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ORIGINAL ARTICLE
ORİJİNAL ARAŞTIRMA

An Etiological Evaluation of Children with Acute Recurrent Pancreatitis: Based on Genetic Analysis and Pancreaticobiliary Maljunction without Biliary Dilatation

Akut Rekürren Pankreatitli Çocukların Etiyolojik Değerlendirmesi: Genetik Analiz ve Biliyer Dilatasyonsuz Pankreatikobiliyer Bileşke Anomalisi Temelinde

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ABSTRACT

Aim: The most common risk factors in acute recurrent pancreatitis (ARP) are genetic and anatomical/obstructive causes. Some cases of ARP have been considered as 'idiopathic' without detailed genetic analysis. The aim of this study is to evaluate the genetic risk factors and the presence of long common channel in cases evaluated as idiopathic ARP.

Material and Method: In this study, 19 patients who were evaluated as idiopathic ARP after primary care evaluation, between January 2017 and January 2022, were included. The CFTR, PRSS1, SPINK1 and CTRC genes were analyzed in these patients. The length of the pancreaticobiliary common channel was measured and compared with normal values for age by the evaluation of MRCP imaging.

Results: The mean age was 11.21 ±3.70 years and 57.9% (n=11) of the patients were female. In 52.6% (n=10) of the cases, the pancreaticobiliary channel length was above the normal values determined for age. Mutations in the CFTR (7/19), PRSS1 (2/19) and CTRC (1/19) genes were detected in 52.6% (n=10) of the patients.

Conclusions: The results of our study showed the presence of genetic risk factors (CFTR, PRSS1 and CTRC) and PBM without biliary dilatation in approximately half of the cases named as idiopathic ARP. In the presence of long common channel without biliary dilatation, diagnostic delay due to the subtle radiological findings may cause gallbladder cancer in adulthood. Therefore, children with ARP should also be evaluated for PBM without biliary dilatation before being labeled as 'idiopathic'.

Keywords: Acute recurrent pancreatitis, CFTR, PRSS1, CTRC, pancreaticobiliary maljunction, long common channel

ÖZ

Amaç: Akut rekürren pankreatitte (ARP) en sık görülen risk faktörleri genetik ve anatomik/obstrüktif nedenlerdir. Bazı ARP vakaları, ayrıntılı genetik analiz yapılmadan 'idiyopatik' olarak kabul edilmektedir. Bu çalışmanın amacı idiyopatik ARP olarak değerlendirilen olgularda genetik risk faktörlerini ve uzun pankreatikobiliyer ortak kanal varlığını değerlendirmektir.

Gereç ve Yöntem: Bu çalışmaya, Ocak 2017-Ocak 2022 tarihleri arasında birinci basamak değerlendirme sonucu idiyopatik ARP olarak değerlendirilen 19 hasta dahil edildi. Bu hastalarda CFTR, PRSS1, SPINK1 ve CTRC genleri analiz edildi. MRCP ile pankreatikobiliyer ortak kanal uzunluğu ölçülerek yaşa göre normal değerlerle karşılaştırıldı.

Bulgular: Hastaların yaş ortalaması 11.21 ±3.70 yıldı ve %57.9 (n=11)'u kızdı. Olguların %52.6 (n=10)'sında pankreatikobiliyer ortak kanal uzunluğu, yaşa göre belirlenmiş normallerin üzerindeydi. Hastaların %52.6 (n=10)'sında CFTR (7/19), PRSS1 (2/19) ve CTRC (1/19) genlerinde mutasyonlar tespit edildi.

Sonuç: Çalışmamız idiyopatik ARP olarak adlandırılmış olguların yaklaşık yarısında genetik risk faktörlerinin (CFTR, PRSS1 ve CTRC) ve biliyer dilatasyonsuz pankreatikobiliyer bileşke anomalisinin varlığını göstermiştir. Biliyer dilatasyonsuz bileşke anomalisi varlığında, radyolojik bulguların belirgin olmamasına bağlı tanısal gecikme, erişkin yaşta safra kesesi kanserine neden olabilir. Bu nedenle ARP'li çocuklar 'idiyopatik' olarak etiketlenmeden önce biliyer dilatasyonsuz bileşke anomalisi açısından da değerlendirilmelidir.

