Muscular Dystrophy (BMD) is a rare and progressive muscle disease characterized by mutations in the dystrophin gene. Although similar to Duchenne Muscular Dystrophy (DMD), BMD usually has a milder course. The aim of this study was to analyze the clinical and genetic profiles of BMD.

Material and Methods: Evaluations were made of 67 patients diagnosed with BMD between 2006 and 2024. Clinical findings, laboratory tests, and genetic analysis of the patients were retrospectively analyzed.

Results: The study group consisted of 67 patients with a mean age of 11.6±4.1 years and age at diagnosis of 4.6±3.3 years. A total of 7.5% of the patients had a history of consanguineous marriage, and 35.8% had a family history of BMD. The most common clinical symptoms were exercise intolerance (44.8%), fatigue (19.4%), and exercise-related pain (14.9%). Cardiac involvement was detected in 11.9% of the patients, and mutations between exons 45-55 were most frequently detected in these patients. Psychiatric problems were detected in 22.4% of patients, and 66.7% of these patients had mutations involving the exon 45 region. There was no significant difference in the clinical and laboratory findings of patients with reading frame mutations.

Conclusion: Although patients with BMD usually have milder clinical features, it was observed that cardiac and psychiatric involvement in particular may be associated with certain genetic mutations. Genetic analysis is considered to be an important tool in the diagnosis and prognosis of BMD, which may reduce the need for muscle biopsy.

Keywords: Becker muscular dystrophy, Clinical characteristics, Genetic mutations, Reading frame mutations

INTRODUCTION

Becker muscular dystrophy (BMD) is an inherited and progressive muscle disease characterized by muscle fibre degeneration and proximal muscle weakness due to a mutation in the dystrophin gene (1). The disease is clinically similar to Duchenne muscular dystrophy (DMD), but it is less common, clinical findings appear later, and the course is milder (2). BMD is caused by mutations in dystrophin protein. In the diagnosis, the evaluation of creatine kinase (CK) levels accompanied by physical examination findings compatible with clinical findings shows high sensitivity (3). Dystrophin has a recessive inheritance pattern linked to the X chromosome, although approximately 30% of cases are due to sporadic new mutations (4,5).

Studies in Europe and North America have shown that the prevalence of Duchenne muscular dystrophy ranges from one in 3500 to one in 5050 live male births, while Becker muscular dystrophy is about one-third of this prevalence (6,7).

Compared with DMD, the age of onset of symptoms in individuals with BMD is generally later, with a wide range from 5 to 60 years of age. Furthermore, the degree of clinical involvement is usually milder (8). Approximately 85% of patients with BMD have dystrophin of abnormal molecular weight, with smaller dystrophin in 80% of cases with deletions and larger dystrophin in 5% of cases with duplications. In these individuals, the amount of dystrophin is usually reduced. The remaining 15% have dystrophin of normal size but reduced quantity (9). Serum CK concentrations in patients with BMD typically rise to ≥5-fold the upper limit of normal (10).

Preserved muscle strength allows clinical distinctions to be made between BMD and DMD. People with BMD are usually able to stand until at least 16 years of age and often well into

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adulthood, with some retaining the ability to walk into old age. Furthermore, cognitive impairment, intellectual disability, behavioral disorders and contractures are less common or less severe in BMD compared to DMD (11). Cardiac involvement is often a prominent feature of the clinical picture in BMD, although muscle involvement is less severe than in DMD (12).

There is no complete cure for BMD, so treatment is based on supportive care and rehabilitation. Many clinical trials of gene therapy are still ongoing (9). BMD is a rare neuromuscular disorder and studies systematically addressing the clinical and laboratory findings of individuals with this disease are very limited in the literature. The main aim of this study was to provide new and in-depth information for a better understanding of the disease by detailing the clinical, demographic and genetic characteristics of patients diagnosed with BMD.

MATERIALS and METHODS

The study retrospectively included 67 patients with BMD who were followed up at Tepecik Training and Research Hospital Muscle Center between 2006 and 2024. Ethical approval was granted by the Tepecik Training and Research Hospital Non-Interventional Ethics Committee (Decision No: 2024/08-05, Date: 02.09.2024). BMD was diagnosed based on clinical and genetic findings. Patients with Gowers sign and Functional Ambulation Scale score of <5 were evaluated as DMD and were excluded from the study. Muscle biopsy was performed in only one patient and the biopsy revealed findings consistent with BMD, which were confirmed by genetic testing. The genetic analysis was performed using the multiplex ligation-dependent probe amplification (MLPA) method for the DMD gene. This method was utilized to investigate large deletions and duplications. Additionally, Sanger sequencing was conducted to identify the presence of nonsense mutations. These analyses confirmed the patient's genetic diagnosis and revealed findings consistent with BMD. The patients were divided into two groups: In-Frame Deletion (-) and In-Frame Deletion (+). Demographic data, clinical symptoms, physical examination findings, laboratory tests and 6-minute walk test results were analyzed.

Muscle strength assessment of the patients was performed using the Medical Research Council (MRC) manual muscle strength scale, which grades muscle strength on a scale from 0 to 5 (13). Psychiatric comorbidities such as learning disabilities, speech delay and/or speech disorders, attention deficit hyperactivity disorder (ADHD) and autism spectrum disorder (ASD) were evaluated and questioned.

The patients were divided into two groups according to the genetic mutations of with and without in-frame mutations. The clinical and laboratory characteristics of patients with and without in-frame mutations were compared.

Statistical analysis

Statistical analysis was performed using IBM SPSS Statistics for Windows, version 25 (IBM Corp., Armonk, N.Y., USA). Descriptive statistics were calculated for demographic, clinical, and laboratory data, including means, standard deviations, medians and interquartile ranges (IQR) for continuous variables, and frequencies and percentages for categorical variables. The comparison of continuous variables between two independent groups was conducted using the independent t-test or Mann-Whitney U test, depending on the distribution of data. The chi-square test or Fisher's exact test was used for the comparison of categorical variables. A p-value of <0.050 was considered statistically significant.

RESULTS

Evaluation was made of 67 patients with BMD, all of whom were male (100%). The mean age of the patients was 11.6±4.1 years (median 12, IQR 9-15 years). The mean age at diagnosis was 4.6±3.3 years (median 4, IQR 2-6 years). There was a history of consanguineous marriage in 5 (7.5%) patients, and a family history of BMD in 24 (35.8%). A maternal carrier test was conducted on the mothers of 42 patients, and carrier status was identified in 34 (80.1%) of them.

Of the total 67 patients, 45 (67.2%) were diagnosed incidentally during the evaluation of elevations in liver function tests, 20 (29.9%) were diagnosed because of a family history of muscle disease, 1 (1.5%) from complaints of fatigue, and 1 (1.5%) during the investigation of chest pain.

The mean age at diagnosis was 4.9±3.5 years for patients with an incidental diagnosis and 3.9±2.9 years for those diagnosed through family history screening. Although patients diagnosed via family history had a lower mean age at diagnosis, the difference was not statistically significant (p=0.238).

The mean age at diagnosis was 4.5±3.4 years in patients with inframe mutations and 5.3±2.9 years in out of frame patients, there was no statistical difference between the two groups (p=0.443).

The most common symptoms were exercise intolerance, fatigue, exercise-induced pain and muscle cramps, respectively. On physical examination, calf hypertrophy was found in 64 patients (95.5%), but no patient had Gowers sign. Muscle strength examination revealed normal strength (5/5) in the upper extremities of all patients. Distal muscle weakness (4/5) in the lower extremities was observed in only two patients, aged 12 and 18 years. The 6-minute walk test results of 48 patients were available and the median value for walking distance was 555 (IQR 457-589) meters (Table I).

The comorbid conditions of the BMD patients were analyzed, and muscular dystrophy-related cardiac involvement was present in 8 patients (11.9%), and psychiatric problems were

Table I: The demographic and clinical characteristics of patients with BMD

Characteristics	Values
Age (years)*	11.6±4.1 (9-15)
Age at diagnosis (years)*	4.6±3.3 (2-6)
Consanguineous marriage†	5 (7.5)
Family history of BMD†	24 (35.8)
Brother	9 (13.4)
Grandfather	1 (1.5)
Uncle	11 (16.4)
Cousin	3 (4.5)
Maternal carrier†	34 (80.1)
Neuromotor development stages	
Age of starting to walk (years)‡	1 (1-2.5)
Delay in walking†	2 (3)
Age of onset of speech (years) ‡	1 (1-5)
Delay in speech†	9 (13.4)
Clinical symptoms†	
Exercise intolerance	30 (44.8)
Early fatigue	13 (19.4)
Pain with exercise	10 (14.9)
Cramp	9 (13.4)
Leg pain	6 (9)
Physical examination findings	
Pseudohypertrophy of calf muscles†	64 (95.5)
Gowers sign	-
Scoliosis†	18 (26.9)
Distal muscle strength of the upper extremity§	67/0/0/0/0/0
Proximal muscle strength of upper extremity§	67/0/0/0/0/0
Distal muscle strength of the lower extremity§	67/0/0/0/0/0
Proximal muscle strength of lower extremity§	65/2/0/0/0/0
6 minutes walking test (m) (n=48)‖	555 (457-589)

***:** *mean±SD (Inter Quantile Range)* ,**†:** *n(%)*, **‡:** *median (min-max)* **§:** *Medical Research Council (MRC) manual muscle strength scale (5/5-4/5-3/5-2/5-1/5-0/5),* **‖:** *median (Inter Quantile Range)*

detected in 15 patients (22.4%). Psychiatric comorbidities included learning disabilities in 7 patients, speech delay and/or speech disorder in 5, ADHD in 2, and ASD in 1. Mutation involving exon 45 was detected in 10 (66.7%) of the patients with psychiatric problems, but this finding was not statistically significant (p=0.622).

Furthermore, no difference in the frequency of psychiatric problems was observed between the two groups when the upstream and downstream regions of DMD exon 45 were compared (p=0.471).

Of the 8 patients with cardiac involvement, 4 had exon 45-48, 3 had exon 45-53 and 1 had exon 45-55 deletion. The genetic mutations and comorbidities of the patients are summarized in Table II.

The patients were divided into two groups as complying and non-complying with the reading frame rule. When the clinical and laboratory findings of these groups were compared, no significant difference was found between them (Table III).

Table II: Genetic mutations and comorbidities of the patients

Mutation region (exon)	n (%)	LD	SD	ASD	ADHD	CI
45-47 deletion (+)	13 (19.4)	2/13	1/13			
45-48 deletion (+)	12 (17.9)		1/12			4/12
45-55 deletion (+)	7 (10.4)		1/7		1/7	1/7
45-53 deletion (+)	5 (7.5)	2/5				3/5
3-4 deletion (+)	3 (4.5)					
3-4 duplication (+)	3 (4.5)		2/3			
46-47 deletion (-)	2 (3)					
48-51 deletion (+)	2 (3)					
19-30 deletion (+)	2 (3)				1/2	
43-53 deletion (+)	2 (3)					
52-53 deletion(+)	1 (1.5)					
51-53 deletion (-)	1 (1.5)					
48-53 deletion (+)	1 (1.5)	1/1				
45-49 deletion (+)	1 (1.5)			1/1		
44-45 deletion (+)	1 (1.5)					
6,7,42-44 duplication (-)	1 (1.5)					
22-29 deletion (-)	1 (1.5)	1/1				
13-15 deletion (+)	1 (1.5)					
11 deletion (-)	1 (1.5)					
5-7 deletion (-)	1(1.5)					
3-5 deletion (+)	1(1.5)					
3-9 deletion (+)	1(1.5)	1/1				
2-7 duplication (+)	1(1.5)					
2 duplication(-)	1(1.5)					
Non-sense mutation	2 (3)					

***LD:** Learning disabilities,* ***SD:** Speech delay and/or speech disorder,* ***ASD:** Autism spectrum disorder,* ***ADHD:** Attention deficit hyperactivity disorder,* ***CI:** Cardiac involvement,* **(+):** *In-Frame Deletion,* **(-):** *In-Frame Deletion*

DISCUSSION

This study analyzed the genetic and clinical characteristics of patients with BMD, focusing on the association between specific genetic mutations and comorbidities. In particular, cases with out-of-frame mutations were identified and some deletions were found to be associated with cardiac or psychiatric involvement. The study also provides new insights into the phenotypic spectrum of BMD, highlighting the earliest age at which certain comorbidities are detected.

In a previous study in literature, the overall carrier frequency in mothers with carrier mutation types of DMD and BMD was found in 80 of 139 mothers (57.6%), and this carrier rate was significantly higher at 89.5% for BMD (14). In the current study,

Table III: Comparisons of the clinical and laboratory findings of BMD patients according to the reading frame rule

	In-Frame Deletion (+) (n=59)	In-Frame Deletion (-) (n=8)	Total (n=67)	p
Clinical symptoms*				
Exercise intolerance	24 (40.7)	6 (76)	30 (44.8)	0.126‡
Early fatigue	10 (16.9)	3 (37.5)	13 (19.4)	0.179‡
Pain with exercise	8 (13.6)	2 (25)	10 (14.9)	0.341‡
Cramp	8 (13.6)	1 (12.5)	9 (13.4)	0.705‡
Leg pain	6 (9)	-	6 (9)	-
Scoliosis*	14 (23.7)	4 (50)	18 (26.9)	0.127‡
Cardiac involvement*	8 (14)	-	8 (11.9)	-
Psychiatric symptoms*	14 (23.7)	1 (12.5)	15 (22.4)	0.672‡
Creatinine kinase (U/L)†				
Highest value	9343 (3400-12751)	16435 (9341-18890)	9422 (3579-13759)	0.069§
Lowest value	2590 (1464-4900)	3476 (1813-7481)	2810 (1482-4900)	0.686§

**: n(%), †: median (Inter Quantile Range), ‡:* Chi-square test, *§:* Mann-Whitney U test

the carrier rate was found to be 80.1%, which was lower than in the literature. This difference was thought to be because, for various reasons, carrier tests could not be performed on all the mothers.

Preserved muscle strength facilitates the differentiation of clinical features between BMD and DMD. BMD patients typically retain ambulation until at least 16 years of age and, in many cases, well into adulthood, with some maintaining this ability into advanced age (9). Diagnosis is most often established incidentally through the detection of elevated CK levels (15).

In the literature, the mean age of diagnosis for DMD is reported to be approximately 5 years; however, due to the milder clinical symptoms in BMD, it is generally expected to be diagnosed at a later age (1,16). In our study, the mean age at diagnosis was 5 years, which is earlier than the age typically observed in BMD patients. Age at diagnosis was lower among patients screened for a family history of the disease; however, this difference was not statistically significant. These findings underscore the critical role of CK screening and emphasize the importance of early diagnostic evaluation in patients with a positive family history.

