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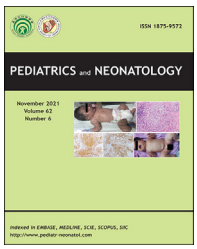

Original Article

Early onset congenital diarrheas; single center experience

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Background: Congenital diarrheal disorders (CDDs) are a rare group of enteropathies that typically present in the early few months of life and pose a diagnostic challenge. We aimed to analyze the clinical findings and outcome of infants with CDDs and share experience about genetic testing.
Methods: Demographic, clinical and genetic findings, and outcome of the patients (n = 24) with CDDs were recorded from hospital files.
Results: The onset of diarrhea was within the neonatal period in 45.8% of the patients. The most frequent causes of CDDs were defects in digestion, absorption and transport of nutrients and electrolytes (DATN) (n = 11, 45.8%) and defects in intestinal immune-related homeostasis (IIH) (n = 6, 25%). Fat malabsorption (n = 6) was the leading cause of defects in DATN. Extra-intestinal manifestations including neurological involvement (25%) and renal involvement (20.8%) were common among the patients. Genetic analyses were performed for 16 patients (targeted gene analysis in 9, congenital diarrhea panel in 3, immune deficiency panel in 1 and whole-exome sequencing in 3 patients). Genetic diagnosis was achieved in 14 of 16 patients (87.5%) with therapeutic consequences in 8 of 16 patients (50%). During the follow-up, 6 patients (25%) died.
Conclusion: The percentage of undefined etiology decreased, and treatment of the patients improved with the increased number of genetic testing in patients with CDDs.

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1. Introduction

Congenital diarrheal disorders (CDDs) are a group of inherited rare enteropathies characterized by severe chronic diarrhea in the first few months with heterogenous etiology and pathogenesis. The differential diagnosis and treatment of CDDs are challenging because of their broad spectrum of clinical presentations ranging from presenting in utero as polyhydramnios and preterm birth to severe diarrhea that leads to dehydration, electrolyte imbalance and intestinal failure soon after birth.[1–3] The exact prevalence of CDDs and frequencies of subgroups remain unknown; because some of the patients may have died due to complications of intestinal failure or septicemia before the definite diagnosis was made. According to network data, congenital chloride diarrhea (67.2%) and Schwachman–Diamond syndrome (11.4%) were leading causes of CDDs among the 61 patients with positive mutations for the causative diseases.[4,5] Furthermore, some selected type CDDs are more common in some ethnic groups due to consanguineous marriages or founder effects such as congenital lactase deficiency (CLD) in Finland, lysinuric protein in tolerance in Finland and Japan, or sucrose isomaltase deficiency in Greenland, Alaska and Canada.[6–8] Ye et al. analyzed the genetic spectrum of 137 children with CDDs and enteropathy in China, *IL10*RA was the most commonly detected pathogenic variant.[9]

The percentage of patients with undefined etiology is decreasing with increasing availability of genetic testing in recent years.[2,9] With next-generation and third-generation sequencing, the number of rare Mendelian diseases with known etiology increased in patients with CDDs. Genetic analysis will not only provide definite and rapid diagnosis but also lead to rapid and targeted therapies in patients with CDDs. Furthermore, the identification of disease-related mutations in the affected probands will help to offer future prenatal diagnosis.

The aim of this study was (i) to review the demographic and clinical findings, laboratory findings and outcome of infants with early-onset (<24 months of age) CDDs and (ii) to share our experience about genetic testing to determine the underlying etiology.

2. Patients and methods

This study included the 24 infants who were hospitalized for early onset-CDDs between since January 2008. Demographic data, clinical and laboratory findings, treatment modalities and outcomes of the patients were recorded. Severe malnutrition was considered as weight for age Z scores were below −3 standard deviations.[10] The diagnoses of anemia, hypogammaglobulinemia, and acid-base disturbance were made as defined elsewhere.[11–13] Serum albumin levels <25 g/L were the criteria used to define hypoalbuminemia. Hyponatremia and hypernatremia were defined as serum sodium <130 and >145 mEq/L, respectively.

Patients were defined and classified according to classification of CDDs defined by Canani et al. and Posovszky.[2,3] They classified congenital diarrheal disorder into four subgroups: (i) defects in digestion, absorption and transport of nutrients and electrolytes (DATN); (ii) defects in enterocyte structure (ES); (iii) defects in enteroendocrine cell differentiation (ECD); and (iv) defects in intestinal immune-related homeostasis (IIH).