Anahtar Kelimeler: Akut rekürren pankreatit, CFTR, PRSS1, CTRC, pankreatikobiliyer bileşke anomalisi, uzun ortak kanal

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INTRODUCTION

Acute recurrent pancreatitis (ARP), defined as 2 or more distinct episodes of pancreatitis, is a relatively rare condition. Most of the data on ARP are obtained from the adult literature (1). Thanks to increasing definition of ARP, the International Study Group for Pediatric Pancreatitis: In Search has focused on the identification, diagnosis and management of risk factors for these cases (2). Risk factors in children are different from adults. Currently, the most common risk factors defined in children include genetic and anatomical factors (3). However, when the INSPPIRE database was evaluated, it was found that many cases were defined as idiopathic ARP, without genetic testing (2).

A normal pancreaticobiliary junction is formed by the joining of the main pancreatic duct and the common bile duct and opening into the duodenum by forming a common channel. The localization of this junction is the submucosal layer of the duodenum. This intramural localization of the common channel provides it to be controlled by the intramurally located sphincter of Oddi. In the case of pancreaticobiliary maljunction (PBM), the bile and pancreatic ducts join outside the duodenal wall, forming a long common channel that is not under the control of the Oddi sphincter (4). In adults, a common channel length greater than 15 mm is defined as PBM (5). However, in children, the maximal length of the common channel varies according to the age. The maximal length of common channel is 3 mm in infants under 1 year of age and increases up to 5 mm until 15 years of age (6).

Pancreaticobiliary maljunction may be with or without biliary dilatation, depending on the diameter of the common bile duct (7). Pancreaticobiliary maljunction without biliary dilatation is seen less frequently than PBM with dilatation. In PBM without biliary dilatation, acute pancreatitis is seen more frequently in pediatric patients compared to adults (8). In PBM with biliary dilatation imaging findings are more definite compared to PBM without biliary dilatation. In cases with PBM without biliary dilatation, diagnosis may be delayed due to indefinite radiological findings. In some cases, the diagnosis can be possible with the detection of biliary cancer in adulthood (9).

The presence of indefinite radiological findings and the frequency of acute pancreatitis in cases with PBM without biliary dilatation suggest that some of the cases defined as idiopathic acute recurrent pancreatitis may be PBM without biliary dilatation. The aim of this study is to investigate genetic risk factors and the presence of PBM without biliary dilatation in children, who had been diagnosed with idiopathic ARP, in the primary care evaluation.

MATERIAL AND METHOD

Study Population

This cross-sectional study was carried out at Necmettin Erbakan University Meram Medical Faculty, Pediatric Gastroenterology clinic between January 2017 and January 2022, prospectively. Patients aged 0-18 years who were diagnosed with ARP and no proven cause (toxic, autoimmune, metabolic, and obstructive) could be found in the primary care evaluation were included in the study.

The diagnosis of acute recurrent pancreatitis was made according to the recommendations of the INSPPIRE (International Study Group of Pediatric Pancreatitis: In Search for a Cure) consortium (10). According to INSPPIRE recommendations, the main criterion was 2 or more episodes of acute pancreatitis. The other criteria include the absence of pain for at least 1 month after the first attack or both the absence of pain regardless of the duration and the return of pancreatic enzyme levels to normalcy (10). The number of patients diagnosed with ARP according to these criteria was 33. The study was conducted on 19 patients who were defined as idiopathic ARP, after excluding 14 patients whose etiology was determined in the first step evaluation (5 patients with gallstones, 4 patients with PBM with biliary dilatation, 2 patients with autoimmune pancreatitis, 1 patient with pancreatic divisum, 1 patient with valproic acid use, 1 patient with 3-Hydroxy-3-Methylglutaryl-CoA Lyase deficiency). In the first step evaluation; Anamnesis (drug use, signs of infection, family history of pancreatic disease, consanguineous history), laboratory evaluation (transaminase, gamma glutamyl transferase (GGT), total/direct bilirubin, calcium, fasting lipids) and imaging methods were performed. Ultrasonography was the primary imaging method and when no anatomical/obstructive cause was detected in ultrasonography, cases with increased GGT were evaluated with Magnetic Resonance Cholangiopancreatography (MRCP).