All patients included in this study were ambulatory, with none exhibiting a positive Gowers sign. Upper extremity muscle strength was entirely preserved in all participants, while only two patients demonstrated mild proximal lower extremity weakness. This finding likely reflects the fact that the study cohort comprised individuals at an age when the initial manifestations of BMD typically emerge.

The most common symptoms in BMD are leg pain, cramping, exercise intolerance and exercise-induced pain (9,17). The most common symptoms in the current study were exercise intolerance, rapid fatigue, exercise-induced pain, cramps, and leg pain, respectively.

A small proportion of patients with dystrophinopathy do not have pathogenic variants in the coding region, which can make the mutation difficult to detect (10). In many cases, molecular genetic testing can confirm a definitive diagnosis of dystrophinopathy without the need for a muscle biopsy (9).

A muscle biopsy may be necessary to differentiate BMD and DMD and for prognostic evaluation. Immunohistochemical analyses have shown a complete absence of dystrophin in patients with DMD, whereas BMD patients have 10% to 40% of the normal amount of dystrophin or a partially functional form of the subsarcolemmal protein (1,18). In severe cases, dystrophin protein levels are usually below 10%, whereas in mild cases with dystrophin levels above 70%, symptoms may be of late onset or even remain asymptomatic (19–22). However, muscle biopsy is not current widely used because it is an invasive method. This increases the importance of studies on BMD. Identifying which mutations lead to which clinical picture and adding them to the literature may have the potential to reduce the need for biopsy.

A muscle biopsy was performed in only 1 patient in the current study, and the results were compatible with BMD. In all the patients, the diagnosis was confirmed by genetic analysis. Monaco et al.(23) first proposed a theory known as the “reading frame rule” based on patient examinations in 1988.

In-frame deletions/duplications in the DMD gene that do not alter the reading frame are generally associated with BMD phenotypes, with exceptions (23). Kesari et al. (24) observed a higher frequency of duplications, a different mutation distribution, and more exceptions to the reading frame rule in BMD patients. Additionally, in the study by Zambon et al. (25), which examined the phenotypic spectrum of dystrophinopathies associated with exon 2 duplications, 30% of the patients exhibited a BMD phenotype. These findings suggest that clinical manifestations can be heterogeneous even in cases with out-of-frame mutations. In the present study, eight patients with out-of-frame mutations were identified, further supporting these observations in the literature.

In patients with BMD, serum CK concentrations can often rise to a level ≥5-fold above the upper limit of normal (9). Consistent

with the literature, the CK levels in this study were ≥5-fold higher. Furthermore, no statistically significant difference was found between patients with and without in-frame mutations.

The most common mutations in BMD are deletions in exons 45-47 (26). In the current study, consistent with the literature, the most common deletions were found in exons 45-47. In addition, most mutations were clustered in the region between exons 45-55.

Although muscle involvement is milder in BMD than in DMD, cardiac involvement is often a prominent feature of the disease (27). The ability of patients with BMD to perform strenuous exercise may cause damage to myocardial cells with abnormal dystrophin by increasing mechanical stress on the heart (9). It has been reported in the literature that cardiac involvement is caused by mutations especially in exons 48 and 49 (28). Mutations involving exons 48 and 49 were found in all the patients with cardiac involvement in this study. Although life-threatening arrhythmias have been reported in patients with exon 3-4 duplications, no arrhythmia was observed in any of the current study patients with exon 3-4 duplication (29).

Different isoforms of dystrophin are also found in the cerebral cortex and Purkinje cells, but the function of all dystrophin isoforms in the brain is not yet fully understood. However, they appear to be involved in myelination during neuronal development, synaptic modulation, neuronal differentiation, and cellular energy metabolism. While cognitive abnormalities in DMD have been well documented, there is limited information on the cognitive profile of BMD patients (30–33).

Although the risk of intellectual disability is increased in children and adults affected by BMD, the average IQ does not differ from the general population (34). BMD has a higher incidence of learning disabilities and behavioral comorbidities compared to the general population, and learning disabilities have been reported in 32% of BMD patients (34). In the current study, psychiatric problems were detected in 15 (22.4%) patients, of which 7 were found to have learning disabilities.

In our study, the lower frequency of psychiatric disorders observed in patients with BMD compared to the general literature was attributed to the relatively short follow-up period and the lack of routine psychiatric evaluations. These limitations may have contributed to an underestimation of the prevalence of psychiatric comorbidities in this patient group (35,36).

Thangarajh et al. (37) reported that psychiatric problems such as ASD, ADHD, and LD were statistically more frequent in the upstream region of DMD exon 45. However, in the present patient group, no significant difference was observed between the two groups (37).

In conclusion, this study highlights the diversity of clinical and psychiatric challenges linked to genetic mutations in BMD patients. The impact of out-of-frame mutations on the phenotypic spectrum reinforces the heterogeneity described in the literature. Our findings emphasize the critical role of early diagnosis and the need for a multidisciplinary approach in the care of these patients. To further improve management strategies, we recommend prioritizing the systematic assessment of comorbidities to inform personalized care. The identification of mutation-specific clinical patterns may improve diagnostic and prognostic tools, reduce dependence on invasive procedures such as muscle biopsies, and ultimately improve the overall quality of care for BMD patients.

Limitations of the study

As this was a retrospective chart review, maternal carrier status could not be examined in all the patients and 6-minute walk test results were not available for all patients. In addition, accurate data on the ages at which comorbid conditions were detected were not available.

Ethics committee approval

This study was conducted in accordance with the Helsinki Declaration Principles. Ethical approval was granted by the Tepecik Training and Research Hospital Non-Interventional Ethics Committee (Decision No: 2024/08-05, Date: 02.09.2024).

Contribution of the authors

Güzin Y: Constructing the hypothesis or idea of research and/or article, Planning methodology to reach the conclusions, Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments, Taking responsibility in logical interpretation and conclusion of the results, Taking responsibility in necessary literature review for the study, Taking responsibility in the writing of the whole or important parts of the study, Reviewing the article before submission scientifically besides spelling and grammar. ***Doğan SD:*** Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments. ***Sarıkaya Uzan G:*** Organizing, supervising the course of progress and taking the responsibility of the research/study. ***Doğan G:*** Taking responsibility in logical interpretation and conclusion of the results. ***Özyılmaz B:*** Taking responsibility in logical interpretation and conclusion of the results. ***Tuncay B:*** Reviewing the article before submission scientifically besides spelling and grammar. ***Baydan F:*** Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments.

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Conflict of interest

The authors declare that there is no conflict of interest.

REFERENCES

1. Thada PK, Bhandari J, Forshaw KC, Umapathi KK. Becker Muscular Dystrophy. Published online 2024:1-10.

2. Andrew LaPelusa; Ria Monica D. Asuncion; Michael Kentris . Muscular Dystrophy. Published online 2024:1-31.
3. Fox H, Millington L, Mahabeer I, van Ruiten H. Duchenne muscular dystrophy. BMJ 2020;368:l7012.https://doi.org/10.1136/bmj.l7012.
4. Cai A, Kong X. Development of CRISPR-Mediated Systems in the Study of Duchenne Muscular Dystrophy. Hum Gene Ther Methods 2019;30:71-80.https://doi.org/10.1089/hgtb.2018.187.
5. Landrum Peay H, Fischer R, Tzeng JP, Hesterlee SE, Morris C, Martin AS, et al. Gene therapy as a potential therapeutic option for Duchenne muscular dystrophy: A qualitative preference study of patients and parents. PLoS One 2019;14:e0213649. https://doi.org/10.1371/journal.pone.0213649.
6. Romitti PA, Zhu Y, Puzhankara S, James KA, Nabukera SK, Zamba GK, et al. Prevalence of Duchenne and Becker muscular dystrophies in the United States. Pediatrics 2015;135:513-21. https://doi.org/10.1542/peds.2014-2044.
7. Ryder S, Leadley RM, Armstrong N, Westwood M, de Kock S, Butt T, et al. The burden, epidemiology, costs and treatment for Duchenne muscular dystrophy: an evidence review. Orphanet J Rare Dis 2017;12:79. https://doi.org/10.1186/s13023-017-0631-3.
8. Bradley WG, Jones MZ, Mussini JM, Fawcett PR. Becker-type muscular dystrophy. Muscle Nerve 1978;1:111-32. https://doi.org/10.1002/mus.880010204.
9. Basil T Darras M. Duchenne and Becker muscular dystrophy: Clinical features and diagnosis. UpToDate Published online 2023.
10. Darras BT, Urion DK, Ghosh PS. Dystrophinopathies Summary Genetic counseling GeneReview Scope. GeneReviews Published online 2018:1-35.
11. Banihani R, Smile S, Yoon G, Dupuis A, Mosleh M, Snider A, et al. Cognitive and Neurobehavioral Profile in Boys With Duchenne Muscular Dystrophy. J Child Neurol 2015;30:1472-82. https://doi.org/10.1177/0883073815570154.
12. Mavrogeni S, Markousis-Mavrogenis G, Papavasiliou A, Kolovou G. Cardiac involvement in Duchenne and Becker muscular dystrophy. World J Cardiol 2015;7:410-4. https://doi.org/10.4330/wjc.v7.i7.410
13. Paternostro-Sluga T, Grim-Stieger M, Posch M, Schuhfried O, Vacariu G, Mittermaier C, et al. Reliability and validity of the Medical Research Council (MRC) scale and a modified scale for testing muscle strength in patients with radial palsy. J Rehabil Med. 2008;40:665-71. https://doi.org/10.2340/16501977-0235.
14. Lee T, Takeshima Y, Kusunoki N, Awano H, Yagi M, Matsuo M, et al. Differences in carrier frequency between mothers of Duchenne and Becker muscular dystrophy patients. J Hum Genet 2014;59:46-50. https://doi.org/10.1038/jhg.2013.119.
15. Zimowski JG, Pilch J, Pawelec M, Purzycka JK, Kubalska J, Ziora-Jakutowicz K, et al. A rare subclinical or mild type of Becker muscular dystrophy caused by a single exon 48 deletion of the dystrophin gene. J Appl Genet 2017;58:343-7. https://doi.org/10.1007/s13353-017-0391-8.
16. Ciafaloni E, Fox DJ, Pandya S, Westfield CP, Puzhankara S, Romitti PA, et al. Delayed diagnosis in duchenne muscular dystrophy: data from the Muscular Dystrophy Surveillance, Tracking, and Research Network (MD STARnet). J Pediatr 2009;155:380-5. https://doi.org/10.1016/j.jpeds.2009.02.007.
17. Angelini C, Marozzo R, Pegoraro V. Current and emerging therapies in Becker muscular dystrophy (BMD). Acta Myol 2019;38:172-9.
18. Hoffman EP, Fischbeck KH, Brown RH, Johnson M, Medori R, Loike JD, et al. Characterization of dystrophin in muscle-biopsy specimens from patients with Duchenne's or Becker's muscular dystrophy. N Engl J Med 1988;318:1363-8. https://doi.org/10.1056/NEJM198805263182104.
19. Gao QQ, McNally EM. The Dystrophin Complex: Structure, Function, and Implications for Therapy. Compr Physiol 2015;5:1223-39. https://doi.org/10.1002/j.2040-4603.2015.tb00638.x.
20. van den Bergen JC, Wokke BH, Janson AA, van Duinen SG, Hulsker MA, Ginjaar HB, et al. Dystrophin levels and clinical severity in Becker muscular dystrophy patients. J Neurol Neurosurg Psychiatry 2014;85:747-53. https://doi.org/10.1136/jnnp-2013-306350.
21. Angelini C, Fanin M, Pegoraro E, Freda MP, Cadaldini M, Martinello F. Clinical-molecular correlation in 104 mild X-linked muscular dystrophy patients: characterization of sub-clinical phenotypes. Neuromuscul Disord 1994;4:349-58. https://doi.org/10.1016/0960-8966(94)90071-X.
22. Angelini C, Fanin M, Freda MP, Martinello F, Miorin M, Melacini P, et al. Prognostic factors in mild dystrophinopathies. J Neurol Sci 1996;142:70-8. https://doi.org/10.1016/0022-510X(96)00144-X.
23. Monaco AP, Bertelson CJ, Liechti-Gallati S, Moser H, Kunkel LM. An explanation for the phenotypic differences between patients bearing partial deletions of the DMD locus. Genomics 1988;2:90-5. https://doi.org/10.1016/0888-7543(88)90113-9.
24. Kesari A, Pirra LN, Bremadesam L, McIntyre O, Gordon E, Dubrovsky AL, et al. Integrated DNA, cDNA, and protein studies in Becker muscular dystrophy show high exception to the reading frame rule. Hum Mutat 2008;29:728-37. https://doi.org/10.1002/humu.20722.
25. Zambon AA, Waldrop MA, Alles R, Weiss RB, Conroy S, Moore-Clingenpeel M, et al. Phenotypic Spectrum of Dystrophinopathy Due to Duchenne Muscular Dystrophy Exon 2 Duplications. Neurology 2022;98:e730-e8. https://doi.org/10.1212/WNL.0000000000013246.
26. Vengalil S, Preethish-Kumar V, Polavarapu K, Mahadevappa M, Sekar D, Purushottom M, et al. Duchenne Muscular Dystrophy and Becker Muscular Dystrophy Confirmed by Multiplex Ligation-Dependent Probe Amplification: Genotype-Phenotype Correlation in a Large Cohort. J Clin Neurol 2017;13:91-7. https://doi.org/10.3988/jcn.2017.13.1.91.
27. Silvestri NJ, Ismail H, Zimetbaum P, Raynor EM. Cardiac involvement in the muscular dystrophies. Muscle Nerve 2018;57:707-15. https://doi.org/10.1002/mus.26014.
28. Melacini P, Fanin M, Danieli GA, Fasoli G, Villanova C, Angelini C, et al. Cardiac involvement in Becker muscular dystrophy. J Am Coll Cardiol 1993;22:1927-34. https://doi.org/10.1016/0735-1097(93)90781-U.
29. Ishizaki M, Fujimoto A, Ueyama H, Nishida Y, Imamura S, Uchino M, et al. Life-threatening Arrhythmias in a Becker Muscular Dystrophy Family due to the Duplication of Exons 3-4 of the Dystrophin Gene. Intern Med 2015;54:3075-8. https://doi.org/10.2169/internalmedicine.54.3986.
30. Aranmolate A, Tse N, Colognato H. Myelination is delayed during postnatal brain development in the mdx mouse model of Duchenne muscular dystrophy. BMC Neurosci 2017;18:63. https://doi.org/10.1186/s12868-017-0381-0.
31. Rae MG, O'Malley D. Cognitive dysfunction in Duchenne muscular dystrophy: a possible role for neuromodulatory immune molecules. J Neurophysiol 2016;116:1304-15. https://doi.org/10.1152/jn.00248.2016.