Genetic testing was performed (i) in patients whose diagnosis could not be made by clinical and laboratory findings, or (ii) to confirm the diagnosis. As a first step, suspected genes were studied (congenital diarrhea or immunodeficiency panel), thereafter exome sequencing was performed when the results were non-diagnostic. Enrichment of exonic DNA fragments was made by using the Agilent Sureselect V6 Exome Kit. Sequencing was performed on a HiSeq 4000 (Illumina) device as 100-bp, 125-bp or 150-bp, paired end reads. Data filtering considered the suggested mode of inheritance and the minor allele frequency (MAF) in approx. 70.000 exomes in public databases (dbSNP-, ESP- und EXAC) (Supplementary data 1).

The study was approved by the Karadeniz Technical University, Faculty of Medicine ethics board (2020–114). Informed consent was obtained from all participants.

Statistical analyses were performed using the SPSS software package (version 16.0 software, SPSS Inc., Chicago, IL, USA). Descriptive analyses were used for statistical evaluation.

3. Results

3.1. General features

The study included 24 patients (50% F) with CDDs. Age at time of admission to our center was 144.3 ± 175.3 days (range; 1 day-23 months, median; 67 days), and the onset of diarrhea was within the neonatal period in 45.8% of the patients. Pregnancy history was remarkable for 3 patients (12.5%) (oligohydramnios, gestational diabetes mellitus and urinary infection one time each). Parental consanguinity was found in 4 patients (16.7%) and premature birth in 5 patients (20.8%). 12 patients had severe malnutrition (50%).

3.2. Diagnosis

The most frequent causes of CDDs in our patients were defects in DATN (n = 11, 45.8%) and defects in IIH (n = 6, 25%) (Fig. 1).

In defects in DATN group, there were 6 patients with fat malabsorption [1 abetalipoproteinemia, 1 cystic fibrosis (CF), 1 CF + Barth syndrome (BS), 1 chylomicron retention disease (CRD), and 1 Johanson-Blizzard syndrome (JBS)]. Additionally, a 5-month old patient had severe steatorrhea, but the etiology could not be identified by laboratory and endoscopic examinations. Three patients had carbohydrate malabsorption [2 CLD and 1 glucose-galactose malabsorption (GGM)]. Two patients had protein losing enteropathy (PLE) where CD55 deficiency was ruled out by genetic analysis. Median age of the patients at the time of admission was 90 days. Anemia (90.9%), metabolic acidosis (45.4%) and hyponatremia (27.2%) were common in patients with defect in DATN.

There were 6 patients with defects in IIH. Three patients were diagnosed with inflammatory bowel disease after colonoscopic examination (2 ulcerative colitis, and 1 Crohn's

disease). On follow-up, one patient with ulcerative colitis was hospitalized for recurrent fever and the diagnosis of familial Mediterranean fever (FMF) was made based on clinical and genetic findings. One patient was diagnosed with autoimmune enteropathy (AIE) based on histopathological examination and anti-enterocyte IgA positivity. One patient who had hypogammaglobulinemia with chronic diarrhea at the time of admission was diagnosed with common variable immune deficiency (CVID) due to persistent of hypogammaglobulinemia and recurrent sinopulmonary infections on follow-up. One patient was admitted with diffuse cytomegalovirus infection, and on the follow-up, she was diagnosed with MHC Class II deficiency. Immunological phenotype classification of the patients with IIH were immune dysregulation for patients with early-onset colitis (n = 2), B-cell defects for patient with CVID (n = 1), combined antibody defects for patient with MHC Class II deficiency (n = 1), autoinflammatory defects for patient with FMF (n = 1) and defects of tolerance induction/regulatory T-cells for patient with AIE (n = 1). Anemia (83.3%) and, hypogammaglobinemia (50%) were common in patients with defects in IHH, whereas neonatal-onset diarrhea was less common (33.3%) compared to other etiologies.

