Diagnosis and Treatment of Newborns Referred to the Metabolism Department from the National Newborn Screening Program in Türkiye: A 5-Year Single-Center Experience

Türkiye'deki Ulusal Yenidoğan Tarama Programı Tarafından Metabolizma Bölümüne Yönlendirilen Yenidoğanların Tanı ve Tedavisi: Beş Yıllık Tek Merkez Deneyimi

Merve KOÇ YEKEDÜZ[1], Fatma Tuba EMİNOĞLU[1,2]

[1]Department of Pediatric Metabolism, Faculty of Medicine, Ankara University, Ankara, Türkiye
[2]Rare Diseases Application and Research Center, Ankara University, Ankara, Türkiye

ABSTRACT

Objective: The aims of this study were to investigate biochemical and genetic tests and treatment plans of newborns referred to our center with inherited metabolic disorders screened in Türkiye National Newborn Screening Program (NNSP).

Material and Methods: The medical records of babies referred by the NNSP between January 2019 and November 2023 were scanned retrospectively. Plasma biotinidase activity and the biotinidase gene (BTD) analysis results for suspected biotinidase deficiency (BD), the plasma phenylalanine and phenylalanine hydroxylase gene (PAH) analysis for a suspicion of phenylketonuria (PKU) were documented with treatment information.

Results: A total of 143 babies, 78 (54.5%) with suspected BD and 65 (45.5%) with suspected PKU were included. A PAH gene analysis was performed on 23 (35.4%) of those had high plasma phenylalanine levels, among which 86.9% were identified with the biallelic variant. Five patients were started on sapropterin-diet combined therapy, three on diet therapy and one on sapropterin therapy. In the first serum biotinidase activity measurement of babies referred with suspected BD, a heterozygous deficiency was detected in 48.7%, partial deficiency in 39.7% and profound deficiency in 10.3%. A BTD gene analysis was performed on 79.5% of those with suspected BD, and biallelic variants were detected in 50%. Forty-six patients (59.0%) underwent biotin treatment.

Conclusion: In our study, approximately one-third of the babies referred from NNSP over the five-year course of the study had biallelic variants of the relevant disease. Our research is one of the few studies on NNSP in our country and presents the diagnosis and treatment process of PKU and BD.

Key Words: Biotinidase deficiency, Neonatal Screening, Phenylketonuria, Türkiye

ÖZ

Amaç: Bu çalışmanın amacı, Türkiye Ulusal Yenidoğan Tarama Programı (UYTP)'de tarama yapılan kalıtsal metabolik hastalık şüphesiyle merkezimize yönlendirilen yenidoğanların biyokimyasal ve genetik testleri ile tedavi planlarını araştırmaktı.

0000-0003-0637-417X : KOÇ YEKEDÜZ M
0000-0002-5880-1113 : EMİNOĞLU FT

Conflict of Interest / *Çıkar Çatışması*: On behalf of all authors, the corresponding author states that there is no conflict of interest.

Ethics Committee Approval / *Etik Kurul Onayı*: This study was conducted in accordance with the Helsinki Declaration Principles. This study was approved by the Ankara University ethics committee (İ01-32-24, 18.01.2024).

Contribution of the Authors / Yazarların katkısı*: *KOÇ YEKEDÜZ M: Constructing the hypothesis or idea of research and/or article, 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, 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. ***EMİNOĞLU FT***: 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 the writing of the whole or important parts of the study, Reviewing the article before submission scientifically besides spelling and grammar.

How to cite / *Atıf yazım şekli* : Koç Yekedüz M and Eminoğlu FT. Diagnosis and Treatment of Newborns Referred to the Metabolism Department From the National Newborn Screening Program in Türkiye: A 5-Year Single-Center Experience. Turkish J Pediatr Dis 2024;18:274-280.

Correspondence Address / Yazışma Adresi:
Merve KOÇ YEKEDÜZ
Department of Pediatric Metabolism,
Ankara University Faculty of Medicine, Ankara, Türkiye
E-posta: drmervekoc13@hotmail.com

Received / Geliş tarihi : 17.03.2024
Accepted / Kabul tarihi : 15.04.2024
Online published : 10.05.2024
Elektronik yayın tarihi
DOI:10.12956/tchd.1454353

Gereç ve Yöntemler: Ocak 2019 ile Kasım 2023 tarihleri arasında UYTP tarafından yönlendirilen bebeklerin tıbbi kayıtları geriye dönük tarandı. Şüpheli biotinidaz eksikliği (BE) için plazma biyotinidaz aktivitesi ve BTD gen analiz sonuçları, fenilketonüri (FKÜ) şüphesi için plazma fenilalanin ve PAH gen analizi ile birlikte tedavi bilgileri araştırıldı.