32. García-Cruz C, Merino-Jiménez C, Ceja V, Aragon J, Siqueiros-Marquez L, Reyes-Grajeda JP, et al. The dystrophin isoform Dp71e(Δ71) is involved in neurite outgrowth and neuronal differentiation of PC12 cells. J Proteomics 2019;191:80-7. https://doi.org/10.1016/j.jprot.2018.03.027.
33. Pezzoni L, Brusa R, Difonzo T, Magri F, Velardo D, Corti S, et al. Cognitive abnormalities in Becker muscular dystrophy: a mysterious link between dystrophin deficiency and executive functions. Neurol Sci 2024;45:1691-8. https://doi.org/10.1007/s10072-023-07169-x.
34. Young HK, Barton BA, Waisbren S, Portales Dale L, Ryan MM, Webster RI, et al. Cognitive and psychological profile of males with Becker muscular dystrophy. J Child Neurol 2008;23:155-62. https://doi.org/10.1177/0883073807307975.
35. Lambert JT, Darmahkasih AJ, Horn PS, Rybalsky I, Shellenbarger KC, Tian C, et al. Neurodevelopmental, behavioral, and emotional symptoms in Becker muscular dystrophy. Muscle Nerve 2020;61:156-62. https://doi.org/10.1002/mus.26750.
36. Mori-Yoshimura M, Mizuno Y, Yoshida S, Yoshida S, Ishihara N, Minami N, et al. Psychiatric and neurodevelopmental aspects of Becker muscular dystrophy. Neuromuscul Disord 2019;29:930-9. https://doi.org/10.1016/j.nmd.2019.09.006.
37. Thangarajh M, Hendriksen J, McDermott MP, Martens W, Hart KA, Griggs RC. Relationships between DMD mutations and neurodevelopment in dystrophinopathy. Neurology 2019;93:e1597-e604. https://doi.org/10.1212/WNL.0000000000008363.

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Original Article

Differential laboratory findings of common respiratory viruses in hospitalized children: a retrospective study

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ABSTRACT

Objective: The objective of this study was to comparatively investigate the effects of five common viral agents causing respiratory infections—influenza virus (IFV), human respiratory syncytial virus (hRSV), human rhinovirus (hRV), human metapneumovirus (hMPV), and human bocavirus (hBoV)—on laboratory parameters and clinical outcomes in children without underlying chronic diseases.

Material and Methods: A total of 983 children aged one month to eighteen years who presented to Ankara Bilkent City Hospital between January 2020 and December 2024 were retrospectively evaluated. Only children hospitalized with a single detected viral agent were included in the study. Clinical data and laboratory parameters were analyzed based on the identified viral pathogens.

Results: Among the 983 patients included, 62% were male. The most commonly detected viral agent was IFV, followed by hBoV, hRSV, hRV, and hMPV. In IFV infections, elevated levels of AST and ALT were observed. Significant elevations in partial pressure of PCO_2 and HCO_3 were detected in hRSV infections. In hBoV infections, inflammatory markers such as CRP, WBC, and NLR reached the highest levels. Furthermore, decreases in pH and increases in PCO_2 were significantly associated with intubation and intensive care admissions.

Conclusion: The distinct biomarker profiles exhibited by different viral agents may aid in guiding the clinical decision-making process. In particular, early assessment of biomarkers such as LDH, pH, PCO_2, and CRP at the time of hospital admission can be valuable for predicting disease severity and determining the need for intensive care in the clinical management of pediatric viral infections.

Keywords: Human bocavirus, Human metapneumovirus, Human respiratory syncytial virus, Human rhinovirus, Influenza virus, Lower respiratory tract infections

INTRODUCTION

Lower respiratory tract infections (LRTIs) are among the most common causes of hospital admissions and prolonged stays during childhood. Viral pathogens, particularly in children under the age of five, are the primary etiological agents of LRTIs. The clinical manifestations of these infections can vary significantly, ranging from mild symptoms to severe illness requiring intensive care (1). In recent years, the identification of viral agents has been greatly enhanced by the widespread adoption of diagnostic techniques such as multiplex real-time PCR (mPCR), enabling a more comprehensive evaluation of the clinical and laboratory profiles associated with viral infections (2).

Common blood tests play a critical role among the parameters frequently utilized in the evaluation of infectious diseases. Hematologic markers such as white blood cells (WBC), lymphocytes (LYM), platelets (PLT), and eosinophils (EO) are widely used laboratory indicators in the clinical assessment of infections (3,4). C-reactive protein (CRP), an acute-phase reactant, can rise rapidly in response to acute inflammatory processes. Although it is well known to increase markedly in bacterial infections, elevated levels may also be observed in viral infections and can serve as an indicator of disease severity (5,6). Blood gas parameters—including pH, partial pressure of oxygen (PO_2), partial pressure of carbon dioxide (PCO_2), bicarbonate (HCO_3), and lactate—are essential in the assessment of respiratory failure, particularly in LRTIs. These parameters are also commonly used to evaluate and monitor the morbidity and potential mortality associated with the disease (7).

This study aimed to investigate the changes in laboratory parameters and their relationship with clinical outcomes in infections caused by common respiratory viruses—such as influenza virus (IFV), human respiratory syncytial virus (hRSV),

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human rhinovirus (hRV), human metapneumovirus (hMPV), and human bocavirus (hBoV)—in children without underlying chronic diseases.

MATERIALS and METHODS

Study group and study design

This study was conducted on 983 children who presented to and were hospitalized at the Pediatric Clinics of Ankara Bilkent City Hospital Children's Hospital. The study population included children aged between one month and 18 years, admitted between January 2020 and December 2024, who underwent multiplex real-time PCR (mPCR) testing on nasopharyngeal swab samples and had no history of chronic disease. Children who presented to the hospital solely with respiratory infection symptoms, had an mPCR sample taken at admission, and had no comorbid conditions were included in the evaluation. Patients who tested positive for IFV, hBoV, hMPV, hRV, or hRSV via mPCR were retrospectively analyzed. Exclusion criteria included a history of chronic disease, improperly collected samples, detection of multiple viral agents on mPCR testing, hospitalization for reasons unrelated to respiratory distress, or detection of non-respiratory pathogens. Laboratory parameters are examined only when clinically indicated. In the study group, the laboratory parameters of patients who underwent these tests were evaluated.

Data collection

In this study, data were retrospectively obtained from the electronic health records system of Ankara Bilkent City Hospital. The analyzed variables included patient age, gender, initial admission location [Pediatric Intensive Care Unit (PICU) or non-intensive care unit (non-PICU)], type of respiratory support provided at admission [invasive mechanical ventilation (IMV), non-invasive ventilation (NIV), high-flow nasal cannula (HFNC), or mask oxygen therapy], and laboratory parameters. The evaluated laboratory parameters included aspartate aminotransferase (AST), alanine aminotransferase (ALT), lactate dehydrogenase (LDH), pH, partial pressure of carbon dioxide (PCO_2), partial pressure of oxygen (PO_2), bicarbonate (HCO_3), lactate (LAC), C-reactive protein (CRP), white blood cell count (WBC), absolute neutrophil count (ANC), absolute lymphocyte count (ALC), eosinophils (EO), platelets (PLT), large unstained cells (LUC), delta neutrophil index (DNI), and neutrophil-to-lymphocyte ratio (NLR).

Multiplex RT-PCR analysis

Respiratory viruses were identified using the multiplex real-time PCR assay (Rotor-Gene Q, QIAGEN, Germantown, Maryland, United States). This technique facilitates the detection of various pathogens, including IFV, hRSV, hCoV (Corana 229E, OC43, NL63, HKU1, SARS-COV2), hPIV, hMPV, hRV, EV, hBoV, hAdV, and human parechovirus. Additionally, bacterial pathogens including *Mycoplasma pneumoniae*, *Bordetella pertussis, Chlamydophila pneumoniae, Haemophilus influenzae,* and *Streptococcus pneumoniae* were also detected.

Statistical analysis

The Statistical Package for the Social Sciences (SPSS) 23.0 (Chicago, Illinois, USA) was implemented for the statistical analysis. The Kolmogorov-Smirnov test and the examination of histograms were used to evaluate the compliance of numerical and continuous variables with normal distribution. Numerical data with a normal distribution were expressed as the mean and standard deviation, while data with a non-normal distribution were expressed as the median and interquartile range (IQR). Percentages (%) and numbers (n) were used to express categorical variables. In contrast, the Mann-Whitney U test was used to compare continuous variables that did not meet the normal distribution. The Kruskal-Wallis test evaluated continuous variables from many groups that did not fit into the normal distribution. Categorical variables were analyzed with the Pearson chi-square or Fisher's Exact Test. When comparing more than one group, p values were calculated using the Bonferroni correction. Binary logistic regression analysis was performed to evaluate the association between age and the likelihood of PICU admission. The significance level was established at $p<0.050$.

RESULTS

This study was conducted on a total of 983 pediatric patients, with males comprising 62% (n=609) of the participants. The distribution of detected viral agents was as follows: IFV accounted for 31.7% (n=317), hRSV 22.5% (n=221), hRV 11.5% (n=113), hMPV 10.0% (n=98), and hBoV 24.3% (n=239). The median age across the study group was established as three (1–5) years. The median ages according to viral agents were as follows: five (2–9) years for IFV, two (1–3) years for hRSV, one (0.4–4) year for hRV, three (2–4) years for hMPV, and 2.5 (1–4) years for hBoV. A statistically significant difference in age was found among the viral agents ($p<0.001$), with IFV infections occurring in older age groups compared to other viruses ($p<0.001$), while hRV and hRSV infections were more frequently observed in younger children ($p<0.001$).

IFV infection was associated with a significant increase in AST and ALT levels compared to hRSV, hRV, hMPV, and hBoV infections ($p<0.001$). hBoV infections were characterized by higher CRP levels compared to IFV infections ($p<0.001$). Significant increases in PCO_2 and HCO_3 levels were observed in hRSV infections, and these parameters were significantly higher compared to hRV, hMPV, and hBoV infections ($p<0.001$). hRV infections were associated with noticeable increases in WBC, EO, and PLT levels ($p<0.001$). hMPV infections were particularly noted for increases in LDH and CRP levels ($p<0.001$). Among

Table I: Laboratory biomarkers by viral respiratory pathogens in pediatric patients

Tests	IFV*	hRSV*	hRV*	hMPV*	hBoV*	Total*	p†
AST (U/L)*	302; 43 (32.7-66)	212; 37 (30-47.7)	109; 34 (28-43)	86; 38 (31.5-51)	227; 33 (28-42)	936; 37 (30-49)	<0.001
ALT (U/L)	302; 23 (17-36)	212; 23 (17-29)	109; 21 (16-27)	87; 20 (15-28)	231; 19 (15-25)	941; 21 (16-30)	<0.001
LDH (U/L)	282; 329 (282.7-386.3)	185; 330 (287.5-371.5)	104; 316 (278.5-376.5)	80; 363.5 (304-404)	202; 337.5 (300.7-383)	853; 333 (291-383)	0.026
pH	194; 7.43 (7.40-7.47)	176; 7.40 (7.36-7.44)	82; 7.40 (7.35-7.43)	68; 7.42 (7.39-7.44)	193; 7.41 (7.37-7.46)	713; 7.42 (7.37-7.45)	<0.001
PCO_2 (mmHg)	194; 31.9 (28.3-35.9)	176; 36.1 (31.6-41.8)	82; 33.8 (29.3-39)	68; 33.8 (29.1-39.3)	191; 30.7 (27.6-36)	712; 33.3 (28.9-38.1)	<0.001
PO_2 (mmHg)	194; 44.7 (37.2-58.3)	176; 47.2 (36.3-59.9)	81; 48.2 (36.1-61.3)	67; 43.8 (37.9-59.8)	194; 49.2 (38.4-64.5)	712; 47.1(37.4-60.8)	0.610
HCO_3 (mEq/L)	193; 21.3 (19.3-22.9)	176; 22.3 (20.1-25)	82; 20.8[d] (18.2-22.9)	68; 21.9 (19.1-25.1)	191; 19.9[d] (17.8-21.9)	710; 21.2 (18.9-23.3)	<0.001
LAC (mmol/L)	193; 1.7 (1.2-2.6)	177; 2 (1.4-2.9)	80; 2 (1.7-2.5)	67; 1.9 (1.5-2.6)	191; 1.8 (1.3-2.4)	708; 1.9 (1.4–2.6)	0.008
CRP (mg/L)	305; 9.1 (3.3-20.4)	212; 4 (1-20)	113; 4 (2-8)	85; 10 (1-40)	224; 11.3 (4-30.7)	939; 8.5 (1.9–20.8)	<0.001
WBC x10^9/L	298; 7.04 (4.71-9.74)	212; 8.85 (6.82-11.49)	112; 11.08 (8.04-14.85)	88; 9.99 (6.60-12.99)	229; 10.69 (8.57-13.29)	939; 9.10 (6.33–12.34)	<0.001
ANC x10^9/L	298; 3.75 (2.01-6.37)	212; 3.24 (1.90-4.99)	112; 5.73 (2.92-9.45)	88; 4.38 (2.78-6.70)	229; 6.59 (4.34-9.78)	939; 4.44 (2.38–7.25)	<0.001
ALC x10^9/L	297; 1.67 (1.04-2820)	212; 4.19 (2.92-5.48)	112; 3.21 (1.76-5.06)	88; 3.79 (2.11-5.63)	229; 2.51 (1.55-3.94)	938; 2.66 (1.50–4.42)	<0.001
EO (/µL)	298; 30 (10-70)	212; 90 (30-230)	112; 110 (40-280)	88; 55 (12.5-177.5)	228; 100 (40-257.5)	938; 60 (20–180)	<0.001
PLT (/µL)	298; 266 (201-350)	212; 427 (334-504)	112; 430 (332-544)	88; 366 (294-507)	229; 355 (285-450)	939; 347 (264–465)	<0.001
LUC (/µL)	298; 160 (100-270)	212; 350 (230-477.5)	112; 245 (142.5-380)	87; 330 (210-460)	228; 150 (40-280)	937; 210 (90–360)	<0.001
DNI	247; 0.1 (0.1-0.5)	212; 0.1 (0.1-0.1)	112; 0.1 (0.1-0.1)	87; 0.1 (0.1-0.1)	222; 0.1 (0.1-0.1)	880; 0.1 (0.1-0.1)	0.161
NLR	298; 2.2 (0.9-4.3)	177; 0.7 (0.3-1.4)	112; 1.5 (0.6-1.5)	55; 1 (0.4-2.2)	217; 2.8 (1.2-5.4)	859; 1.7 (0.7–4.1)	<0.001

:** *n; median (min-max),* **†:** *Kruskal-Wallis test,* ***AST: *Aspartate Aminotransferase,* ***ALT:*** *Alanine Aminotransferase,* ***LDH:*** *Lactate Dehydrogenase,* ***CRP:*** *C-Reaktif Protein* ***WBC:*** *White Blood Cell,* ***ANC:*** *Absolute Neutrophil,* ***ALC:*** *Absolute Lymphocyte,* ***EO:*** *Eosinophil,* ***PLT:*** *Platelet,* ***LUC:*** *Large Unstained Cells,* ***DNI:*** *Delta Neutrophil Index,* ***NLR:*** *Neutrophil-to-Lymphocyte Ratio,* ***LAC:*** *Lactate*

all viral agents, hBoV infections showed the highest levels of CRP and WBC (p<0.001) (Table I).