There were 2 patients with microvillous inclusion disease (MVID) and 1 patient with trichohepatoenteric syndrome (THES) in the group of defects in ES. Additionally, two patients were admitted with chronic diarrhea, neurological findings, pancytopenia, and hyponatremia, and both were diagnosed as familial hemophagocytic lymphohistiocytosis type-5 (FHL-5) by genetic testing. Patients with the defects in ES were commonly admitted in the neonatal period (60%) with anemia (100%) and hyponatremia (100%).

Two siblings with proprotein convertase 1/3 deficiency (PC1/3D) had defects in ECD. Neonatal-onset diarrhea, severe malnutrition, anemia, metabolic acidosis and hypernatremia were present in both of them. Clinical and laboratory findings of the patients with CDDs are shown in Table 1.

3.3. Associated findings

Other system involvements were as follows: neuromuscular involvement in 6 patients (25%) (epilepsy in 1, psychomotor retardation in 1, acute infarct and cerebral effusion in 1, symmetric lesions in neuroimaging in 2 and myopathy in 1), renal involvement in 5 patients (20.8%) (tubulopathy in 4 and renal stones in 1), endocrinological disorders in 4 patients (16.6%) (congenital hypothyroidism in 2 and central diabetes insipidus in 2), skeletal anomaly in 3 patients (12.5%) (nasal agenesis in 1, polydactyl in 1 and dolichocephaly in 1), hepatobiliary involvement in 3 patients (12.5%) (acute hepatitis in 1, cirrhosis in 1 and hepatosplenomegaly in 1), anal atresia, pulmonary involvement (interstitial pneumonia), mucocutaneous involvement (wooly hair), severe neutropenia, pancreatic lipomatosis and cardiological involvement in 1 patient (4.1%) (cardiomegaly) (Fig. 2).

3.4. Genetic analysis

Genetic analyses were performed on 16 patients. Clinical and genetic findings of 13 patients are shown in Table 2 (Supplementary data 2). Targeted gene analysis (TGA) was performed in 8 patients, where congenital diarrhea panel was needed in 3 patients and immune deficiency panel in 1 patient. The definite diagnosis could be made by whole-exome sequencing in 1 patient. Apart from these patients, whole-exome sequence analysis was performed in 2 patients (1 with AIE and 1 with early onset Crohn's disease) but none of them were diagnostic. CF was confirmed by genetic analysis in 1 patient. Genetic diagnosis was achieved in 14 of 16 patients (87.5%). Medical treatment was directed according to genetic study in 8 of 16 patients (50%); hematopoietic stem cell transplantation (HSCT) in patients #1, #12 and #13, colchicine in patient #2 and special diet in patients #6, #8, #9 and #11.

3.5. Treatment modalities

Special personalized diet was prescribed in patients with abetalipoproteinemia (n = 1), CRD (n = 1) and JBS (n = 1). Fructose-free formula was used for patient with GGM (n = 1), lactose-free formula for patients with CLD (n = 2) and carbohydrate-free formula for PC1/3D (n = 2). Pancreatic enzyme replacement was given in patients with fat malabsorption. Corticosteroid was used for patients with inflammatory bowel disease (n = 2) and AIE (n = 1),

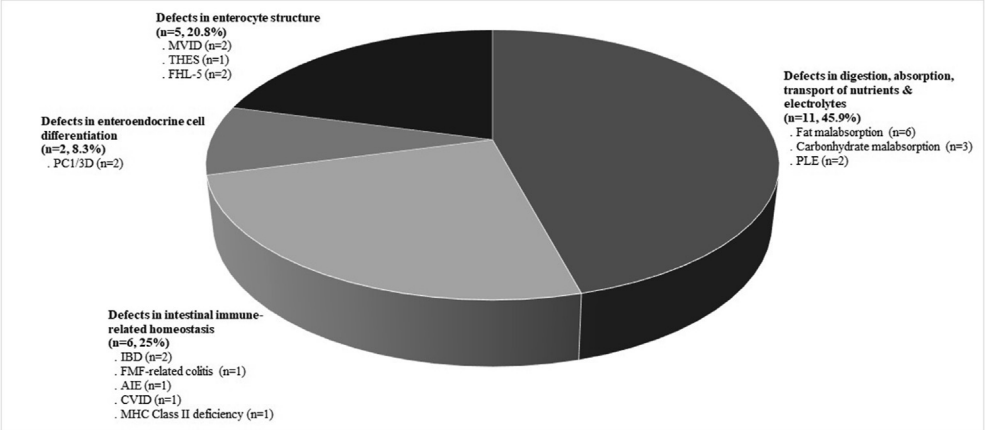

Figure 1 Distribution of etiologies for CDDs (n = 24).