Bulgular: Yetmiş-sekizi (%54.5) şüpheli BE ve 65'i (%45.5) şüpheli FKÜ olmak üzere toplam 143 bebek dahil edildi. Yüksek plazma fenilalanin seviyelerine sahip olanların 23'üne (%35.4) PAH gen analizi yapılmış, bunların %86.9'unda biallelik variant saptanmıştı. Beş hastada sapropterin-diyet kombinasyon tedavisine, üç hastada diyet tedavisine ve bir hastada sapropterin tedavisine başlanmıştı. Şüpheli BE ile yönlendirilen bebeklerin ilk serum biyotinidaz aktivite ölçümünde, %48.7'sinde heterozigot eksiklik, %39.7'sinde kısmi eksiklik ve %10.3'ünde derin eksiklik saptanmıştı. Şüpheli BE olan bebeklerin %79.5'ine BTD gen analizi yapılmış ve %50 oranında biallelik varyant saptanmıştı. Kırk altı hastaya (%59) biyotin tedavisi başlanmıştı.

Sonuç: Çalışmamızda, beş yıllık süre boyunca UYTP tarafından yönlendirilen bebeklerin yaklaşık üçte birinde ilgili hastalığın biallelik varyantları bulunduğu gösterildi. Araştırmamız, ülkemizde UYTP üzerine yapılan az sayıdaki çalışmalardan biridir ve FKÜ ve BE'nin tanı ve tedavi sürecini sunmaktadır.

Anahtar Sözcükler: Biotinidaz eksikliği, Yenidoğan Tarama, Fenilketonüri, Türkiye

INTRODUCTION

Inherited metabolic disorders (IMD), while rare, are more common in Türkiye where the rate of consanguineous marriages is higher than in other countries (1). In Türkiye, two IMDs, namely phenylketonuria (PKU) and biotinidase deficiency (BD), are screened in the National Newborn Screening Program (NNSP). The Phenylketonuria Screening Program started regionally in 1987 and was expanded to all of Türkiye in 1993. In 2008, BD was added to NNSP (2). While the newborn screening rate in Türkiye was 4.7% in 1987, it has reached almost a total rate of 95% since 2008 (3). It is estimated that the application of the NNSP has led to the prevention of disability in approximately 4,500 babies per year through early diagnosis in Türkiye (2).

Phenylketonuria is an autosomal recessive IMD associated with high phenylalanine levels, 98% of which are caused by PAH gene mutations and 2% by mutations in the BH4 metabolism. Phenylketonuria is frequently associated with developmental delay, progressive cognitive deterioration, neuropsychiatric findings, autism, dysmyelination, and light skin, hair and eye color. Early diagnosis and treatment can support the patient in developing in line with their peers (4, 5). Biotinidase deficiency is another autosomal recessively inherited neurocutaneous IMD that is associated with such symptoms as seizures, sensorineural hearing loss, skin rash, skin dryness, alopecia and acidosis. Since these findings can be confused with many different diagnoses in the neonatal period, an accurate diagnosis would be difficult in the absence of a screening program. Early diagnosis and rapid treatment can prevent the development of neurological sequelae and other clinical findings, and for this reason, BD and PKU are both included in the NNSP in Türkiye (6-8).

Phenylketonuria is most frequently seen in Türkiye, with a prevalence of 0.0167% in the world (9). In a regional study conducted in Türkiye, the highest and lowest incidences of screened diagnoses for the last 10 years were reported as 1:657–1:8375 for PKU and 1:1861–1:6815 for biotinidase deficiency (10). In a 10-year study conducted in another region of Türkiye, the incidence of PKU was reported as 1:7878, and the incidence of BD as 1:2359. A further study found that 4.7% of babies referred with suspected PKU had the diagnosis confirmed, while 18.9% had hyperphenylalaninemia (HPA), and 29.7% of those referred with suspected BD had the diagnosis confirmed (11). In the limited number of studies conducted for the single regions, final diagnoses were made based on biochemical examinations.