A total of 84.5% (n=831) of the patients were admitted to non-PICU units, while 15.5% (n=152) were admitted to the PICU. The most common viral agent associated with intensive care admission was hRSV, followed by hMPV, hBoV, hRV, and IFV (p<0.001). When laboratory data were compared in relation to intensive care admissions, a decrease in pH levels and increases in PCO_2 and HCO_3 levels were significantly associated with the need for intensive care (p<0.001 for each). Additionally, elevated LDH levels (p=0.013) and higher LUC values (p=0.012) were also significantly associated with PICU admission. Higher rates of intensive care admission were observed in younger age groups, with the frequency decreasing as age increased. Each one-year increase in age was associated with a 14% decrease in the likelihood of PICU admission (Exp(B)=0.853; 95% CI: 0.794–0.915; p<0.001).

During the treatment process, 2.6% (n=26) of the patients received IMV, 14.2% (n=140) received NIV, 9.9% (n=97) received HFNC, and 73.2% (n=720) received oxygen support via mask. Among all biomarkers, pH and partial pressure of PCO_2 were the parameters that showed the most consistent and significant differences across the respiratory support groups. In patients who underwent IMV, pH levels were significantly lower compared to the NIV (p=0.003), HFNC (p<0.001), and mask (p<0.001) groups. Similarly, PCO_2 levels in the IMV group were significantly higher compared to the NIV (p=0.004), HFNC (p<0.001), and mask (p<0.001) groups. Although significant differences were observed in LDH, HCO_3, and LUC levels—particularly between the NIV and mask groups—these differences were not consistent across all groups. Parameters such as CRP, ANC, and PLT did not show statistically significant differences between the groups. Furthermore, no significant difference was found between patients treated with HFNC and those treated with a mask in terms of the biomarkers analyzed.

DISCUSSION

Viral agents causing LRTIs may result in clinical presentations ranging from mild symptoms to severe morbidity and mortality. In this study, laboratory parameters associated with hospital admissions due to common viral agents were evaluated in a large cohort of children without underlying chronic diseases. Furthermore, differences in laboratory findings among various viral agents and their potential effects on the clinical course were investigated. The findings of this study contribute to the identification of virus-specific biomarkers and support improved risk stratification in the management of pediatric patients.

The literature indicates that viral agents are most commonly detected in children under the age of four (8–11). The median age findings in our study are consistent with these reports. It was found that the median age of children with IFV infection was higher compared to those with other viral agents, in line with previous studies (12,13). The median ages associated with other viral agents also showed similarities to previously reported data (2). The findings suggest that careful clinical evaluation is essential for children under the age of three in cases of hRV, hRSV, hMPV, and hBoV, and for older children in cases of IFV. In particular, when assessing hospitalized children without underlying conditions, clinicians should consider that viral agents may play a significant role in hospital admissions among those under five years of age. Therefore, special attention should be given during the initial clinical assessment of patients within this age group.

Increases in ALT and AST levels are important biomarkers indicating potential organ failure, while LDH reflects tissue damage and the presence of hypoxia (14). In the literature, some studies have reported elevated liver enzyme levels in 19.4% of patients, with the highest frequencies observed in infections caused by hRSV (50%) and IFV (35.8%) (15). Another study found that the highest prevalence of elevated ALT and AST levels occurred in IFV cases (33.7%), followed by hRSV infections (30.9%) (16). Consistent with these findings, the present study also observed the most frequent elevations of ALT and AST in IFV infections. LDH has been identified as a significant prognostic marker in infectious diseases such as COVID-19 and RSV in various studies (17,18). In adult patients, elevated LDH levels in pneumonia have been directly associated with increased mortality (19). In pediatric cases, LDH levels were found to be significantly higher in intensive care admissions related to COVID-19 (20). In a study involving infants, LDH levels were reported to be higher in human metapneumovirus (hMPV) infections compared to hRSV infections (21). In the present study, we observed the most notable LDH elevations in hMPV infections, followed by human bocavirus (hBoV). Although elevations in ALT and AST were not significantly associated with intensive care admissions, LDH levels, consistent with previous reports, showed a significant association. Therefore, assessing ALT and AST levels upon admission for evaluating the risk of organ failure, and monitoring LDH levels as a predictor of tissue damage and the potential need for intensive care, is crucial in clinical management. Patients with elevated biomarker levels, particularly LDH, should be closely monitored for morbidity and mortality.

In the literature, hRSV has been reported as the viral agent most frequently associated with intensive care admissions and the need for intubation (22,23). Consistently, in the present study, hRSV infections constituted the highest-risk group. Analysis of blood gas parameters revealed significant increases in partial pressure of PCO_2 and HCO_3 levels in hRSV infections. The bronchiolitis commonly observed in hRSV cases may reduce alveolar ventilation, leading to the development of hypercapnia. This condition is often accompanied by a metabolic compensation, which results in elevated bicarbonate levels. Additionally, the lowest pH values were observed in patients requiring intubation—particularly those with hRSV infections—rather than in patients with other viral agents. A decrease in pH and an increase in PCO_2 are considered important biomarkers for predicting respiratory failure (24,25). In the current study, these parameters showed the most pronounced differences in the intubation group relative to other patient groups. Therefore, rapid and careful assessment of pH and PCO_2 levels is critical in making decisions regarding intensive care admission or intubation, and may significantly enhance clinical management.

The literature indicates that CRP levels can serve as an important early biomarker for sepsis and mortality. In particular, CRP levels at the time of hospital admission are considered a key indicator for evaluating disease severity. Several studies have demonstrated statistically significant associations between CRP and other inflammatory markers with mortality (26,27). In the present study, the highest CRP levels were observed in infections caused by human bocavirus (hBoV) and human metapneumovirus (hMPV). Notably, the pronounced elevation

of CRP in hBoV infections suggests that this virus may trigger a systemic inflammatory response. Supporting this finding, previous reports have stated that hBoV infections may present with elevated CRP levels and thus mimic bacterial infections (28). Therefore, in cases with high CRP levels at admission, the exclusion of viral etiologies should be carefully considered prior to initiating antibiotic therapy, as part of the clinical decision-making process.

White blood cell (WBC) count is one of the most commonly used laboratory parameters in the evaluation of childhood infections. In the present study, significant increases in WBC, EO, and PLT levels were observed in infections caused by hRV and hBoV. The eosinophilia observed in hRV infections may reflect not only viral inflammation but also suggest that these infections occur more frequently or more severely in children with a history of atopy or asthma (29). In contrast, lower EO levels in hRSV and IFV infections support this distinction. Furthermore, previous studies have indicated that hRV may lead to a more severe clinical course in children with underlying allergic conditions (30). The increase in platelet count is generally interpreted as reactive thrombocytosis and is recognized as a marker of the systemic inflammatory response to infection (31,32). In our study, the highest platelet levels were observed in hRV and hRSV infections, suggesting that both viruses may elicit a strong inflammatory response. Lymphocyte subgroups, such as ANC and ALC, can provide valuable insights into the nature and progression of an infection. The finding that the highest ANC levels were observed in hBoV and hRV infections suggests that these viruses may be associated with neutrophil-dominant inflammation. Conversely, the highest ALC levels were detected in hRSV infections, indicating a predominantly lymphocytic response. In this context, evaluating both ANC and ALC levels in combination may serve as a useful clinical tool for distinguishing viral from bacterial infections.

The neutrophil-to-lymphocyte ratio (NLR) is commonly used as an indicator of systemic inflammation in adult populations (33). In pediatric cases, several studies have reported elevated NLR levels during severe viral infections (34,35). In the present study, the highest NLR levels were observed in infections caused by hBoV, suggesting that hBoV may be associated with a more aggressive inflammatory response. However, viral infections can present with varying hematological profiles, including not only neutrophilia and lymphopenia but also neutropenia accompanied by lymphopenia. These variations indicate that NLR may not consistently serve as a reliable standalone marker for assessing the severity of viral infections in children.

Some studies have reported that decreased levels of large unstained cells (LUC) are significantly associated with intensive care admissions, suggesting that an inadequate production of activated lymphocytes and a compromised immune response may contribute to severe clinical outcomes (34). Conversely, the present study found that increased LUC levels were significantly associated with both intubation and intensive care admissions. LUC represents immature cell populations and typically rises during viral infections. This elevation is particularly notable in cases where the immune system is strongly stimulated and may serve as a useful biomarker in clinical presentations resembling sepsis (36,37). Although the prognostic use of LUC levels in pediatric patients remains limited, it is considered to have potential for further investigation in future research.

While this study provides important findings, several limitations should be acknowledged. First, its single-center and retrospective design restricts the generalizability of the results to broader populations. Nevertheless, the inclusion of a large pediatric cohort and the exclusion of children with chronic diseases are notable strengths that enhance the study's internal validity. In addition, the study did not assess the impact of viral load, as detected by multiplex real-time PCR (mPCR), on clinical outcomes. This omission may limit the clinical interpretation of certain laboratory parameters. The absence of sputum cultures also complicates the exclusion of secondary bacterial infections. However, the mPCR assay covers several common pneumonia pathogens, and any patients in whom these pathogens were identified were excluded from the study.

This study demonstrates that common viral respiratory infections in pediatric patients are associated with distinct biomarker profiles. Notably, elevated AST and ALT levels in IFV infections, as well as the significant association of increased partial pressure of PCO_2 and HCO_3 levels in hRSV infections with the need for intensive care, are particularly striking. Human bocavirus (hBoV) infections exhibited the highest values in inflammatory markers such as CRP, WBC, and NLR, suggesting a more pronounced systemic inflammatory response. The findings indicate that certain biomarkers measured at the time of admission—such as pH, PCO_2, LDH, and CRP—may serve as valuable tools in predicting disease severity. Therefore, early evaluation of these biomarkers can significantly contribute to risk stratification and decision-making regarding intensive care needs in the clinical management of pediatric viral infections.

Ethics committee approval

This study was conducted in accordance with the Declaration of Helsinki and received ethical approval from the Ethics Committee of Ankara Bilkent City Hospital (10.07.2024/TABED 2-24-299).

Contribution of the authors

Kalaycı F : Constructing the hypothesis or idea of research and/or article, Planning methodology to reach the conclusions, Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in logical interpretation and conclusion of the results, Taking responsibility in the writing of the whole or important parts of the study, Reviewing the article before submission scientifically besides spelling and grammar. ***Çelebier K:*** Taking responsibility in patient follow-up, collection of relevant biological materials,

data management and reporting, execution of the experiments, Taking responsibility in logical interpretation and conclusion of the results, Taking responsibility in necessary literature review for the study, Reviewing the article before submission scientifically besides spelling and grammar.

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Conflict of interest

The authors declare that there is no conflict of interest.

REFERENCES

1. Kurskaya O, Ryabichenko T, Leonova N, Shi W, Bi H, Sharshov K, et al. Viral etiology of acute respiratory infections in hospitalized children in Novosibirsk City, Russia (2013 - 2017). PLoS One. 2018;13(9): e0200117. https://doi.org/10.1371/journal.pone.0200117
2. Schober T, Wong K, Delisle G, Caya C, Brendish NJ, Clark TW, et al. Clinical Outcomes of Rapid Respiratory Virus Testing in Emergency Departments: A Systematic Review and Meta-Analysis. JAMA Intern Med. 2024 ;184(5):528-36. https://doi.org/10.1001/jamainternmed.2024.0037
3. Honda T, Uehara T, Matsumoto G, Arai S, Sugano M. Neutrophil left shift and white blood cell count as markers of bacterial infection. Clin Chim Acta. 2016;457:46-53. https://doi.org/10.1016/j.cca.2016.03.017
4. Zeng F, Li L, Zeng J, Deng Y, Huang H, Chen B, et al. Can we predict the severity of coronavirus disease 2019 with a routine blood test? Pol Arch Intern Med. 2020;130(5):400-6.
5. Ticinesi A, Lauretani F, Nouvenne A, Porro E, Fanelli G, Maggio M, et al. C-reactive protein (CRP) measurement in geriatric patients hospitalized for acute infection. Eur J Intern Med. 2017;37:7-12. https://doi.org/10.1016/j.ejim.2016.08.026
6. Li Y, Min L, Zhang X. Usefulness of procalcitonin (PCT), C-reactive protein (CRP), and white blood cell (WBC) levels in the differential diagnosis of acute bacterial, viral, and mycoplasmal respiratory tract infections in children. BMC Pulm Med. 2021;21(1):386. https://doi.org/10.1186/s12890-021-01756-4
7. Yildizdaş D, Yapicioğlu H, Yilmaz HL, Sertdemir Y. Correlation of simultaneously obtained capillary, venous, and arterial blood gases of patients in a paediatric intensive care unit. Arch Dis Child. 2004;89(2):176-80. https://doi.org/10.1136/adc.2002.016261
8. Cilla G, Oñate E, Perez-Yarza EG, Montes M, Vicente D, Perez-Trallero E. Viruses in community-acquired pneumonia in children aged less than 3 years old: High rate of viral coinfection. J Med Virol. 2008 ;80(10):1843-9. https://doi.org/10.1002/jmv.21271
9. Drews AL, Atmar RL, Glezen WP, Baxter BD, Piedra PA, Greenberg SB. Dual Respiratory Virus Infections. Clin Infect Dis. 1997;25(6):1421. https://doi.org/10.1086/516137
10. Chorazy ML, Lebeck MG, McCarthy TA, Richter SS, Torner JC, Gray GC. Polymicrobial Acute Respiratory Infections in a Hospital-Based Pediatric Population. Pediatr Infect Dis J. 2013;32(5):460-6. https://doi.org/10.1097/INF.0b013e31828683ce
11. Wei L, Liu W, Zhang XA, Liu EM, Wo Y, Cowling BJ, et al. Detection of Viral and Bacterial Pathogens in Hospitalized Children With Acute Respiratory Illnesses, Chongqing, 2009-2013. Medicine. 2015;94(16):e742. https://doi.org/10.1097/MD.0000000000000742
12. Wang X, Li Y, O'Brien KL, Madhi SA, Widdowson MA, Byass P, et al. Global burden of respiratory infections associated with seasonal influenza in children under 5 years in 2018: a systematic review and modelling study. Lancet Glob Health. 2020 ;8(4):e497-e510.
13. Nair H, Brooks WA, Katz M, Roca A, Berkley JA, Madhi SA, et al. Global burden of respiratory infections due to seasonal influenza in young children: a systematic review and meta-analysis. Lancet. 2011;378(9807):1917-30. https://doi.org/10.1016/S0140-6736(11)61051-9
14. Khan AA, Allemailem KS, Alhumaydhi FA, Gowder SJT, Rahmani AH. The Biochemical and Clinical Perspectives of Lactate Dehydrogenase: An Enzyme of Active Metabolism. Endocr Metab Immune Disord Drug Targets. 2020 ;20(6):855-68. https://doi.org/10.2174/1871530320666191230141110
15. Jeon NL, Kim BS, Kim YK, Hong SJ. Etiology and Clinical Features of Severe Acute Viral Lower Respiratory Tract Infections in Children. Journal of the Korean Pediatric Society. 2000;43(12):1558-68.
16. Oh JS, Choi JS, Lee YH, Ko KO, Lim JW, Cheon EJ, et al. The Relationships between Respiratory Virus Infection and Aminotransferase in Children. Pediatr Gastroenterol Hepatol Nutr. 2016;19(4):243. https://doi.org/10.5223/pghn.2016.19.4.243
17. Mehta R, Scheffler M, Tapia L, Aideyan L, Patel KD, Jewell AM, et al. Lactate dehydrogenase and caspase activity in nasopharyngeal secretions are predictors of bronchiolitis severity. Influenza Other Respir Viruses. 2014;8(6):617. https://doi.org/10.1111/irv.12276
18. Gupta GS. The Lactate and the Lactate Dehydrogenase in Inflammatory Diseases and Major Risk Factors in COVID-19 Patients. Inflammation. 2022;45(6):2091. https://doi.org/10.1007/s10753-022-01680-7
19. Tao RJ, Luo XL, Xu W, Mao B, Dai RX, Li CW, et al. Viral infection in community acquired pneumonia patients with fever: a prospective observational study. J Thorac Dis. 2018 ;10(7):4387-95.https://doi.org/10.21037/jtd.2018.06.33
20. Mania A, Faltin K, Mazur-melewska K, Małecki P, Jończyk-potoczna K, Lubarski K, et al. Clinical Picture and Risk Factors of Severe Respiratory Symptoms in COVID-19 in Children. Viruses. 2021;13(12):2366. https://doi.org/10.3390/v13122366
21. Taniguchi A, Kawada JI, Go K, Fujishiro N, Hosokawa Y, Maki Y, et al. Comparison of Clinical Characteristics of Human Metapneumovirus and Respiratory Syncytial Virus Infections in Hospitalized Young Children. Jpn J Infect Dis. 2019;72:237-42. https://doi.org/10.7883/yoken.JJID.2018.480
22. Al-Eyadhy A, Almazyad M, Hasan G, Almuhaideb Q, AbuDujain N, Alhaboob AAN, et al. The burden of viral infections in pediatric intensive care unit between endemic and pandemic coronavirus infections: A tertiary care center experience. J Infect Chemother 2022;29(1):20-5. https://doi.org/10.1016/j.jiac.2022.09.007
23. Tsou P, Vadivelan A, Kovvuri M, Garg N, Thangavelu M, Wang Y, et al. Association between multiple respiratory viral infections and pediatric intensive care unit admission among infants with bronchiolitis. Archives De Pediatrie. 2019;27(1):39-44. https://doi.org/10.1016/j.arcped.2019.11.006
24. Bilan N, Behbahan AG, Khosroshahi AJ. Validity of venous blood gas analysis for diagnosis of acid-base imbalance in children admitted to pediatric intensive care unit. World J Pediatr 2008 ;4(2):114-7. https://doi.org/10.1007/s12519-008-0022-x
25. Wrotek A, Jackowska T. Capillary Blood Gas in Children Hospitalized Due to Influenza Predicts the Risk of Lower Respiratory Tract Infection. Diagnostics. 2022 ;12(10):2412. https://doi.org/10.3390/diagnostics12102412