Table 1 Some clinical parameters and laboratory findings of the patients with CDDs[a].

Parameters	Defects in DATN (n = 11)	Defects in IIH (n = 6)	Defects in ES (n = 5)	Defects in ECD (n = 2)	Total (n = 24)
Gender, female, n (%)	3 (27.2)	4 (66.6)	3 (60)	2 (100)	12 (50)
Age at admission, median days	90	91	24	26	67
Neonatal onset of diarrhea, n (%)	4 (36.3)	2 (33.3)	3 (60)	2 (100)	11 (45.8)
Consanguinity, n (%)	1 (9)	2 (33.3)	1 (20)	0	4 (16.6)
Prematurity, n (%)	2 (18.1)	0	2 (40)	1 (50)	5 (20.8)
Severe malnutrition, n (%)	4 (36.3)	4 (66.6)	2 (40)	2 (100)	12 (50)
Anemia, n (%)	10 (90.9)	5 (83.3)	5 (100)	2 (100)	22 (91.6)
Hypoalbuminemia, n (%)	5 (45.4)	4 (66.6)	4 (80)	1 (50)	14 (58.3)
Hypogammaglobulinemia, n (%)	2 (18.1)	3 (50)	2 (40)	0	7 (29.1)
Metabolic acidosis, n (%)	5 (45.4)	0	3 (60)	2 (100)	10 (41.6)
Metabolic alkalosis, n (%)	0	1 (16.6)	1 (20)	0	2 (8.3)
Hyponatremia, n (%)	3 (27.2)	0	5 (100)	0	8 (33.3)
Hypernatremia, n (%)	1 (9)	0	0	2 (100)	2 (8.3)
Genetic testing, positive/tested (%)	5/5 (100)	2/4 (50)	5/5 (100)	2/2 (100)	14/16 (87.5)

[a] CDDs: congenital diarrheal disorders, DATN: digestion, absorption and transport of nutrients and electrolytes, ECD: enteroendocrine cell differentiation, ES: enterocyte structure, IHH: intestinal immune-related homeostasis.

periodic intravenous immunoglobulin for patients with CVID (n = 1), THES (n = 1) and PLE (n = 2), and colchicine for patient with FMF (n = 1). HSCT was performed in two patients with FHL-5 and planned in patient with MHC Class II deficiency.

Prolonged total parenteral nutrition (TPN) was required in 9 patients [(37.5%), 2 MVID, 2 PC1/3D, 1 GGM, 1 FHL-5, 1 AIE, 1 MHC Class II deficiency, 1 THES] for 135.3 ± 78.5 days (range; 15–320 days). The longest duration (>250 days) of TPN was given in patients with PC1/3D (n = 2), THES (n = 1) and FHL-5 (n = 1). Full enteral nutrition was tolerated in 5 patients (55.5%) during the follow-up (2 PC1/3D, 1 GGM, 1 FHL-5 and 1 MHC Class II deficiency).

3.6. Follow-up and outcome of the patients

Patients were followed for 4.7 ± 3.4 years (range 2 days–10 years). Despite diet, liver cirrhosis and renal stones developed in the patient with abetalipoproteinemia. Obesity developed in two siblings with PC1/3D. TPN-related liver diseases developed in 7 patients (elevated liver enzymes in 5, fatty liver in 2, cirrhosis in 1, gall stones in 1). 6 patients (25%) died. The diagnoses of deceased patients were MVID in two patients, and THES, CF + BS, AIE, FHL-5 (after BMT) each in one patient. Causes of death were septic-shock (n = 5) and metabolic decompensation (n = 1).

4. Discussion

In this study, we share our experience about CDDs, and we found that (i) defects in DATN were the leading cause of CDDs, and CDDs may be the initial manifestation of rare diseases such as FHL-5 or MHC Class II deficiency; (ii) extraintestinal system involvement especially neuromuscular, renal and skeletal involvement may be seen with CDDs; (iii) genetic analysis, both targeted and whole-exome, is needed in some cases for the definite diagnosis and management; and (iv) mortality is about 25% in patients with CDDs.