Two of the patients had been diagnosed with cystic fibrosis (CF) before the ARP clinic developed and CFTR gene analysis had already been performed. The remaining 17 patients underwent gene analysis for Serine Protease 1 (PRSS1), CF Transmembrane Conductance Regulator (CFTR), Serine Peptidase Inhibitor Kazal type 1 (SPINK1), and Chymotrypsin C (CTRC). Common bile duct diameter and pancreaticobiliary common channel length were measured by MRCP in all cases.

Genetic Analysis

The next-generation sequencing analysis of the SPINK1, PRSS1, CTRC, and CFTR genes were performed using the MiSeq NGS platform (Illumina, San Diego, CA, USA). The extraction of genomic DNA was performed following the manufacturer's standard procedure and using the QIAamp DNA Blood Mini Kit (Qiagen, Hilden, Germany). The exome sequencing necessary for the synthesis of the primers to be used in the study was taken from the www.ensembl.

org genetic data banks. To design the primers from those sequences, the www.ncbi.nlm.nih.gov/tools/primer-blast/ database was used. Gene pools were prepared for all patients and each pool was measured at Qubit according to the standard Qubit protocol. The library was prepared in accordance with the Nextera XT DNA Library Prep Kit (Illumina Inc.) procedure and by following the manufacturer's instructions. The library preparation phase included the genomic DNA fragmentation, labeled DNA amplification, DNA purification, and denaturation that were performed respectively. The "Integrative Genomics Viewer (IGV)" program was used to evaluated the variants of interested genes in this study. Sanger Sequencing was conducted along with 3500DX Genetic Analyzer (Applied Biosystems, United States) as a verification method.

In this study, we analyzed the coding regions and splice sites of four genes in a group of ARP patients. Since we aimed to be conservative in our estimate of the major genetic cause of ARP, we excluded synonymous, intronic, and 5'- or 3'-untranslated region variants in the four genes from consideration except where there was persuasive evidence of a functional consequence. The pathogenicity classification of variants was evaluated in accordance with ACMG criteria (11).

Evaluation of Biliary Dilatation and Common Channel Length

MRCP imaging was performed with 1.5 Tesla Siemens (Allegra, Germany) by using body coil. First three planes True FISP imaging was taken for localizing the abdominal structures. Axial images must include the liver from the anterior wall to the posterior wall of the abdomen. T2 HASTE images with fat saturation were performed first in axial plane. Then coronal oblique imaging was performed for whole liver from diaphragm to the third and fourth segment of the duodenum to include all biliary system. For oblique coronal imaging the block was positioned across the common bile duct. These images were taken while the patients hold their breath. The diameter of common bile duct was measured for assessment of congenital biliary dilatation. Biliary dilatation was defined using reference values for age in the diagnostic criteria for congenital biliary dilatation defined by the Japanese Study Group on Pancreaticobiliary Maljunction (JSGPM) in 2015 (12).The length of the common channel was measured from the coronal oblique T2 HASTE images. Measurements were analyzed using reference values for age that previously reported by Guelrud et al and subsequently used in several studies (6, 13, 14).

Statistical analysis

Statistical analyzes of the study were performed by using SPSS 20.0 (IBM Inc, Chicago, IL, USA). The conformity of the variables to the normal distribution was evaluated by the Shapiro Wilk test. Descriptive statistics were presented as mean ±SD and median (Q1-Q3) for numerical variables and frequency (percentage ratio) for categorical variables. Comparisons of attack number and attack age according to Common Channel scores were performed by Mann-Whitney U test. A value of $p<0.05$ was considered statistically significant for the type-I error rate of 5% in the analyses.

RESULTS

A total of 19 pancreatitis patients were included in the study. The mean age was 11.21 ±3.70 years and 57.9% (n=11) of the patients were female and 42.1% were males (n=8). The mean weight z score was -0.17 ±1.36 and the mean height z score was -0.43 ±1.06. The median BMI z score was -0.40 (-0.88-1.33). The mean follow-up period was 29.79 ±15.10 months, the age at first attack was 8.37 ±3.77 years, and the number of attacks was 5.16 ±3.07 (**Table 1**). In 31.6% (n=6) of the patients a family history of pancreatitis was present and in 31.6% (n=6) of the patients a family history of cystic fibrosis was present.