26. Wang D, Hu B, Hu C, Zhu F, Liu X, Zhang J, et al. Clinical Characteristics of 138 Hospitalized Patients With 2019 Novel Coronavirus-Infected Pneumonia in Wuhan, China. JAMA. 2020;323(11):1061-9. https://doi.org/10.1001/jama.2020.1585
27. Luo X, Zhou W, Yan X, Guo T, Wang B, Xia H, et al. Prognostic value of C-reactive protein in patients with COVID-19. Clin Infect Dis. 2020;71(16):ciaa641. https://doi.org/10.1101/2020.03.21.20040360
28. Jaya Lakshmi SS, Nisha V, Leela KV, Harsha Vardhini N, Subash S. Uncovering the Presentation and Diagnosis of Human Bocavirus in a Patient at a Tertiary Care Center: A Case Report. Cureus. 2024;16(8):e66550.
29. Jackson DJ, Gangnon RE, Evans MD, Roberg KA, Anderson EL, Pappas TE, et al. Wheezing rhinovirus illnesses in early life predict asthma development in high-risk children. Am J Respir Crit Care Med. 2008;178(7):667-72. https://doi.org/10.1164/rccm.200802-309OC
30. Jackson DJ, Gern JE. Rhinovirus Infections and Their Roles in Asthma: Etiology and Exacerbations. J Allergy Clin Immunol Pract. 2022;10(3):673-81. https://doi.org/10.1016/j.jaip.2022.01.006
31. Schattner A, Kadi J, Dubin I. Reactive thrombocytosis in acute infectious diseases: Prevalence, characteristics and timing. Eur J Intern Med. 2019;63:42-5. https://doi.org/10.1016/j.ejim.2019.02.010
32. Kurt F, Mustafaoğlu Ö, Alaboyun ES, Gürsoy C, Yakut Hİ, Dibek Mısırlıoğlu E, et al. Evaluation of Changes in Mean Platelet Volume in Acute Bronchiolitis Cases.Turkish J Pediatr Dis. 2020;14(4):352-8.
33. Huang Z, Fu Z, Huang W, Huang K. Prognostic value of neutrophil-to-lymphocyte ratio in sepsis: A meta-analysis. Am J Emerg Med. 2020;38(3):641-7. https://doi.org/10.1016/j.ajem.2019.10.023
34. Burrack N, Adar A, Goldbart A, Weissmann S, Cohen B, Hazan I, et al. Monocyte and neutrophil to lymphocyte ratios in hospitalized children with RSV bronchiolitis. Pediatr Pulmonol. 2023;58(12):3530-41. https://doi.org/10.1002/ppul.26687
35. Prozan L, Shusterman E, Ablin J, Mitelpunkt A, Weiss-Meilik A, Adler A, et al. Prognostic value of neutrophil-to-lymphocyte ratio in COVID-19 compared with Influenza and respiratory syncytial virus infection. Sci Rep. 2021;11(1):21519. https://doi.org/10.1038/s41598-021-00927-x
36. Shin D, Lee MS, Kim DY, Lee MG, Kim DS. Increased large unstained cells value in varicella patients: A valuable parameter to aid rapid diagnosis of varicella infection. J Dermatol. 2015;42(8):795-9. https://doi.org/10.1111/1346-8138.12902
37. Vanker N, Ipp H. Large unstained cells: a potentially valuable parameter in the assessment of immune activation levels in HIV infection. Acta Haematol. 2014;131(4):208-12. https://doi.org/10.1159/000355184

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Original Article

Prognosis of chronic urticaria in children

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ABSTRACT

Objective: Chronic urticaria (CU) is defined as the recurring presence of urticaria, angioedema, or both, for a duration of six weeks or more. In most patients, it is a self-limiting disease. The study aimed to investigate the clinical findings, comorbidities, laboratory results, response to treatment and prognosis in children with CU.

Material and Methods: Patients aged 0-18 years with CU followed up at the Pediatric Allergy and Immunology Outpatient Clinic of Selcuk University Faculty of Medicine Hospital between January 2022 and March 2024 were included, and their medical records were retrospectively analyzed.

Results: The study sample included 74 patients with CU. About 55.4% of the patients were girls. The mean age of the patients was 11 year (2.3-17.9). The mean follow-up period was 475 days (129-925). Sixteen patients (21.6%) had angioedema, while 15 patients (20.2%) reported presence of a trigger. Eleven patients (14.8%) had dermographism. Four patients (5.4%) were anti-thyroid peroxidase positive and seven patients (9.4%) were antinuclear antibody positive. Eosinopenia was present in 10 patients (13.5%), whereas basopenia was present in only two (2.7%) patients. Skin prick test detected positivity in 24 patients (32.4%). Sixty-eight patients (91.9%) responded to antihistamine and/or montelukast treatment, and omalizumab was prescribed to six patients (8.1%) showing no response to the conventional treatment.

Conclusion: In pediatric CU patients, the disease is often self-limiting without the need for treatment with omalizumab. Drug compliance should be evaluated in patients with poor disease control.

Keywords: Antihistamine, Chronic urticaria, Eosinopenia, Prognosis

INTRODUCTION

Urticaria is characterized by edematous papules/plaques involving the upper layers of the dermis, which are raised from the skin, fade with pressure, pink or red in color, with clear borders and usually surrounded by a ring of erythema (1). Chronic urticaria (CU) is broadly described as continuous or intermittent urticaria present for at least 6 weeks. The incidence of CU is estimated to be 1.4% per year, affecting 2-3% of the population and significantly impairing their quality of life. Previous studies indicate that the prevalence of CU is generally higher in females with a female-to-male ratio ranging from 7:3 to 4:1 (2,3).

Chronic spontaneous urticaria (CSU) accounts for around 50-75% of chronic urticaria cases, whereas chronic inducible urticaria (CIU) is responsible for approximately one-third of all cases (1). Epidemiological data derived from adult populations suggest that certain subtypes of CSU may have an underlying autoimmune etiology (3). In pediatric CSU patients, spontaneous remission occurs in 30% to 50% of cases within the first three-years following diagnosis (4).

The diagnostic workup for all urticaria patients should involve a comprehensive clinical history, the initial step crucial for accurate diagnosis and subsequent management. Clinicians should thoroughly collect all available information regarding the temporal aspects of symptoms (onset, frequency, and pattern), potential triggering factors, environmental exposures, presence of angioedema or other systemic manifestations, current and past medication use, and known allergies. Besides, urticarial lesions and angioedema often display an ephemeral nature they may not be evident during physical examination so it is of great importance to review documents detailing signs and symptoms, including photographic evidence of urticaria and/or angioedema (4-6). The third step in the diagnostic algorithm for CU involves a basic diagnostic workup with limited laboratory testing. Subsequent to these three steps, further diagnostic

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investigations may be performed, individually tailored based on previous findings, the specific type and subtype of urticaria identified, such as provocation tests for inducible urticaria (5,7).

The current treatment options for urticaria mainly focus on targeting mast cell mediators, such as histamine, or their activators including autoantibodies. The first line of treatment, as recommended by international guidelines, involves the administration of second-generation H1-antihistamines at standard doses for symptomatic relief (5). Leukotriene receptor antagonists (LTRA) particularly montelukast, which is the most extensively studied in this class have so far shown a favorable safety profile in numerous clinical trials, including those involving pediatric patients. However, their efficacy in CSU still remains rather limited, as we have no robust evidence to encourage LTRA use as monotherapy (8). For patients failing to achieve adequate symptomatic control with second-generation H1-antihistamines, omalizumab stands out as the only licensed drug therapy, thus representing the next echelon in the treatment algorithm. A monoclonal antibody targeting free immunoglobulin E (anti-IgE), omalizumab has been shown to be highly effective and safe in the management of CSU with reduced urticaria activity scores (9).

In this study, we aimed to investigate the factors affecting prognosis in children with chronic urticaria.

MATERIALS and METHODS

The sample of this retrospective study included pediatric patients aged 0-18 years who were evaluated and followed at the Pediatric Allergy and Immunology Outpatient Clinic of Selcuk University Faculty of Medicine Hospital from January 2022 to March 2024. Research data were retrieved from medical records and the hospital's electronic health record system. The following parameters were recorded for analyses: gender, age, family history, chronic diseases, duration of urticaria, type of urticaria, laboratory findings (complete blood count results, erythrocyte sedimentation rate (ESR), C-reactive protein (CRP) level, thyroid function tests, antithyroglobulin and antithyroid peroxidase autoantibodies (anti-TPO), antinuclearantibody (ANA) level, urine analysis).

Drug therapies: Treatment regimens administered to the study cohort were also documented. In order to assess current therapeutic status and remission outcomes, patients were contacted via telephone for a standardized follow-up interview. The Urticaria Activity Score over seven days (UAS7) was employed as a validated instrument to quantify disease activity and evaluate remission status.

Hematological and immunological parameters were analyzed through established cut-off values from the literature: an eosinophil count below 0.05×10^9/L was considered eosinopenia, while basophil values below 0.01×10^9/L were interpreted as basopenia (10,11). Serum total IgE levels were classified as low if below 40 kU/L and high if above 100 kU/L (12).

Diagnostic procedures: Results of skin prick tests for common aeroallergens and food allergens, and provocation tests for physical urticaria were collected and included in the analyses. Skin prick testing was performed through standardized allergen extracts from Credisol® (Maharashtra, India) and Lofarma® (Milano, Italy). To reduce potential confounding effects, patients were instructed to discontinue antihistamines 15 days prior to testing, antidepressants seven days prior, and montelukast three days prior. Positive control agent was a histamine solution (10 mg/mL) and the negative control substance was physiological saline. Allergen solutions were applied to the volar aspect of the forearm through the prick-lancet technique. Measured 15 minutes after allergen application, a positive reaction was defined as the development of a wheal measuring 3 mm or larger in diameter at the allergen test site as compared to the negative control site (13). Inhalation allergens used in the skin prick test included house dust mites (Dermatophagoides farinea, Dermotophagoides pteronyssinus), pollen allergens representing various sources like trees (redwood, birch), grasses (rye, grass), and weeds (xanthium or cocklebur, common plantain, lamb's quarters, and other prevalent weeds), cat epithelium, dog epithelium, molds (Alternaria alternata, Aspergillus and Cladosporium) and cockroach debris. Food allergens included common allergenic foods like milk, eggs, tree nuts, peanuts, wheat flour, fish, tomatoes, and soy. In addition to the skin prick tests, the ice cube test was used to evaluate for cold urticaria, which involved placing an ice cube on the inner side of the forearm for five minutes and observing for the development of hives, which would indicate an allergic reaction to cold.

Statistical analysis

Statistical analyses were performed via IBM IBM SPSS Statistics for Windows, version 22 (IBM Corp., Armonk, N.Y., USA). After evaluating the conformity of the data to normal distribution by the Kolmogorov Smirnov test, an after evaluating the conformity of the data to normal distribution by the Kolmogorov Smirnov test, median (IQR or minimum-maximum) or mean (standard deviation) values were used to evaluate numerical data; frequency distributions and percentages were used to summarize categorical data. Mann-Whitney U tests were conducted to explore the differences between groups for data not conforming to normal distribution. Chi-square test was used to compare categorical data. Statistical significance level was accepted as $p<0.050$ in all tests.