In our study conducted in a tertiary center in Ankara, the capital of Türkiye, babies referred by the NNSP with suspected PKU or BD were subjected to both biochemical and genetic examinations to investigate not only the diagnosis, but also the treatment status.

MATERIALS and METHODS

This study was approved by the Ankara University ethics committee (İ01-32-24, 18.01.2024).

The medical records of babies referred to Ankara University Faculty of Medicine, Department of Pediatric Metabolism between January 2019 and November 2023 were reviewed retrospectively, and the baseline characteristics, screening values, and biochemical and genetic examination results, treatment status of the patients were documented. This study was descriptive study.

In the NNSP, if the initial phenylalanine value is ≥4mg/dL, or ≥2.1 mg/dL twice for PKU, and if the biotinidase activity is ≤65 MRU (microplate response units) twice for BD, referral to department of metabolism is recommended (2).

The plasma blood amino acids of babies referred to our center with suspected PKU were studied and a PAH gene analysis was performed in the presence of abnormal phenylalanine values. Additionally, patients with high phenylalanine levels were assessed for BH4 (tetrahydrobiopterin) metabolism disorders. Phenylalanine levels of >20 mg/dL are classified as classical PKU, 15–20 mg/dL as moderate PKU, 10–15 mg/dL as mild PKU, 6–10 mg/dL as mild hyperphenylalaninemia and 2–6 mg/dL as benign mild HPA (12). If a phenylalanine level of >6 mg/dL is recorded in newborns, the urgent onset of treatment

is recommended (13). Before initiating BH4 treatment, we evaluated our patient's variants on the BioPKU website to predict BH4 responsiveness. We conducted a 48-hour BH4 loading test and monitored the responsiveness of phenylalanine levels. For the present study, these data were adopted as the classification and treatment indication limits.

Plasma biotinidase activity was measured and a BTD gene analysis was performed on babies referred to our center with suspected BD. At our center, the level of biotinidase enzyme is studied using the fluorometric method. In which <0.7 U/L (<10%) was considered a profound deficiency, 0.7–2.1 U/L (10-30%) a partial deficiency, 2.1–5.1 U/L (30-70%) a heterozygous deficiency and>5.1 U/L (>70%) normal activity (14). Biotin treatment was started in patients who have a profound deficiency and partial deficiency for biotinidase activity. For the present study, these data were accepted as the classification and treatment indication limits.

The complete gene sequence analysis was conducted for PAH and BTD genes, and in the event of identifying two different variants in a patient, confirmation was carried out on their parents using Sanger sequencing. Patients with variants detected in two different alleles were presented as compound heterozygotes.

Statistical Analysis

IBM SPSS Statistics (Version 28.0. Armonk, NY: IBM Corp.) was used for the statistical analysis, for which numbers, percentages, 25th-75th quarters, mean, standard deviation, median, minimum, and maximum values were calculated.

RESULTS

Between January 2019 and November 2023, 143 babies were referred to our department by the NNSP, of whom 59 were female (41.3%) and 84 (58.7%) were male. The median (25–75th percentile) age of the patients upon presentation to our center was 23.0 (17.0–30.0) days, while the median (25–75th percentile) gestational age was 38.3 (37.4–39.40) weeks. Of the total, 27 (18.9%) were preterm, 112 (78.3%) were term and four (2.8%) were post-term. The median (25–75th percentile) birth weight was 3080 (2770–3500) grams, and the weight of 90.9% of the sample was within the normal range. Of the total, 13.3% of the patients had a history of hospitalization, 23.1% had a history of icterus, 24.5% were from consanguineous marriages, and 3.5% had a sibling history with the same diagnosis. Finally, 78 (54.5%) of the patients were referred to our center with suspected BD and 65 (45.5%) with suspected PKU (Figure 1) (Table I) .

The median values of the first, second and third samples garnered from the NNSP records of the patients referred with suspected PKU were 2.3 mg/dL (2.0–3.4), 2.4 (2.2–3.4) and 2.4 (2.1–2.7), respectively. The median values of the first, second samples of patients referred with suspected BD were 54.4 MRU [46.7–60.8] and 57.3 [46.1–62.0], respectively. Only one patient in the sample was examined for the third time for BD, with a recorded value of 52.8 MRU (Table II).