Alterations in DATN represent the most prevalent and well-known group of CDDs. Alterations in epithelial nutrient/electrolyte transporters or disturbance in epithelial enzymes causes congenital diarrhea in most cases.[2,3,14] Mutations in pure electrolyte transporters such as congenital chloride (*SLC26A3*) or sodium (*SLC9A3*) diarrhea or in electrolyte-nutrient co-transporters such as glucose galactose malabsorption (*SLC5A1*) or in regulatory proteins (*GUCY2C*) and other transporters (*SLC10A2*) induce electrolyte rich gastrointestinal fluid loss. Defects in brush-border enzymes involved in carbohydrate digestion such as lactase (*LCT*) and sucrose-isomaltase (*SI*) result in diet-induced diarrhea after the intake of carbohydrate containing foods. Mutations in the proteins involved in fat transport or metabolism cause failure in the absorption or transport of the fats such as abetalipoproteinemia (*MTTP*) or chylomicron retention disease (*SAR1B*). Apart from these disorders, mutations of approximately 20 disorders associated with the defect in DATN have been defined up to date.[1,3,14,15] Recently, a novel genetic entity caused by mutations in *DGAT1* which is involved in cellular triglyceride formation has been defined characterized by electrolyte transport-related diarrhea, vomiting and PLE induced by enteral intake of lipids.[16,17] Typically, histological or ultrastructural features of the enterocytes or villus in DATN are normal except for the patients with fat malabsorption; lipid-laden vacuoles may be seen.[15] In these CDDs, the onset of diarrhea may vary; neonatal onset in carbohydrate malabsorption or electrolyte transporter disorders and late onset in fat malabsorption. Most of the patients had osmotic diarrhea and acid-base and electrolyte disturbances are common. Fat malabsorption encompasses a major subgroup of DATN in our study.

Early-onset CDDs may be result from mutations in genes encoding proteins related to IIH. Altered immune response to pathogens, inflammation or autoimmunity may cause bloody or watery diarrhea with perianal disease, and