RESULTS

Our study included 74 patients. The mean age at presentation was 11 year (2.3-17.9). Female patients constituted the majority with a rate of 55.4% (n=41). Patients were followed up for a mean duration of 475 days (129-925) after diagnosis.

Sixteen patients (21.6%) had angioedema associated with urticaria. When chronic diseases were questioned, asthma was the most common comorbidity. Among the comorbidities were allergic rhinitis, hypothyroidism, familial mediterranean fever (FMF), idiopathic thrombocytopenic purpura (ITP), and diabetes mellitus (DM). Fifteen patients (20.2%) reported urticaria after a trigger (hot water, water, cold, pressure). Of all CU patients, 60 (81.1%) were diagnosed with CSU and 14 (18.9%) with Chronic inducible urticaria. According to diagnostic provocation tests, the most common type among the 14 patients with CIU was symptomatic dermographism (78.6%), followed by aquagenic urticaria (21.4%). Demographic characteristics of the patients are summarized in Table I.

Laboratory results showed that 10 of 74 (13.5%) patients had eosinopenia, four (5.4%) eosinophilia and two (2.7%) basopenia. There were 24 patients (32.4%) with a positive skin prick test. Allergic sensitization via inhalation was detected in 13 (17.6%), food sensitization in eight (10.8%), and combined inhalation and food sensitization in three (4%) patients. ANA positivity was found in seven (9.4%) of all patients, and no patient developed rheumatologic disease during the follow-up period. Antithyroid peroxidase antibody positivity was detected in four patients (5.4%) and no autoimmune thyroiditis was detected during follow-up. Thirteen patients (4%) showed elevated ESR and seven (9.4%) elevated CRP as acute phase reactants. Urinalysis detected signs of infection in 11 patients (14.9%). The laboratory test results are presented in Table II.

Table I: Clinical characteristics of children with chronic urticaria

Variables	Children with Chronic Urticaria (n=74, %)
Age at presentation (year)*	11±5.1 (2.3-17.9)
Mean follow-up time (days)*	475±237.2 (129-925)
Gender†	
Female	41 (55.4)
Male	33 (44.6)
Presence of angioedema†	16 (21.6)
Presence of a trigger (hot water, water, cold, pressure)†	10 (13.5)
Hot water	5 (6.7)
Cold	2 (2.7)
Water	2 (2.7)
Stress	1 (1.3)
Dermographism†	11 (14.8)
Comorbidities†	
Asthma	7 (9.4)
Allergic rhinitis	2 (2.7)
Hypothyroidism	2 (2.7)
Familial mediterranean fever	1 (1.3)
Idiopathic thrombocytopenic purpura	1 (1.3)
Diabetes mellitus	1 (1.3)
Family history†	
Chronic urticaria (mother/father)	4 (5.4)
Asthma/Allergic rhinitis (mother/father)	2 (2.7)
Rheumatologic disease (mother/father)	1 (1.35)
Chronic spontaneous urticaria†	60 (81.1)
Chronic inducible urticaria†	14 (18.9)
Dermographism	11 (14.9)
Aquagenic (cold) urticaria	3 (4)

***:** *mean±standard deviation (min-max),* **†:** *n(%)*

Table II: Laboratory results of children with chronic urticaria

Variables	Children with chronic urticaria (n=74, %)
Eosinopenia	10 (13.5)
Eosinophilia	4 (5.4)
Basopenia	2 (2.7)
Mean IgE level, average*	133.87±152 (17-688)
Low IgE <40 kU/L	16 (21.6)
High IgE >100 kU/L	21 (28.4)
Positive for antithyroid peroxidase antibodies	4 (5.4)
Positive for antinuclear antibodies	7 (9.4)
Skin prick test positivity	24 (32.4)
Inhalant allergen sensitivity	13 (17.6)
Food sensitivity	8 (10.8)
Inhalation and food sensitivity	3 (4)
Elevated erythrocyte sedimentation rate	3 (4)
Elevated C-reactive protein	7 (9.4)
Urine analysis	
Leukocyturia	11 (14.7)
Hematuria	2 (2.7)
Proteinuria	2 (2.7)

***:** *mean±standard deviation (min-max)*

As a first line of treatment, all 74 patients received antihistamine therapy at the standard dose. For patients who did not respond, the dose was increased incrementally up to two, three, or four times the initial dose. Montelukast was added to the drug regimen in 28 patients with no partial response despite the increased antihistamine doses. Twenty-two patients responded well to combined therapy with antihistamine and montelukast. Omalizumab was commenced in six patients with partial response. The details of drug regimens administered to patients are presented in Figure 1. Among the six patients who received omalizumab therapy, one (16.7%) presented with angioedema, one (16.7%) asthma, one (16.7%) exhibited both eosinopenia and basopenia, two (33.3%) tested positive for ANA, and one (16.7%) patient showed elevated levels of both ESR and CRP.

We managed to contact 44 patients (59.4%) via telephone, and 22 (50%) of these patients had achieved complete remission. Of the other 22 patients, nine (20.4%) had well-controlled symptoms, six (13.6%) partially controlled and seven (16%) uncontrolled. When the patients were called by phone, 9-12 months had passed. All patients with partially controlled or uncontrolled signs and symptoms reported to experience drug compliance issues.

Table III presents a comparison of laboratory results across different disease control categories: remission-well-controlled

Table III: Comparison of laboratory parameters in children with chronic urticaria by disease control status: remission, good control, partial control, and no control

Variables	Remission and well controlled (n=31)	Partially controlled and uncontrolled (n=13)	p
Eosinopenia*	10 (13.5)	3 (23.1)	0.650
Eosinophilia*	-	3 (23.1)	
Basopenia*	2 (6.4)	-	
Elevated ESR*	2 (6.4)	-	
Elevated CRP*	3 (9.7)	1 (7.7)	
Positive for antithyroid peroxidase antibodies	2 (6.4)	1 (7.7)	
Positive for antinuclear antibodies	3 (9.7)	2 (15.4)	0.780
Skin prick test positivity*	13 (61.9)	5 (71.4)	0.690
IgE level†	104 (45.2-202)	68 (42-235)	0.700

**: n(%), †: median (interquartile range)*

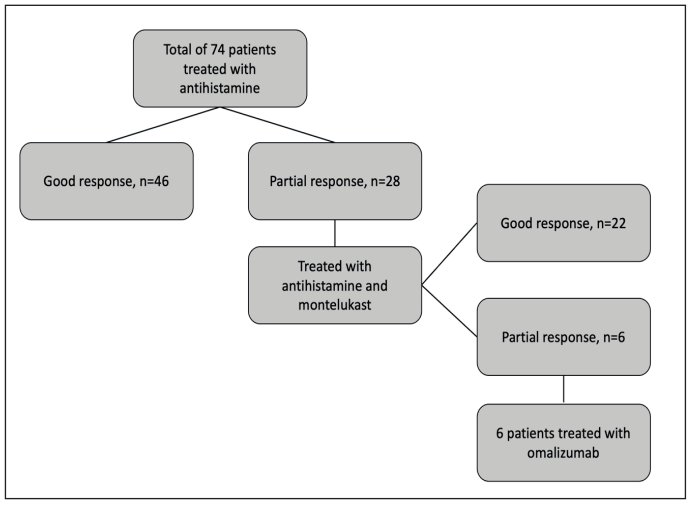

Figure 1: Treatment regimen flow chart for children with chronic urticaria}

and partially controlled-uncontrolled. Eosinopenia, ANA positivity, anti-TPO positivity were proportionally higher in patients achieving remission and exhibiting well-controlled chronic urticaria, although there was no statistically significant difference. Serum IgE levels were similarly higher in both patient groups, albeit with no significant statistical difference.

DISCUSSION

In this study, we evaluated the clinical and etiologic features, laboratory findings, and treatment response of 74 CU patients who were followed up in our clinic. The female patients constituted the majority of the sample. The most common accompanying symptom was angioedema, while the most common comorbidity was asthma. Family history included chronic urticaria, asthma and rheumatologic diseases. CSU was detected in about 80% of our patients, and the remaining ones were diagnosed with CIU by provocation tests. Evidence-based information from clinical trials or real-life studies on epidemiology, comorbidities and treatment outcomes in the pediatric population is scarce, thus there are gaps in the management of CU in children.

In literature, pediatric patients receiving a diagnosis of CU are predominantly female, with a mean age range of 8-11 years (14, 15). In our cohort, the majority of patients were female (55.4%), and the mean age was in accordance with the findings reported in the literature. Angioedema was observed in 21.6% of our patients. Prior research has documented comparable incidences of angioedema ranging from 30% to 50%, which renders it as the most frequently occurring symptom associated with CU (15) .

While the association between CU and atopy remains to be fully elucidated, some studies suggest a potential link. Previous research has demonstrated correlations between atopy, comorbid allergic conditions, and CU, with children diagnosed with CU exhibiting a higher prevalence of allergic diseases or sensitization (16). In our case series, atopy was observed in 32.7% of patients with CU, a figure consistent with previous findings (17). Furthermore, 14.9% of our cohort had a diagnosis of asthma and/or allergic rhinitis, while 32.4% demonstrated sensitization on skin prick testing. A family history of atopy was noted in 8.1% of patients. Although food sensitization was identified in some individuals, no specific association with food-induced urticaria was reported; therefore, food challenge testing was not conducted.

The potential role of autoimmunity in chronic urticaria should also be investigated. In line with the EAACI/WAO guideline diagnostic algorithm, assessment for autoimmunity (chronic autoimmune or autoantibody-associated CU) is recommended in selected patients with chronic spontaneous urticaria who exhibit an inadequate response to standard treatment (1). Within our sample, three patients (4%) presented with concomitant autoimmune conditions, including DM, FMF, and ITP; TPO antibodies were detected in four patients (5.4%), and seven (9.4%) tested positive for ANA. No patients developed any new autoimmune diseases during the follow-up period.

CSU accounts for 50-75% of chronic urticaria cases, and CIU represents about one-third of this subset. Dermographism is the most common manifestation of CIU (18). Our case series closely align with these literature findings, as 81.1% had CSU and 18.9% CIU, predominantly dermographism.

Current guidelines recommend antihistamines as first-line therapy for the management of chronic urticaria, while second-line treatment should involve up-titration to a maximum of four times the standard dose. Omalizumab is recommended for patients aged over 12 not responding to such therapies (7). Previous studies have reported that 7% to 10.7% of pediatric patients diagnosed with chronic urticaria require omalizumab (19, 20). In support of these data, 8.1% of our patients received omalizumab.

Even though chronic urticaria is often self-limiting, about 20% of patients experience symptoms for more than 5 years (21). In our study, 29.5% of patients remained partially controlled or uncontrolled after a mean follow-up period of 1.5 years, underscoring the importance of continued monitoring and improvement of treatment strategies.

In patients with chronic spontaneous urticaria, complete blood count results typically fall within the normal range, however the presence of eosinopenia (<50 mm^3) is often associated with a poorer response to antihistamines and omalizumab (11). Likewise, although the majority of CSU patients exhibit normal CRP and ESR levels, significant elevations in either marker could be linked to worse outcomes, including reduced quality of life and diminished antihistamine efficacy (22). For patients failing to respond to omalizumab therapy, generally cyclosporine is recommended, but none of our patients required cyclosporine as their urticaria was effectively managed with omalizumab.

This study is subject to several limitations. First of all, its retrospective design can inherently cause some recall bias. Secondly, failure to contact some patients for follow-up may have impacted the overall findings. Finally, the relatively small sample size should limit the generalizability of our results.

CONCLUSION

The findings suggest that chronic urticaria is often self-limiting without the need for omalizumab in pediatric patients. Drug compliance should be evaluated in patients with poor disease control. Although the number of patients treated with omalizumab was relatively low, they responded well to this therapy. More studies are needed to evaluate etiology and treatment response in patients with chronic urticaria.

Ethics committee approval

This study was conducted in accordance with the Helsinki Declaration Principles. Ethical approval was obtained from the ethics committee of Selcuk University Faculty of Medicine (approval no: 2024/116 date: 27.02.2024).

Contribution of the authors

Yılmaz SY: Planning methodology to reach the conclusions, Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments, Taking responsibility in logical interpretation and conclusion of the results, Taking responsibility in necessary literature review for the study, Taking responsibility in the writing of the whole or important parts of the study, Reviewing the article before submission scientifically besides spelling and grammar. ***Külhaş Çelik İ:*** Planning methodology to reach the conclusions, Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments, Taking responsibility in logical interpretation and conclusion of the results, Taking responsibility in necessary literature review for the study, Reviewing the article before submission scientifically besides spelling and grammar. ***Artaç H:*** Constructing the hypothesis or idea of research and/or article, Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments, Taking responsibility in logical interpretation and conclusion of the results, Taking responsibility in necessary literature review for the study, Taking responsibility in the writing of the whole or important parts of the study, Reviewing the article before submission scientifically besides spelling and grammar.

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Conflict of interest

The authors declare that there is no conflict of interest.