Table 1. The demographic characteristics, clinical and radiological findings of patients with acute recurrent pancreatitis

	N (%) / Mean ±SD / Median (Q1-Q3)
Gender	
Female	11 (57.9)
Male	8 (42.1)
Age (year)	11.21 ±3.70
Weight z score	-0.17 ±1.36
Height z score	-0.43 ±1.06
BMI z score	-0.40 (-0.88-1.33)
Follow-up time (month)	29.79 ±15.10
Age at first pancreatitis attack (year)	8.37 ±3.77
Number of pancreatitis attacks	5.16 ±3.07
Common bile duct diameter (mm)	2.47 ±0.64
Length of the common channel (mm)	4.94±0.63
Long common channel	
Yes	10 (52.6)
No	9 (47.4)

Radiologic Measurements

The mean common bile duct diameter measured on MRCP was found to be 2.47 ±0.64 mm and it was within the normal range for age in all cases. Common channel length was found to be 4.94 ±0.63mm. Pancreaticobiliary channel length was above the normal values determined for age in 52.6% (n=10) of the cases (**Table 2**). The mean age at first attack (6.70 ±3.88 years) in patients with long common channel was smaller than those with normal common channel length (10.22 ±2.77 years); however, the difference was not statistically significant (p=0.053). The mean number of attacks was 5.40 ±3.98 in cases with long common channel and 4.89 ±1.83 in those with normal common channel length (p=0.72).

Table 2. The radiologic findings and detected mutations in patients with acute recurrent pancreatitis

Case	CFTR	PRSS1	CTRC	Sweat test (mEq/L)	Diameter of CBD (mm)	Maximal diameter of CBD for age (mm)	Length of PBC (mm)	Maximal length of PBC for age (mm)
1	-/-	K170E*/S181N†	-/-	27	3.5	4.5	5.3	4.4
2	-/-	S181N†/-	-/-	22	2	5.0	5.0	5.0
3	G85E/ L568F	-/-	-/-	42.8	2.8	4.5	5.7	4.4
4	G85E/ L568F	-/-	-/-	47	2.5	4.8	5.0	5.0
5	F508del/-	-/-	-/-	61	3.6	5.0	5.7	5.0
6	-/-	K170E*/S181N†	-/-	28	2.1	4.8	4.8	5.0
7	-/-	-/-	-/-	24	2	3.7	4.6	3.6
8	L233V‡/-	-/-	-/-	25.7	1.9	3.7	4.1	3.6
9	-/-	R122C/-	-/-	26.5	2	5.3	5.0	5.0
10	-/-	-/-	-/-	26.7	2.4	4.3	3,5	4.1
11	-/-	-/-	-/-	21	2.1	4.9	4.2	5.0
12	D1152H/ D1152H I1051V/I1051V	-/-	-/-	89	2.6	4.3	5.5	4.1
13	F508del / R334W	-/-	-/-	92.1	2.1	5.2	5.0	5.0
14	-/-	Q98PfsTer10/-	-/-	37.6	1.8	3.9	5.4	4.1
15	-/-	-/-	-/-	32	2.8	4.0	4.5	4.1
16	F334W/ N1303K	-/-	-/-	135	1.5	5.1	4.3	5.0
17	G1069R/-	-/-	-/-	48.6	3.2	5.0	6,0	5.0
18	-/-	-/-	-/-	29.4	3.7	4.9	5.0	5.0
19	E217G†/-	-/-	R254W/-	22.2	2.3	4.7	5.4	5.0

*Likely benign †Benign ‡Variant of uncertain significance

Mutation Data Analysis and Classification

The mutations in CFTR, PRSS1 and CTRC genes were detected in 52.63% (n=10) of patients with ARP. Heterozygous variants contributed to the development of ARP were detected in the PRSS1 gene in 10.52% (n=2) of patients, and in the CTRC gene in 5.26% (n=1) of patients. CFTR genotypes contributed to the development of ARP in 36.84 % of cases, heterozygous, compound heterozygous and homozygous variants in the CFTR gene were identified in 7 of the 19 ARP patients.

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Detected CFTR Variants

CFTR genotypes contributed to the development of ARP in 36.84% of cases. The heterozygous, compound heterozygous and homozygous variants in the CFTR gene were identified in 7 of the 19 ARP patients.