Table I: Baseline characteristics of the research group

Sex*	
Female	59 (41.3)
Male	84 (58.7)
Age at hospital admission, days	
mean±SD	26.43±14.8
median (min-max)	23.0 (4.0-89.0)
25–75th percentile	17.0-30.0
Gestational age group*	
Preterm	27 (18.9)
Term	112 (78.3)
Post-term	4 (2.8)
Gestational age, weeks	
mean±SD	38.28±1.78
median (min-max)	38.3 (32.0-42.3)
25–75th percentile	37.4-39.40
Birth weight group*	
<2500 grams	9 (6.3)
2500–4000 grams	130 (90.9)
>4000 grams	4 (2.8)
Birth weight, grams	
mean±SD	3096±575
median (min-max)	3080 (2770-3500)
25–75th percentile	2770-3500
Hospitalization history*	
Yes*	19 (13.3)
No	124 (86.7)
Jaundice history*	
Yes	33 (23.1)
No	110 (76.9)
Consanguineous marriage*	
Yes*	35 (24.5)
No	108 (75.5)
Sibling with the same diagnosis*	
Yes*	5 (3.5)
No	138 (96.5)
Disease suspicion*	
Biotinidase deficiency	78 (54.5)
Phenylketonuria	65 (45.5)

*: *n(%)*

The examinations performed at our center revealed median phenylalanine values in the first sample in the serum of patients referred with suspected PKU of 1.9 mg/dL (1.2–3.3). Biotinidase activity in the serum of patients referred with suspected BD were compatible with a partial deficiency in 39.7% and a profound deficiency in 8% in the first sample, while in the second sample, 25.6% were consistent with partial deficiency and 3.8% with profound deficiency (Table II).

A PAH gene analysis was performed on 23 (35.4%) babies who were found to have high phenylalanine levels. Of the patients subjected to a PAH gene analysis, a compound heterozygous variant was detected in 14 (60.9%) and a homozygous variant in six (26.0%). Of the six patients with the homozygous

Table II: National Newborn Screening Program Results and Serum Measurements in Our Center

	1st sample		2nd sample		3rd sample	
	A*	B†	A*	B†	A*	B†
PKU suspected babies, Phe level, mg/dL						
Sample number	63	65	55	40	18	23
mean±SD	3.2±3.0	3.7±6.6	2.8±3.0	3.7±4.4	2.5±0.6	4.0±3.0
median (min-max)	2.3 (0.8–17.0)	1.9 (0.65–46.0)	2.4 (2.0–6.1)	1.9 (0.5–22.0)	2.4 (1.8–4.8)	3.5 (0.9–12.4)
25–75th percentile	2.0–3.4	1.2–3.3	2.2–3.4	1.2–4.4	2.1–2.7	1.6–5.4
BD suspected babies, biotinidase activity, MRU						
The number of the sample	76	78	68	59	1	38
mean±SD	49.5±16.5	2.01±0.9	53.7±19.2	2.3±1.0	52.8	3.0±1.6
median (min-max)	54.4 (0.0–65)	2.0 (0.1–5.0)	57.3 (0.0–122)	2.2 (0.1–5.6)	-	2.8 (0.4–9.7)
25–75th percentile	46.7–60.8	1.3–2.6	46.1–62.0	1.8–2.8	-	2.0–3.7
The group of biotinidase activity, n (%)						
Profound deficiency (<0.7)		8 (10.3)		3 (3.8)		1 (1.3)
Partial deficiency (0.7–2.1)	-	31 (39.7)		20 (25.6)		11 (14.1)
Heterozygous deficiency (2.1–5.1)		38 (48.7)		33 (42.3)		22 (28.2)
Normal (>5.1)		1 (1.3)	-	1 (1.3)	-	3 (3.8)

***A:** National Newborn Screening Program Results, †**B:** Serum Measurements in Our Center, **BD:** Biotinidase deficiency, **Phe:** Phenylalanine, **PKU:** Phenylketonuria*

Table III: PAH and BTD gene analysis

	Total analysis	Homozygous variant	Compound heterozygous variant	Heterozygous variant	No variant
In the group of babies referred with suspected PKU PAH gene analysis)	23 (35.4)	6 (26.0)	14 (60.9)	2 (8.7)	1 (4.4)
In the group of babies referred with suspected BD BTD gene analysis*	62 (79.5)	21 (33.9)	10 (16.1)	27 (43.5)	4 (6.5)