REFERENCES

1. Arga M, Asılsoy S, Cetinkaya F, Oner Erkekol F, Gelıncık A, Gul U, et al. Turkish National Society of Allergy and Clinical Immunology Position Paper: Diagnosis and Management of Urticaria. Asthma Allergy Immunol 2018; 16:97-122.
2. Bernstein JA, Lang DM, Khan DA, Craig T, Dreyfus D, Hsieh F, et al. The diagnosis and management of acute and chronic urticaria: 2014 update. J Allergy Clin Immunol 2014;133:1270-7. https://doi.org/10.1016/j.jaci.2014.02.036
3. Powell RJ, Leech SC, Till S, Huber PA, Nasser SM, Clark AT. British Society for Allergy and Clinical Immunology. BSACI guideline for the management of chronic urticaria and angioedema. Clin Exp Allergy 2015;45:547-65. https://doi.org/10.1111/cea.12494
4. Caffarelli C, Paravati, F, El Hachem M, Duse M, Bergamini M, Simeone G, et al. Management of chronic urticaria in children: A clinical guideline. Ital J Pediatr. 2019; 45:101. https://doi.org/10.1186/s13052-019-0695-x
5. Zuberbier T, Abdul Latiff AH, Abuzakouk M, Aquilina S, Asero R, Baker D, et al. The international EAACI/GA2LEN/EuroGuiDerm/APAACI guideline for the definition, classification, diagnosis, and management of urticaria. Allergy 2022;77:734-66. https://doi.org/10.1111/all.15090

6. Greiner B, Nicks S, Adame M, McCracken J. Pathophysiology, Diagnosis, and Management of Chronic Spontaneous Urticaria: A Literature Review. Clin Rev Allergy Immunol 2022 ;63:381-9. https://doi.org/10.1007/s12016-022-08952-y
7. Saini SS, Kaplan AP. Chronic Spontaneous Urticaria: The Devil's itch. J Allergy Clin Immunol Pract 2018;6:1097-106. https://doi.org/10.1016/j.jaip.2018.04.013
8. Chin ML. It Comes and Goes. Pediatric Chronic Spontaneous Urticaria. Cureus 2023;15: e 40006. https://doi.org/10.7759/cureus.40006
9. Zhao ZT, Ji CM, Yu WJ, Meng L, Hawro T, Wei JF, et al. Omalizumab for the treatment of chronic spontaneous urticaria: a meta-analysis of randomized clinical trials. J Allergy Clin Immunol 2016;137:1742-50. https://doi.org/10.1016/j.jaci.2015.12.1342
10. Shah AD, Denaxas S, Nicholas O, Hingorani AD, Hemingway H. Low eosin- ophil and low lymphocyte counts and the incidence of 12 cardiovascular dis- eases: a CALIBER cohort study. Open Heart 2016;3:e000477. https://doi.org/10.1136/openhrt-2016-000477
11. Kolkhir P, Church MK, Altrichter S, Skov PS, Hawro T, Frischbutter S, et al. Eosinopenia, in Chronic Spontaneous Urticaria, Is Associated with High Disease Activity, Autoimmunity, and Poor Response to Treatment. J Allergy Clin Immunol Pract 2020; 8:318-325.e15. https://doi.org/10.1016/j.jaip.2019.08.025
12. Altrichter S, Fok JS, Jiao Q, Kolkhir P, Pyatilova P, Romero SM, et al. Total IgE as a marker for c hronic spontaneous urticaria. Allergy Asthma Immunol Res 2021;13:206-18. https://doi.org/10.4168/aair.2021.13.2.206
13. Ansotegui IJ, Melioli G, Canonica GW, Caraballo L, Villa E, Ebisawa M, et al. IgE allergy diagnostics and other relevant tests in allergy, a World Allergy Organization position paper. World Allergy Organ J 2020;25;13:100080. https://doi.org/10.1016/j.waojou.2019.100080
14. Chansakulporn S, Pongpreuksa S, Sangacharoenkit P, Pacharn P, Visitsunthorn N, Vichyanond P, et al. The natural history of chronic urticaria in childhood: a prospective study. J Am Acad Dermatol 2014;71:663-8. https://doi.org/10.1016/j.jaad.2014.05.069
15. Lachover-Roth I, Rabie A, Cohen-Engler A, Rosman Y, Meir-Shafrir K, Confino-Cohen R. Chronic urticaria in children - new insights from a large cohort. Pediatr Allergy Immunol 2021;32:999-1005. https://doi.org/10.1111/pai.13457
16. Park YM, Oh MS, Kwon JW. Predicting inadequate treatment response in children with chronic spontaneous urticaria. Pediatr. Allergy Immunol 2020;31:946-53. https://doi.org/10.1111/pai.13319
17. Sahiner UM, Civelek E, Tuncer A, Yavuz ST, Karabulut E, Sackesen C, et al. Chronic urticaria: Etiology and natural course in children. Int Arch Allergy Immunol 2011;156, 224-30. https://doi.org/10.1159/000322349
18. Sánchez-Borges M, Asero R, Ansotegui IJ, Baiardini I, Bernstein JA, Canonica GW, et al; WAO Scientific and Clinical Issues Council. Diagnosis and treatment of urticaria and angioedema: a worldwide perspective. World Allergy Organ J 2012;5:125-47. https://doi.org/10.1097/WOX.0b013e3182758d6c
19. Özçeker D, Can PK, Terzi Ö, Ornek SA, Degirmentepe EN, Kızıltac K, et al. Differences between adult and pediatric chronic spontaneous urticaria from a cohort of 751 patients: Clinical features, associated conditions and indicators of treatment response. Pediatric Allergy and Immunology 2023;34:e13925. https://doi.org/10.1111/pai.13925
20. Özhan AK, Arıkoğlu T. Evaluation of Pediatric Chronic Urticaria with Emphasis on Clinical and Laboratory Characteristics and Treatment Response to Omalizumab: A Real-Life Experience from a Tertiary Allergy Center. Children (Basel) 2024;11:86. https://doi.org/10.3390/children11010086
21. Sarbjit S Saini, Allen P Kaplan. Chronic Spontaneous Urticaria: The Devil's Itch. Clin Immunol Pract 2018;6:1097-106. https://doi.org/10.1016/j.jaip.2018.04.013
22. Kolkhir P, Altrichter S, Hawro T, Maurer M. C-reactive protein is linked to disease activity, impact, and response to treatment in patients with chronic spontaneous urticaria. Allergy 2018;73:940-8. https://doi.org/10.1111/all.13352

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Review

An overview of the etiology of vaccine hesitancy and refusal

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ABSTRACT

Vaccination is among the most cost-effective interventions for protecting populations against infectious diseases. Individuals exhibiting vaccine hesitancy may eventually accept some or all vaccines, may delay them, or may reject specific ones. In contrast, vaccine refusal denotes the complete voluntary rejection of all vaccines in the current immunization program. Vaccine refusal represents a significant public health con-cern, threatening both individual and community health. Opposition to vaccines has existed since their inception and continues to grow, particularly with the influence of the internet and social media. Some of the reasons for vaccine refusal include concerns about the vaccine components, distrust in vaccines, religious beliefs, reservations about the pharmaceutical industry, and fear of adverse effects. This review aims to explore the etiology of vaccine refusal based on current literature.

Keywords: Child, Etiology, Vaccine Hesitancy, Vaccine Refusal

INTRODUCTION

Vaccines are biological products that confer protection against targeted diseases and are derived from attenuated microorganisms, their toxins, or surface antigens (1). Immunization programs rank among the most cost-effective public health strategies, significantly decreasing morbidity and mortality, preventing and eradicating infectious diseases, and promoting public health (2). According to the World Health Organization (WHO), vaccine hesitancy is the delay in acceptance or refusal of vaccines despite the availability of vaccination services. It is a complex and context-specific issue influenced by factors such as complacency, convenience, and confidence. Individuals exhibiting vaccine hesitancy may eventually accept some or all vaccines, may delay them, or may reject specific ones. In contrast, vaccine refusal denotes the complete voluntary rejection of all vaccines in the current immunization program. Figure 1 shows vaccine hesitancy process (3).

1- History of anti-vaccination movements

To understand the origins of vaccine refusal and hesitancy, it is essential to consider the historical background.

Early anti-vaccination movements

Resistance to vaccination emerged in 18th-century England, where religious leaders argued that disease was a divine punishment and that preventing it was a defiance of God's will. Between 1840 and 1853, mandatory smallpox vaccination led to the establishment of the anti-vaccination league in London, which opposed compulsory immunization on grounds of personal liberty (4). Sanctions, including imprisonment, against those who refused vaccination generated public outrage (2). In 1867, the vaccination mandate was extended to children up to 14 years old, prompting the formation of the anti-compulsory vaccination league (5). Anti-vaccine publications in the 1870s and 1880s reduced vaccine uptake across Europe and the United States of America (USA), resulting in outbreaks such as the 1874 smallpox epidemic in Stockholm, which claimed over 4.000 lives (4,6). In 1898, under growing pressure, the British Parliament introduced the concept of the "conscientious objector," allowing parents to exempt their children from vaccination (7).

Anti-vaccination movements in USA

The first American anti-vaccine conference was held in 1907, led by J. Pitcairn, who later founded the Anti-vaccination league of

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America in Philadelphia. A parallel movement in Brazil, led by O. Cruz, also resisted vaccination (8). Despite these oppositions, the 1950s and 1960s are regarded as the golden era of vaccine acceptance, marked by successful immunization campaigns against polio, measles, and rubella (9).

Polio vaccine: the cutter incedent

In 1955, a polio vaccine produced by Cutter Laboratories was found to contain live poliovirus, resulting in approximately 70.000 mild infections, 200 cases of paralysis, and 10 deaths. This incident significantly eroded public trust in vaccine safety (10).

The diphtheria, tetanus, and pertussis (DTP) vaccine controversy

Concerns about the DTP vaccine arose in 1974 following a report linking it to neurological complications in children (11). In 1982, the documentary "DTP Vaccine Roulette" and the book "Shots in the Dark" further fueled fears, alleging that the vaccine caused seizures and permanent brain damage. These claims led to a substantial decline in vaccination coverage and a pertussis outbreak in the England (10).

Swine flu vaccine and Guillain-Barré Syndrome (GBS)

In 1976, the USA launched a nationwide vaccination campaign against swine flu. Shortly thereafter, reports emerged of an increased incidence of GBS. Although subsequent studies found that the risk was minimal, public anxiety persisted (12,13). Similar allegations surfaced in France in the 1990s, where hepatitis B vaccination was erroneously linked to multiple sclerosis. Later research disproved this association (12,14).

The measles, mumps, and rubella (MMR) vaccine controversy

In 1998, Andrew Wakefield published a now-retracted study in The Lancet that falsely claimed a link between the MMR vaccine and autism in children. The article received extensive media coverage and significantly contributed to vaccine refusal worldwide, despite being discredited (15).

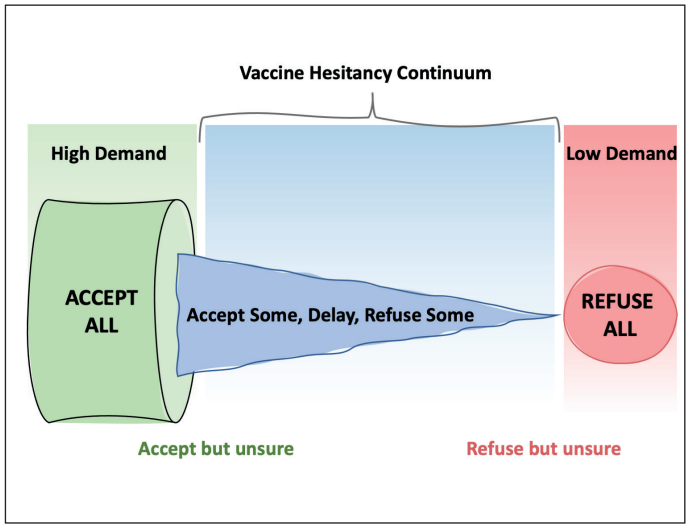

Figure 1: Vaccine hesitancy process

Vaccine and infertility

In 2003–2004, Muslim leaders and politicians in Northern Nigeria alleged that oral polio vaccines were part of a Western plot to induce infertility and spread HIV. This resulted in widespread vaccine refusal and a resurgence of polio cases (11,16).

Vaccine hesitancy/refusal in the modern era

Since the 2000s, the rise of the internet and social media has facilitated the rapid spread of anti-vaccine rhetoric. Public figures expressing anti-vaccine views have further legitimized these beliefs. Online communities and blogs dedicated to vaccine skepticism have emerged, influencing public opinion and reinforcing vaccine hesitancy (17). Figure 2 shows the timeline of vaccine refusal (10).

2- Etiology of vaccine hesitancy/refusal

The introduction of vaccination has consistently been accompanied by opposition. The earliest documented resistance emerged in 18th-century England, driven by religious objections (4). Over time, the underlying causes of vaccine hesitancy/refusal have diversified and are now categorized into three main domains: contextual influences, individual and group influences, and vaccine- or vaccination-specific issues (Table I) (3).

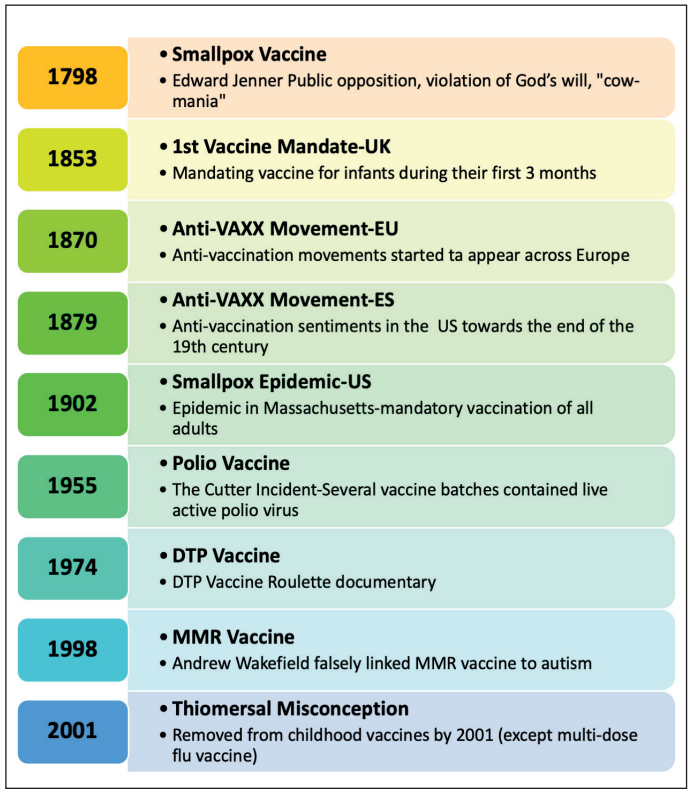

Figure 2: Vaccine hesitancy/refusal timeline

Vaccine and mercury

Contrary to widespread misconceptions, the form of mercury used in vaccines is thiomersal, an ethylmercury compound. The toxic effects associated with mercury arise from methylmercury, which is not present in vaccines. Scientific studies have shown no evidence of neurotoxicity or chronic accumulation of thiomersal. Due to the financial and logistical burdens of producing thiomersal-free single-dose vials, the WHO supports the continued use of multi-dose thiomersal-containing vaccines (18).

Vaccine and autism

A primary argument of anti-vaccine groups is the alleged link between vaccines and autism spectrum disorder (ASD), often based on methodologically flawed studies. Although rising ASD prevalence is frequently blamed on vaccines, it is more accurately attributed to expanded diagnostic criteria, improved detection methods, and increased awareness among families and professionals. A large-scale meta-analysis encompassing over 1.2 million children found no correlation between ASD and either thiomersal exposure or MMR vaccination (19,20,21).

Vaccines, pharmaceutical industry and conflict of interest

Public skepticism toward the pharmaceutical industry is frequently cited in vaccine refusal (22). Reports indicate that drug promotion expenditures totaled $28 billion in the USA, $20 billion across several European nations, and over $26 billion in Japan as of 2012 (23). Despite the industry's contributions to medical advancement, mistrust arises due to perceived commercial motives and inadequate physician awareness regarding drug pricing (24). In Türkiye, all vaccines are imported and undergo regulatory testing; however, public concerns about pharmaceutical companies' influence persist, contributing to vaccine hesitancy/refusal (22).