In two patients who were siblings (Case 3 and 4), p.G85E/p.L568F variants were detected in the compound heterozygous state. These patients had normal pulmonary functions and equivocal sweat tests (42.8 and 50 mEq/L). p.Gly85Glu variant (NM_000492.4; c.254G>A; chr7:117149177) causes the substitution of a Glycine into an Glutamine residue at position 85 in exon 3. This variant position has a submission in ClinVar (ID: 7143), previously been reported in publications associated with CFTR gene (15). The detected variant is classified as "Pathogenic" (PP5, PM1, PM5, PP3, PM2). p.Leu568Phe variant (NM_000492.4; c.1704G>T; chr7:117230431) causes the substitution of a Leucine into an Phenylalanine residue at position 568 in exon 13. This variant has been reported in publications associated with CFTR gene (16). The detected variant is classified as "Likely Pathogenic" (PM1, PP3, PM2).

In the Case 5, a previously described in frame p.Phe508del variant was detected (NM_000492.4; c.1521_1523del; chr7:117199645) in the heterozygous state. This patient had normal pulmonary functions and abnormal sweat tests (61 mEq/L). The detected variant causes the substitution of a Phenylalanine into a del residue at position 508 in exon 11. This variant has submissions in ClinVar (ID: 634837), previously been reported in publications (17, 18). The detected variant is classified as "Pathogenic" (PS3, PM1, PP5, PM4, PM2).

In the case 12, p.Asp1152His/p.Asp1152His and p.Ile1051Val/p.Ile1051Val variants in the homozygous state had detected already. This patient had been previously diagnosed with cystic fibrosis and the sweat test were 89mEq/L. p.Asp1152His is a missense

variant (NM_000492.4; c.3454G>C; p.Asp1152His; chr7:117254753) causes the substitution of a Aspartic acid into an Histidine residue at position 152 in exon 21. This variant has a submission in ClinVar (ID: 35867), previously been reported in publications (19) and classified as "Pathogenic" (PM2, PP3, PP2, PP5). p.Ile1051Val is a missense variant (NM_000492.4; c.3151A>G; p.Ile1051Val; chr7:117251646) causes the substitution of a Isoleucine into an Valine residue at position 1051 in exon 20. This variant has been reported in publication (20) and classified as "Likely Pathogenic" (PM2, PM1, PP2, PP3).

In the Case 13, p.Phe508del/p.Arg334Trp variants in the compound heterozygous state had detected already. This patient had been previously diagnosed with cystic fibrosis and the sweat test were 92.1 mEq/L. The p.Arg334Trp variant (NM_000492.4; c.1000C>T; p.Arg334Trp; chr7:117180284) causes the substitution of a Arginine into an Tryptophan residue at position 334 in exon 8. This variant has a submission in ClinVar (ID: 7139), previously been reported in publication (21) and classified as "Pathogenic" (PM1, PP2, PM2, PM4).

In the Case 16, p.Asn1303Lys/p.Arg334Trp variants were detected in the compound heterozygous state. The sweat test 135 mEq/L was detected in this patient and bronchiectasis developed in the follow-up of the patient. The p.Arg334Trp variant is classified as "Pathogenic" (PM1, PP2, PM2, PM4). p.Asn1303Lys variant (NM_000492.4; c.3909C>G; chr7:117292931) causes the substitution of a Asparagine into an Lysine residue at position 1303 in exon 24. This variant has a submission in ClinVar (ID: 7136), previously been reported in publication (22). The detected variant is classified as "Pathogenic" (PS1, PM5, PM1, PP2, PM2).

In case 17, p.Gly1069Arg variant was detected in the heterozygous state. This patient had normal pulmonary functions and equivocal sweat test (48.6 mEq/L). p.Gly1069Arg is a missense variant (NM_000492.4; c.3205G>A; p.Gly1069Arg; chr7:117251700) causes the substitution of a Glycine into an Arginine residue at position 1069 in exon 75. This variant has been reported in publication (23). The detected variant is classified as "Likely Pathogenic" (PVS1, PP5, PM2).

Detected PRSS1 Variants

The heterozygous variants in the PRSS1 gene were identified in 2 of the 20 ICP patients.