***:** n (%), **BD:** Biotinidase deficiency, **PKU:** Phenylketonuria*

Table IV: Treatment approach to patients with suspected PKU and BD

	Suspected PKU, treatment*	Suspected BD, treatment*
Total number	9 (13.8)	46 (59.0)
Diet	3 (4.6)	-
Saptopterin	1(1.5)	-
Diet + saptopterin	5 (7.7)	-
No treatment	56 (86.2)	1 (1.3)
Biotin	-	46 (59.0)
Biotin treatment discontinued during follow-up	-	31 (39.7)

***:** n(%), **BD:** Biotinidase deficiency, **PKU:** Phenylketonuria*

variant in the PAH gene, treatment had already been started in five as a phenylalanine level of >6 mg/dL was recorded at admission. While three patients with the homozygous variant were receiving combined saptopterin and diet treatment, two were being monitored with diet therapy alone. Although the variant of a male patient with a homozygous variant in the PAH gene [PAH: NM_000277.3 c.533A>G (p.Glu178Gly)] is pathogenic, treatment has not been started as phenylalanine levels of 1.8–2.1 mg/dL have been recorded, but follow-up continues. Of the 14 patients with a compound heterozygous variant in the PAH gene, 10 were found to be compatible with benign mild HPA (phenylalanine levels of between 2–6 mg/dL) and were being monitored without treatment. Of the four patients with the compound heterozygous variant, two were continued with a saptopterin and diet combined treatment, one with diet and one with saptopterin (Tables III and IV). A total of nine cases of PKU and 21 cases of HPA were diagnosed. All patients were evaluated for BH4 metabolism disorders, and no abnormal results were found. One patient with no variant in the PAH gene was diagnosed with citrin deficiency based on the identification of high levels of citrulline and arginine in a blood amino acid analysis revealing simultaneous cholestasis and hyperammonemia findings. The patient's diagnosis was confirmed with the detection of a pathogenic homozygous variant in the SLC25A13 gene, for which the patient was started on a combined diet and pharmacological treatment based on a diagnosis of neonatal intrahepatic cholestasis caused by citrin deficiency (NICCD) type citrin deficiency.

A BTD gene analysis was performed on 62 (79.5%) of the patients referred with suspected BD, revealing a heterozygous variant in 27 (43.5%), a homozygous variant in 21 (33.9%) and a compound heterozygous variant in 10 (16.1%), while no BTD variant was identified in four (5.1%). All patients were started on biotin except one baby with a normal initial biotinidase level. The biotin treatment was subsequently discontinued in 31 babies (39.7%) identified with no biallelic variant after a BTD