Table I: Factors affecting vaccine hesitancy/refusal

Contextual Influences
• Communication and media environment
• Influential leaders, immunization program gatekeepers and anti or pro-vaccination lobbies.
• Historical influences
• Religion/culture/ gender/socio-economic
• Politics/policies
• Geographic barriers
• Perception of the pharmaceutical industry
Individual and group influences
• Personal, family and/or community members' experience with vaccination, including pain
• Beliefs, attitudes about health and prevention
• Knowledge/awareness
• Health system and providers-trust and personal experience.
• Risk/benefit (perceived, heuristic)
• Immunisation as a social norm etc. not needed/ harmful
Vaccine/vaccination spesific issues
• Risk/ benefit (epidemiological and scientific evidence)
• Introduction of a new vaccine or new formulation or a new recommendation for an existing vaccine
• Mode of administration
• Design of vaccination program/mode of delivery (e.g., routine program or mass vaccination campaign)
• Reliability and/or source of supply of vaccine and/or vaccination equipment
• Vaccination schedule
• Costs
• The strength of the recommendation and/or knowledge base and/or attitude of healthcare professionals

The role of social media

In the digital era, social media platforms have become a primary source of health-related information. Parents often seek vaccine-related advice on platforms such as facebook and instagram, where misinformation from non-expert individuals is prevalent. The amplification of biased content, often promoted by celebrities and influencers, undermines public trust in vaccination (25,26). A 2023 report from the Turkish Statistical Institute highlighted whatsApp, youtube, and instagram as the most commonly used platforms (27). Unlike traditional media, social media allows rapid dissemination of unverified content, exacerbating information pollution (26). Enhancing digital health literacy and actively monitoring social media are vital to combating misinformation.

Vaccine hesistancy/refusal and religion

Religious concerns, particularly in Muslim communities, have centered around the origin of gelatin used in vaccines. Gelatin, employed to stabilize vaccine components, can be sourced from cattle, poultry, or pigs. Due to religious sensitivities, vaccines administered in Türkiye use only bovine-derived gelatin (28). Furthermore, in 1995, the World Islamic Health Federation stated that gelatin, even when derived from pigs, undergoes transformation processes that render it religiously permissible (29). Another religious objection to vaccination stems from the belief that it interferes with divine will (30). However, no religious texts explicitly forbid vaccination, and immunization is mandated for participation in Hajj and Umrah pilgrimages.

Perceived unnecessity and ineffectiveness of vaccines

Some individuals argue that vaccines are unnecessary, citing natural immunity, the low prevalence of diseases, or reliance on traditional therapies such as herbal medicine or cupping. Others believe that vaccine-induced immunity is passive and short-lived, requiring repeated booster doses (31). Parents who experienced illnesses such as measles or mumps without complications may prefer natural infection for their children, believing it confers lifelong benefits (32). Mistrust in public health data and skepticism toward reported efficacy rates also contribute to vaccine refusal (33). However, data from the Centers for Disease Control and Prevention (CDC) demonstrate substantial reductions in disease incidence post-vaccination, with declines of 96% to 100% for several vaccine-preventable diseases (Table II) (34).

Table II: Morbidity before and after vaccination

Disease	Pre vaccine era estimated annuel morbidity	After vaccıne estimates u.s.a cases	Percent decrease (%)
Polio (paralytic)	16.316	0[2]	100
Diphtheria	21.053	2[2]	>99
H. influenzae serotype B (invasive, <5 years of age)	20.000	18[2]	>99
Hepatitis A	117.333	37.700[3]	68
Measles	530.217	1.275[2]	>99
Meningococcal disease (all serotypes)	2.886[4]	371[2]	87
Pertussis	200.752	18,.617[2]	91
Pneumococcal disease (invasive, <5 years of age)	16.069	1.700[5]	89
Rotavirus (hospitalizations, <3 years of age)	62.500[6]	30.625[7]	51
Rubella	47.745	6[2]	>99
Smallpox	29.005	0[2]	100
Varicella	4.085.210	8.297[8]	>99
Tetanus	580	26[2]	96

Vaccine hesistancy/refusal and complementary/ alternative medicine

The popularity of complementary and alternative medicine has increased significantly, driven by media coverage and cultural preferences. These approaches are perceived as natural, low-risk, and free from corporate influence (35). Many individuals believe such treatments contain fewer synthetic chemicals and are safer than conventional pharmaceuticals. Parents may choose alternative therapies due to concerns about vaccine ingredients, side effects, or doubts regarding vaccine efficacy (35,36).

Vaccine hesistancy/refusal and population reduction policy

In certain low-income and minority communities, conspiracy theories suggest that vaccines are used by Western countries as tools of population control, particularly to reduce fertility (37). A study in Pakistan found that vaccine refusal was attributed to fears of infertility (3.3%), perceived ineffectiveness (21.6%), and general distrust (40.2%) (38).

Vaccine hesitancy/refusal in in Türkiye

Although vaccination is mandatory in Türkiye, immunization rates were 75% before 2007 but rose to 95% thereafter due to improved access. Since 2010, however, vaccine refusal has gained attention. A pivotal event occurred in 2015, when a court ruled in favor of a prosecutor who refused vaccination for his twins, citing individual rights and consent. The media portrayed this as a legal victory, fueling public debate and encouraging vaccine refusal (2,39). Consequently, the number of families refusing vaccines increased from 183 in 2011 to 23.600 by 2018 (40).

CONCLUSION

In conclusion, addressing vaccine hesitancy and refusal requires a multifaceted, evidence-based approach that integrates transparent risk communication, trust-building with healthcare professionals, and proactive countering of misinformation particularly on digital platforms (41,42). Ensuring equitable vaccine access, especially for underserved populations, remains essential for promoting uptake (43). Educational interventions tailored to cultural and social contexts, along with the integration of vaccine literacy into broader health education, have also shown promise in fostering informed decision-making (43,44). A coordinated strategy involving health authorities, educators, and local communities is vital to restoring public confidence and maintaining high vaccination coverage (41,42).

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REFERENCES

1. World Health Organization (WHO). How do vaccines work? [Internet]. [cited 2023 Jul 12]. Available from: https://www.who.int/news-room/feature-stories/detail/how-do-vaccines-work
2. Bekis Bozkurt H. An overview of vaccine rejection and review of literature. Kafkas J Med Sci. 2018;8(1):71-6. https://doi.org/10.5505/kjms.2018.12754
3. Report of the SAGE Working Group on Vaccine Hesitancy [Internet]. 2014 Nov 12. [cited 2023 Jul 12].
4. Nelson MC, Rogers J. The right to die? Anti-vaccination activity and the 1874 smallpox epidemic in Stockholm. Soc Hist Med. 1992;5(3):369-88. https://doi.org/10.1093/shm/5.3.369
5. Durbach N. Bodily matters: The anti-vaccination movement in England, 1853-1907. Durham, NC: Duke University Press; 2005. https://doi.org/10.1215/9780822386506
6. Prabhu M. Ye olde anti-vaxxers. Gavi, the Vaccine Alliance [Internet]. [cited 2023 Jul 19]. Available from:
7. Swales JD. The Leicester anti-vaccination movement. Lancet. 1992;340(8826):1019-21. https://doi.org/10.1016/0140-6736(92)93021-E
8. Wolfe RM, Sharp LK. Anti-vaccinationists past and present. BMJ. 2002;325(7361):430-2. https://doi.org/10.1136/bmj.325.7361.430
9. Ołpiński M. Anti-vaccination movement and parental refusals of immunization of children in USA. Pediatr Pol. 2012;87(4):381-5. https://doi.org/10.1016/j.pepo.2012.05.003
10. Nuwarda RF, Ramzan I, Weekes L, Kayser V. Vaccine hesitancy: Contemporary issues and historical background. Vaccines (Basel). 2022;10(2):247. https://doi.org/10.3390/vaccines10101595

11. Kulenkampff M, Schwartzman JS, Wilson J. Neurological complications of pertussis inoculation. Arch Dis Child. 1974;49(2):46-9. https://doi.org/10.1136/adc.49.1.46
12. Badur S. Anti-vaccination: vaccine hesitancy and refusal. Ankem Derg. 2011;25(2):82-6.
13. Centers for Disease Control and Prevention (CDC). Guillain-Barré Syndrome and vaccines [Internet]. [cited 2023 Jul 19]. Available from: https://www.cdc.gov/vaccinesafety/concerns/guillain-barre-syndrome.html
14. Confavreux C, Suissa S, Saddier P, Bourdes V, Vukusic S. Vaccinations and the risk of relapse in multiple sclerosis. N Engl J Med. 2001;344(5):319-26. https://doi.org/10.1056/NEJM200102013440501
15. Wakefield AJ, Murch SH, Anthony A, Linnell J, Casson DM, Malik M, et al. Retracted: Ileal-lymphoid-nodular hyperplasia, non-specific colitis, and pervasive developmental disorder in children. Lancet. 1998;351(9103):637-41. https://doi.org/10.1016/S0140-6736(97)11096-0
16. Jegede AS. What led to the Nigerian boycott of the polio vaccination campaign? PLoS Med. 2007;4(3):e73. https://doi.org/10.1371/journal.pmed.0040073
17. Smith TC. Vaccine rejection and hesitancy: A review and call to action. Open Forum Infect Dis. 2017;4(3):ofx146. https://doi.org/10.1093/ofid/ofx146
18. Kara A, Çiftçi E, Tezer H, Somer A, editors. Aşı ve Bağışıklama. Aşılar ve Civa İddiaları. 1st ed. Ankara: Selen Medya Yayıncılık. 2021; 877-878.
19. Kara A, Çiftçi E, Tezer H, Somer A, editors. Aşı ve Bağışıklama. Aşılar ve Otizm İddiaları. 1st ed. Ankara: Selen Medya Yayıncılık; 2021:897-904.
20. Taylor LE, Swerdfeger AL, Eslick GD. Vaccines are not associated with autism: An evidence-based meta-analysis of case-control and cohort studies. Vaccine. 2014 ;32(29):3623-9. https://doi.org/10.1016/j.vaccine.2014.04.085
21. T.C. Sağlık Bakanlığı. Aşılar ile otizm arasında ilişki var mıdır? [Internet]. [cited 2023 Jul 25]. Available from: https://asi.saglik.gov.tr
22. Bults M, Beaujean DJMA, Richardus JH, van Steenbergen JE, Voeten HACM. Pandemic influenza A (H1N1) vaccination in The Netherlands: Parental reasoning underlying child vaccination choices. Vaccine. 2011;29(37):6226-35. https://doi.org/10.1016/j.vaccine.2011.06.075
23. Yağcı-Küpeli B, Kiper N. Interactions between physicians and drugindustry. Cukurova Med J. 2016;41:777-81. https://doi.org/10.17826/cutf.254526
24. Ağalar C, Ağalar F, Sayek İ. The interactions between doctorsand pharmaceutical companies. Çocuk Sağlığı ve Hastalıkları Dergisi. 2005;48:273-83.
25. Taşçı Ö, Gökler ME. Aşı karşıtlarının sosyal medya paylaşımlarının incelenmesi: Instagram örneği. Med Res Rep. 2021;4(2):23-30.
26. Etesaminia S, Bağcı Derinpınar K. The role of social media in vaccıne hesitation. Usaysad Derg. 2021;7(2):377-90.
27. Türkiye İstatistik Kurumu (TÜİK). Hanehalkı Bilişim Teknolojileri Kullanım Araştırması 2022 [Internet]. [cited 2023 Jul 27]. Available from: https://data.tuik.gov.tr
28. T.C. Sağlık Bakanlığı. Aşı içerikleri [Internet]. [cited 2023 Jul 13]. Available from: https://covid19asi.saglik.gov.tr
29. Büyükkalay BN. Vaccine Rejection and Motivation Sources of Vaccine Rejected People. Int Soc Mentality Res Thinkers J. 2022;8(65):2116-26. https://doi.org/10.29228/smryj.65987
30. Fourn L, Haddad S, Fournier P, Gansey R. Determinants of parents' reticence toward vaccination in urban areas in Benin (West Africa). BMC Int Health Hum Rights. 2009;9(Suppl 1):S1-5. https://doi.org/10.1186/1472-698X-9-S1-S14
31. The Liberty Beacon. The vaccine illusion: How vaccination compromises our natural immunity and what we can do to regain our health [Internet]. [cited 2023 Aug 10]. Available from: https://www.thelibertybeacon.com
32. McKee CP, Bohannon KB. Exploring the reasons behind parental refusal of vaccines. J Pediatr Pharmacol Ther. 2016;21(2):104-9. https://doi.org/10.5863/1551-6776-21.2.104
33. Dirlik R. Aşıyı ve tapınak tıbbını savunan katil. [Internet]. [cited 2023 Aug 10]. Available from: https://ruhadirlik.blogspot.com/2016/01/asiyi-ve-tapinak-tibbini-savunan.html
34. Immunization Action Coalition. Vaccine work [Internet]. Available from: https://www.immunize.org/catg.d/p4037.pdf
35. Hızel Bülbül S, Turgut M, Köylüoğlu S. Çocuklarda tıp dışı alternatif uygulamalar konusunda ailelerin görüşleri. Çocuk Sağlığı ve Hastalıkları Dergisi. 2009;52:195-202.
36. Çıtak G, Aksoy Duran Ö. An Important Obstacle in Vaccination: Vaccination Rejection. Erciyes Univ Health Sci Fac J. 2020;7(2):15-20.
37. Cobos Muñoz D, Monzón Llamas L, Bosch-Capblanch X. Exposing concerns about vaccination in low- and middle-income countries: A systematic review. Int J Public Health. 2015;60(7):767-80. https://doi.org/10.1007/s00038-015-0715-6
38. Khowaja AR, Khan SA, Nizam N, Bin Omer S, Zaidi A. Parental perceptions surrounding polio and self-reported non-participation in polio supplementary immunization activities in Karachi, Pakistan: A mixed methods study. Bull World Health Organ. 2012;90:822-30. https://doi.org/10.2471/BLT.12.106260
39. Cumhuriyet. Yargıtay'dan aşı kararına ikinci ret [Internet]. [cited 2023 Jul 19]. Available from: https://www.cumhuriyet.com.tr/haber/yargitaydan-asi-kararina-ikinci-ret-531726
40. Gür E. Vaccine hesitancy - vaccine refusal. Turk Pediatri Ars. 2019;54(1):1-2. https://doi.org/ 10.14744/TurkPediatriArs.2019.79990
41. Betsch C, Böhm R, Chapma GB. Using behavioral insights to increase vaccination policy effectiveness. Policy Insights from the Behavioral and Brain Sciences, 2018;5(1):61-9. https://doi.org/10.1177/2372732217752066
42. Larson HJ, Gakidou E, Murray CJL. The vaccine-hesitant moment. New England Journal of Medicine, 202;387(1):58-65. https://doi.org/10.1056/NEJMra2106441
43. World Health Organization. Behavioural and social drivers of vaccination: Tools and practical guidance for achieving high uptake. 2023
44. Kempe A, Saville AW, Albertin C, Zimet G, Breck A, Helmkamp L, Dickinson LM. Parental hesitancy about routine childhood and influenza vaccinations: A national survey. Pediatrics 2020;146(1):e20193852. https://doi.org/10.1542/peds.2019-3852