In the Case 9, p.Arg122Cys variant was detected in the heterozygous state. The p.Arg122Cys variant (NM_002769.5; c.364C>T; chr7-142459788) causes the substitution of a Arginine into an Cysteine residue at position 122 in exon 3. This variant has a submission in ClinVar (ID: 11883), previously been reported in publication (24). The detected variant is classified as "Pathogenic" (PS3, PM1, PM5, PP3, PP5).

In the Case 14, p.Gln98ProfsTer10 variant was detected in the heterozygous state. p.Gln98ProfsTer10 variant (NM_002769.5; c.292dup; chr7:142459712) causes the substitution of a Glutamine into an Proline residue at position 98 in exon 3. It is a frameshift variant and gives rise to termination after 10 codons. There is no recorded publication in the literature. To our knowledge, it was detected for the first time in this study. The detected variant is classified as "Likely Pathogenic" (PVS1, PM2).

Detected CTRC Variants

CTRC genotype contributed to the development of ICP in 5% of cases. Heterozygous variant in the CTRC gene was identified in 1 of the 20 ICP patients.

In the Case 19, p.Arg254Trp variant was detected in the heterozygous state. p.Arg254Trp variant (NM_007272.3; c.760C>T; p.Arg254Trp; chr1:15772212) causes the substitution of a Arginine into an Tryptophan residue at position 254 in exon 7. This variant position has been reported in publications (25). The detected variant is classified as "Likely Pathogenic" (PM2, PP3, PP5).

DISCUSSION

Pancreaticobiliary maljunction is the joining of the pancreas and bile duct outside the duodenal wall and in PBM the common channel is longer than normal. Since the control of the sphincter of Oddi over the common channel is lost, the junction cannot be occluded by sphincter contraction. This situation may cause mutual reflux of biliary and pancreatic fluid, and may cause pancreatitis, cholangitis, and biliary tract cancers. PBM may be with (congenital choledochal cyst) or without biliary dilatation (26).

Acute pancreatitis is more common in children with PBM than in adults. Pancreatitis may recur and cause chronic pancreatitis (8). Miyake et al. in their study, in which they evaluated long common channel cases without biliary dilatation retrospectively, reported that abdominal pain and hyperamylasemia were more common in cases with long common channel without biliary dilatation compared to cases with biliary dilatation (27). In our study, we showed that PBM without biliary dilatation with MRCP was present in 52.6% of the cases with ARP.

The gold standard method in the diagnosis of PBM is ERCP. However, since it is an invasive method, MRCP should be preferred when there is a therapeutic necessity or a strong suspicion of ductal anomaly (1, 28). In the literature, the diagnostic roles of MRCP and ERCP in the diagnosis of PBM have been compared and the diagnostic rates of MRCP were reported to be 75-82% (29, 30). However, common channel length is not routinely evaluated in radiological imaging (31). In the radiologic evaluation, the presence of bile duct dilatation is an indicator for the diagnosis of PBM

with biliary dilatation and the presence of gallbladder wall thickening is an indicator for PBM without biliary dilatation. However, cases without biliary dilatation are often diagnosed with gallbladder cancer in adulthood since gallbladder wall thickening is a condition that develops in advancing age due to diffuse epithelial hyperplasia and dysplasia of the gallbladder and in the absence of biliary dilatation, the radiological findings are indefinite (9). In this study, we found that the common channel length was longer than the upper limit determined for age in 52.6% of the cases who had been considered normal, in the primary level radiological evaluations. Although not statistically significant, it is observed that recurrent pancreatitis attacks started at earlier ages in the presence of long common channel. We suggest that this difference may be more significant if the number of patients was higher.

Kumar et al. evaluated risk factors for ARP and chronic pancreatitis (CP) in the INSPPIRE cohort, and found that 48% of children with ARP had mutations in at least 1 gene. They reported that the identified mutations were in the PRSS1 gene (46%) in chronic pancreatitis and in the CFTR gene (34%) in ARP (3). Similar to the INSPPIRE cohort, in our cohort, we identified mutations in at least one gene in 52.63% of patients with ARP. In addition, we found that 36.84% of the cases had CFTR gene mutations.