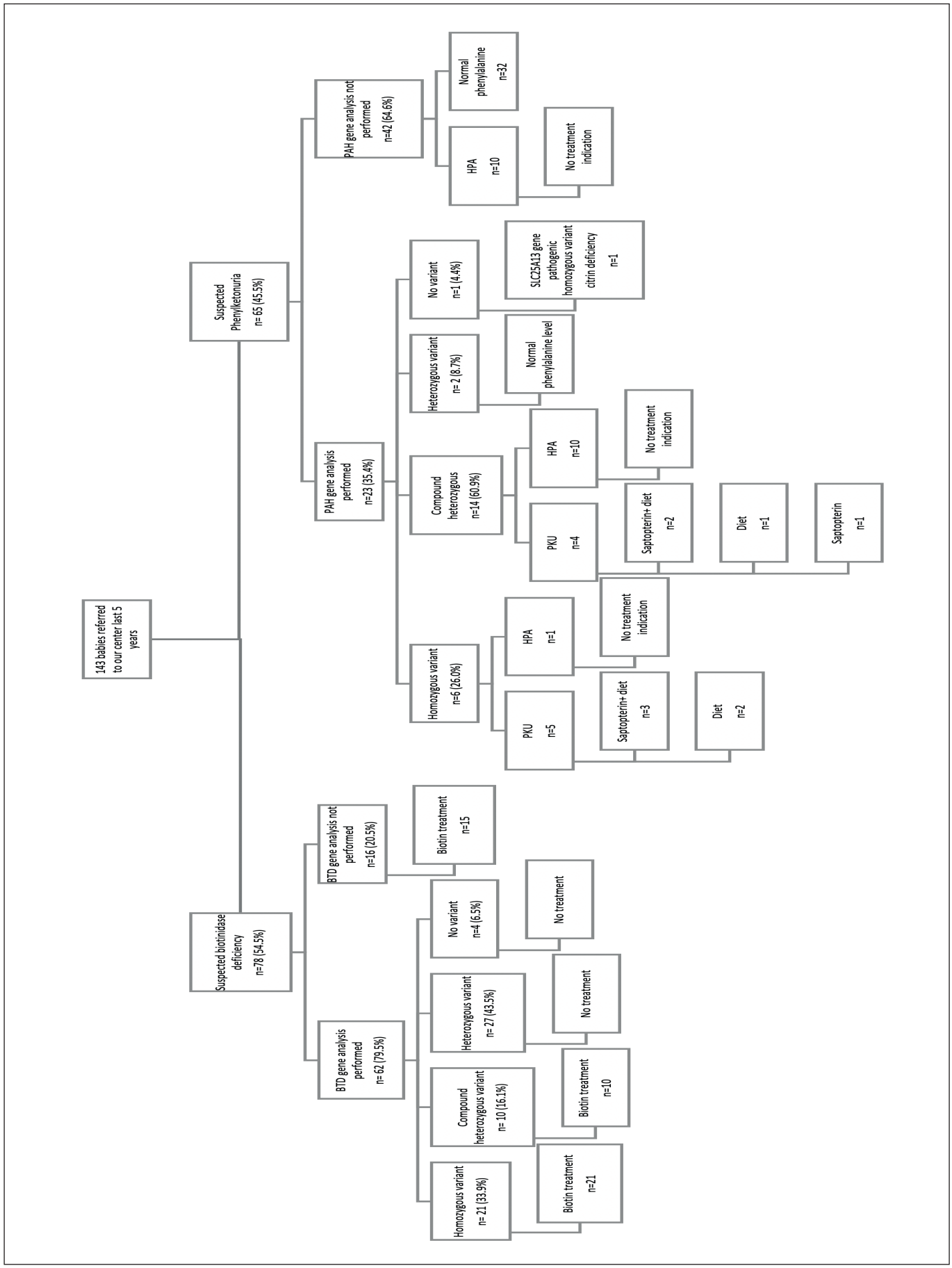

Figure 1: The flowchart of the study

gene analysis, and whose follow-up biotinidase activities were compatible with the carrier level. Finally, the biotin therapy of 46 patients (59.0%) was continued (Tables III and IV).

DISCUSSION

In this study, carried out in a tertiary center in Ankara, the capital of Türkiye, approximately one-third of the babies referred to our center by the NNSP over the last five years were found to be carrying biallelic variants of the relevant disease. Treatment indication was determined for 13.8% of the babies referred with suspected PKU and 59% of those with suspected BD.

A rate of consanguineous marriage in Türkiye of 23.2% has been reported, although rates of 42.6% have been reported in the Southeastern Anatolia region (15). The rate of consanguineous marriage in the present study was found to be 24.5%, which is similar to the general country rate. Türkiye is a country where a higher incidence of PKU and BD can be expected than in the rest of the world, and there are few studies in the field of NNSP (9-11). The present study is the first to include genetic examination and treatment processes, in addition to biochemical evaluations of babies with suspected PKU and BD.

Assuming a global prevalence of 1:23930, there are an estimated 0.45 million cases of PKU around the world, and prevalence studies have been conducted in many countries to date reporting different figures, including Italy (1:4000), Ireland (1:4545), Denmark (1:13434), Finland (1:112000), Thailand (1:227273), Japan (1:125000) and the Philippines. (1:116006) (4). Studies for PKU conducted in our country have produced different rates of 1:657, 1:8375, 1:6667 and 1:1861, reported regionally and yearly (4, 9-11). It is estimated at 1:60000 live births in BD (16). A study conducted in Italy reported a prevalence of BD of 1:61000, while as 1:28316 in Saudi Arabia, which is the rate of consanguineous marriage was reported similar to our country (17,18). Regional studies in our country have reported a prevalence of BD of 1:1861, 1:2359 and 1:6815, depending on the year and region (10,11). In the present study, the general research population was babies referred to our center with suspected PKU or BD, however this cannot be considered a prevalence study, as there is a lack of data on the number of positive cases among the screened patients, and what percentage of them applied to our center. Since our research population is a center in the capital that accepts patients from all over the country, accessing accurate data is also challenging, and this can be considered a limitation of our research.

A study conducted in a single region in our country reported that 4.7% of the babies referred with suspected PKU had the diagnosis confirmed, while 18.9% were diagnosed with hyperphenylalaninemia, and 29.7% of the babies referred with suspected biotinidase deficiency were diagnosed with BD (11). In the present study, 13.8% of the babies referred with suspected PKU had diagnosis confirmed, while 32.3% were diagnosed with hyperphenylalaninemia. A BTD gene analysis was performed on 79.5% of the babies referred with suspected BD in the present study, and biallelic variants were detected in half of them. The higher rates of prevalence reported in the present study indicate the potential for regional variations, revealing a need to garner data from each region in the country to identify the prevalence and distribution of positive cases geographically. The creation of a national registry would provide a clear understanding of the regions in Türkiye where pediatric metabolism specialists are needed.

Zeybek et al. (19) reported a case of citrin deficiency detected incidentally in NNSP upon the identification of a compound heterozygous variant in the SLC25A13 gene in whom cholestasis, coagulopathy, hyperammonemia and high citrulline were detected during follow-up for suspected PKU. In the present study, a female baby was examined for the development of cholestasis while being investigated for suspected PKU and was diagnosed with citrin deficiency due to the very similar clinical findings and the detection of a homozygous pathogenic variant in the SLC25A13 gene. Our study also revealed the second case in Türkiye to be referred by NNSP with suspected PKU and diagnosed with citrin deficiency (19). Also, sister of the patient was examined due to the history of cholestasis in infancy, and she was also diagnosed with citrin deficiency. Two siblings were started on a high-protein, low-carbohydrate diet and followed up. Ünal et al. (20) reported diagnoses of Maple Syrup Urine Disease (MSUD) in four patients, galactosemia in two patients and tyrosinemia type 1 in one patient referred from NNSP with high phenylalanine levels who underwent a detailed examination. It has been suggested that any condition that causes liver dysfunction can increase plasma phenylalanine levels (20). In the light of the above cases diagnosed with IMD other than PKU, we recommend that measurements of phenylalanine levels should not be carried out with limited parameters in patients coming from NNSP with suspected PKU, all plasma amino acids should be analyzed, and patients should be examined in detail and evaluated holistically.

Since the incidence of autosomal recessive diseases is higher in regions where consanguineous marriages are common, patients need to be diagnosed and started on treatments early in the asymptomatic period. The launch of an expanded newborn screening program in our country would contribute to a decrease in the death and sequelae rates of many patients, as only two IMDs (PKU and BD) are currently being screened for today.

Newborns with phenylalanine levels >6 mg/dL should be started on treatment as soon as possible (13). In the present study, this cut-off value was adopted as the treatment indication, and led to the start of treatment in nine (13.8%) patients. Since patients with BD can have neurocutaneous consequences if left

untreated, prompt treatment decisions are essential (7, 21, 22). Biotin is a safe and non-toxic drug (23). Biotin was started in all of the patients in the present study whose biotinidase levels were lower than normal. The importance of repeated measurements of biotinidase activity and genetic analysis for a final diagnosis has been demonstrated (14). Biotin treatment was started and discontinued only after excluding a BD diagnosis in babies with no biallelic variant in their genetic analysis, and whose repeated measurements of biotinidase activity were within the normal range during follow-up.

This is one of the few studies of the NNSP to date in our country and has provided genetic analysis and treatment follow-up data related to babies. Multi-center, large-scale studies would provide a clearer understanding of the current situation in the country.