All of the mutations we identified in the CFTR gene were previously identified and reported to be associated with cystic fibrosis. We found that 4 patients were compound heterozygous (G85E/ L568F, R334W/F508del, F334W/ N1303K), one patient with 2 homozygous mutations in the CFTR gene (D1152H/D1152H, I1051V/I1051V), and 2 patients with mutations heterozygous for CFTR (F508del/- and G1069R/-). The importance of a single heterozygous mutation in the CFTR gene in the pathogenesis of ARP is controversial (32). However, in cases without respiratory findings, CFTR mutations have been associated with conditions such as congenital bilateral absence of the vas deferens and pancreatitis. Cohn et al. showed that the risk of pancreatitis increased fourfold in cystic fibrosis patients with a normal CFTR allele (33). Sobczyn´ska-Tomaszewska et al. showed the F508del/- variant in 3 patients with ARP and chronic pancreatitis. They reported that there may be additional genetic or environmental risk factors in cases with ARP with a single heterozygous CFTR variant (33). In our study, long common channel was present in 2 patients with a single heterozygous mutation. We suggest that, this can be considered as an additional risk factor for the development of long common channel ARP in these 2 patients.

In our study, we detected R122C and Q98PfsTer10 mutations in the analysis of the PRSS1 gene. Le Maréchal et al. reported that the R122C mutation causes hereditary pancreatitis, for the first time in 2001 and this was later supported by other studies (24,35,36). Q98PfsTer10 mutation, defined as “Likely Pathogenic” (PVS1, PP5, PM2), has not been reported in the literature before. To the best of our knowledge, this is the first study to report the Q98PfsTer10 mutation.

In the INSPPIRE cohort, including 301 children with ARP and chronic pancreatitis, CTRC mutation rates of 10% and 5% (respectively) were reported (3). Palermo et al., in their study analyzed genetic variants in children with ARP and CP, found R254W mutation in the CTRC gene in 7% of the cohort (37). In our study, we detected the R254W mutation in 5.26% of our cohort.

One of the limitations of our study is that, although we have shown that ARP attacks begin at an earlier age in long common channel cases, we could not show a statistically significant difference due to the small number of patients. However, one of the aims of our study was to evaluate the rate of the long common channel without biliary dilatation cases without anatomic/obstructive pathology in the primary care evaluation. Therefore, cases with an obstructive/ anatomical diagnosis were not included in the study. Although our focus on this specific group resulted in the small number of patients, it was a good single-center number for this highly specific group. In this context, we think that our study will guide multicenter studies with larger number of patients. Another limitation of our study was that channel lengths measured by MRCP were not confirmed by ERCP. However, according to the EPC/HPSG (The European Pancreatic Club/The Hungarian Pancreatic Study Group) guideline recommendations, MRCP should be the first-choice imaging method in children unless there is a therapeutic necessity (38). In previous studies, it has been shown that MRCP is 75-82% diagnostic in the diagnosis of PBM (29, 30). In this context, we planned to follow up long common channel cases without biliary dilatation in terms of gallbladder cancer that may occur in adulthood. Pre-surgical ERCP can be considered when there is a need for surgery based on the clinical course. Additionally, the study population could be evaluated using endoscopic ultrasound (EUS). However, we do not have the opportunity to perform endoscopic ultrasound in this age group in our clinic. EUS is used safely in adults to detect the presence of chronic pancreatitis and the etiology of acute recurrent pancreatitis. However, data on the use of EUS in children with acute recurrent pancreatitis are limited. In the study of Singh et al. in which they evaluated children with ARP using endoscopic ultrasound, it was reported that EUS can be used safely in the detection of changes due to chronic pancreatitis in children. However, the evaluation of common channel length in children using EUS remains uncertain (39).

CONCLUSION

In this study, we showed the presence of genetic risk factors (CFTR, PRSS1 and CTRC) and PBM without biliary dilatation in a significant proportion of cases labeled as idiopathic ARP. In the pediatric age group, data of long common channel cases without biliary dilatation is insufficient. Considering the possibility of diagnostic delay due to the lack of definite radiological findings in the presence of long common channel without biliary dilatation, we suggest that our study will provide a guide for clinicians and radiologists.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was conducted in accordance with the Declaration of Helsinki, and approved by the Institutional Review Board (or Ethics Committee) of the Ethics Committee of Necmettin Erbakan University Medical Faculty (protocol code:2017/885 and date of approval: 14.04.2017).

Informed Consent: All patients signed the free and informed consent form.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

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Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

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