REFERENCES

1. Kaplan S, Pinar G, Kaplan B, Aslantekin F, Karabulut E, Ayar B, et al. The Prevalence of Consanguineous Marriages and Affecting Factors in Turkey: A National Survey. J Biosoc Sci 2016;48:616-30.
2. https://hsgm.saglik.gov.tr/tr/tarama-programlari/ntp.html. Access Date: January 20, 2024
3. Tezel B, Dilli D, Bolat H, Sahman H, Ozbas S, Acican D, et al. The development and organization of newborn screening programs in Turkey. J Clin Lab Anal. 2014;28:63-9.
4. Hillert A, Anikster Y, Belanger-Quintana A, Burlina A, Burton BK, Carducci C, et al. The Genetic Landscape and Epidemiology of Phenylketonuria. Am J Hum Genet. 2020;107:234-50.
5. Rajabi F, Rohr F, Wessel A, Martell L, Dobrowolski SF, Guldberg P, et al. Phenylalanine hydroxylase genotype-phenotype associations in the United States: A single center study. Mol Genet Metab 2019;128:415-21.
6. Karaca M, Ozgul RK, Unal O, Yucel-Yilmaz D, Kilic M, Hismi B, et al. Detection of biotinidase gene mutations in Turkish patients ascertained by newborn and family screening. Eur J Pediatr 2015;174:1077-84.
7. Canda E, Kalkan Ucar S, Coker M. Biotinidase Deficiency: Prevalence, Impact And Management Strategies. Pediatric Health Med Ther 2020;11:127-33.
8. Funghini S, Tonin R, Malvagia S, Caciotti A, Donati MA, Morrone A, et al. High frequency of biotinidase deficiency in Italian population identified by newborn screening. Mol Genet Metab Rep 2020;25:100689.
9. El-Metwally A, Yousef Al-Ahaidib L, Ayman Sunqurah A, Al-Surimi K, Househ M, Alshehri A, et al. The Prevalence of Phenylketonuria in Arab Countries, Turkey, and Iran: A Systematic Review. Biomed Res Int 2018;2018:7697210.
10. Akova İ, Kılıç E, Koşaroğlu NE. Evaluation of the results of the 10-year screening program for neonatal metabolic and endocrine diseases: The case of Sivas province, Türkiye. Turk J Public Health 2022; 20:410-22.
11. Toktas I, Saribas S, Canpolat S, Erdem O, Ozbek MN. Evaluation of patients diagnosed with phenylketonuria and biotinidase deficiency by the newborn screening program: a ten-year retrospective study. Turk J Pediatr 2022;64:985-92.
12. Camp KM, Parisi MA, Acosta PB, Berry GT, Bilder DA, Blau N, et al. Phenylketonuria Scientific Review Conference: state of the science and future research needs. Mol Genet Metab 2014;112:87-122.
13. Vockley J, Andersson HC, Antshel KM, Braverman NE, Burton BK, Frazier DM, et al. Phenylalanine hydroxylase deficiency: diagnosis and management guideline. Genet Med 2014;16:188-200.
14. Surucu Kara I, Kose E, Koc Yekeduz M, Eminoglu FT. A different approach to the evaluation of the genotype-phenotype relationship in biotinidase deficiency: repeated measurement of biotinidase enzyme activity. J Pediatr Endocrinol Metab 2023;36:1061-71.
15. Tezel B, Aydın Ş. Infant Mortality Situation Report in Turkey on the 100th Anniversary of the Establishment of the Ministry of Health. Ankara: Turkish Republic of Ministry of Health, General Directorate of Public Health, 2021.
16. Wolf B. Clinical issues and frequent questions about biotinidase deficiency. Mol Genet Metab 2010;100:6-13.
17. Porta F, Pagliardini V, Celestino I, Pavanello E, Pagliardini S, Guardamagna O, et al. Neonatal screening for biotinidase deficiency: A 30-year single center experience. Mol Genet Metab Rep 2017;13:80-2.
18. Mohamed S, Elsheikh W, Al-Aqeel AI, Alhashem AM, Alodaib A, Alahaideb L, et al. Incidence of newborn screening disorders among 56632 infants in Central Saudi Arabia. A 6-year study. Saudi Med J 2020;41:703-8.
19. Zeybek AC, Kiykim E, Zubarioglu T, Cansever MS, Ceylaner S, Erkan T. Citrin Deficiency: An Infant Incidentally Detected by Phenylketonuria Screening with a Novel Mutation in Slc25a13 Gene. Genet Couns 2015;26:409-13.
20. Unal O, Ozturk-Hismi B, Coskun T, Tokatli A, Dursun A, Sivri HS. Detection of other inborn errors of metabolism in hyperphenylalaninemic babies picked up on narrow-spectrum screening programs. Turk J Pediatr 2012;54:409-12.
21. Sondhi V, Sharma S. Vitamin-Responsive Movement Disorders in Children. Ann Indian Acad Neurol 2020;23:325-31.
22. Ferreira P, Chan A, Wolf B. Irreversibility of Symptoms with Biotin Therapy in an Adult with Profound Biotinidase Deficiency. JIMD Rep 2017;36:117-20.
23. Wolf B. High doses of biotin can interfere with immunoassays that use biotin-strept(avidin) technologies: Implications for individuals with biotin-responsive inherited metabolic disorders. Mol Genet Metab 2019;127:321-4.