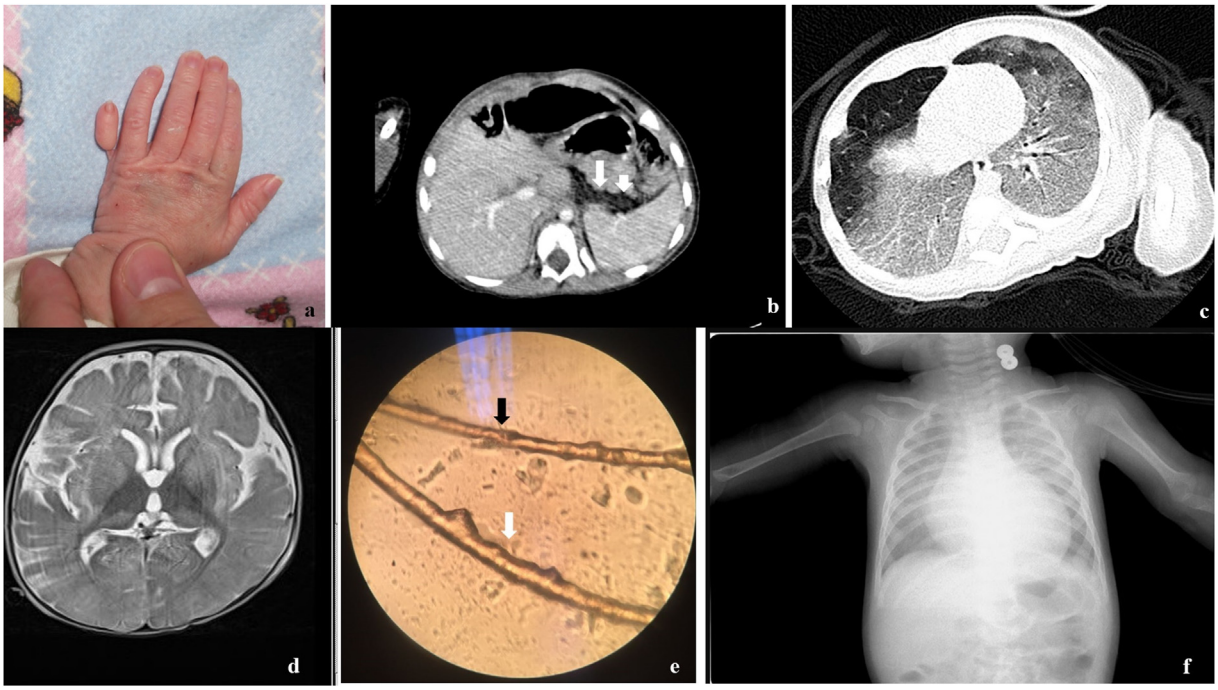

Figure 2 Extraintestinal involvement of the patients with CDDs. a: Postaxial polydactyl in patient with MVID, b: Abdominal CT showed pancreatic lipomatosis (white arrows) in a patient with JBS, c: Interstitial pneumonia with severe diarrhea due to cytomegalovirus infection as initial presentation of the patient with MHC Class II deficiency, d: Axial T2 cranial MR imaging of a patient with severe diarrhea, hyponatremia and convulsion showed bilaterally symmetrical hyperintense lesions in the white and grey matter suggesting hemophagocytic syndrome, e: Microscopic examination of scalp hair of the patient with THES (black arrow) and age and gender-matched healthy control (white arrow) revealed wooly hair, f: X-ray graph of the patient with CF + BS revealed cardiomegaly.

systemic disease and multiorgan involvement in some cases. Early-onset colitis (*IL10*, *IL10*RA and *IL10*RB), IPEX (*FOXP3*) and IPEX-like disorders, APECED, chronic granulomatous disease, antibody defects and hyper- and autoinflammatory defects are the major subgroups of defects of IIH.[2,3,14] Neonatal-onset diarrhea is uncommon and hypogammaglobulinemia may be seen in some cases. Contrary to defects in IHH, patients with the defects in ES commonly present in the neonatal period with anemia and hyponatremia. Most of the patients have extraintestinal manifestations. The three most well described disorders are MVID (*MYO5B* and *STX3*), congenital tufting enteropathy (*EPCAM* and *SPINT2*) and THES (*TTC37* and *SKIV2L*).[2,3] Recently, mutations of *STXBP2* gene (FHL-5) were found to be associated with severe malabsorptive diarrhea. Pagel et al. reported that 14 of 37 patients with FHL-5 had severe diarrhea and malnutrition.[18] Diarrhea was mainly recognized before the development of classical symptoms of FHL-5, and it was common in patients without exon 15 splice-site mutations. The loss of function of *STXBP2* leads to cargo-selective mis-localization of brush-border components and a subapical accumulation of cargo vesicles, as it is known from the loss of polarity phenotype in MVID.[19,20] Patients with defects in ECD mainly presented with congenital generalized osmotic diarrhea in the neonatal period with systemic endocrinopathies such as diabetes insipidus, hypothyroidism and adrenal insufficiency. Four diseases have been defined in this group: enteric anendocrinosis (*NEUROG3*), Mitchell–Riley syndrome (RFX6), X-linked lissencephaly and mental retardation (*ARX*) and PC1/3D (*PCSK1*).[1–3] The latter is associated with obesity in the adolescent period.

The diagnosis of CDDs is based on clinical and laboratory findings and histopathological examinations. A specific diagnosis could not be established in a minority of the cases despite detailed examinations. Guarino et al. noted that it can vary from 0% to 100% according to diagnostic techniques.[21] Genetic analysis including targeted candidate sequencing or whole exome/genome sequencing is an option for diagnosis. Thiagarajah et al. proposed to perform genetic analysis in early stages of evaluation in order to reduce the time required for a definitive diagnosis.[15] In many cases, histopathological and clinical findings may highly suggest a specific etiology, and the diagnosis may be confirmed by targeted testing that may allow for appropriate early treatment. Sometimes these genomic tests may be adjusted according to common mutations in the populations; but whole-exome sequencing should be performed where the diagnosis based on clinical evaluation is unclear. In our study, we performed specific genetic analysis in the majority of cases in order to confirm the diagnosis. TGA was performed in four patients (patient no #1, 2, 4 and 5). Immune deficiency panel was performed in case #1 due to diffuse CMV infection, and congenital diarrhea panel testing for cases #2, 4 and 5. Whole-exome was performed in patient #12 because clinical and laboratory findings did not reveal any specific diagnosis. Apart from these cases, however, whole-exome sequencing was performed in 2 patients to find the underlying genetic defect but it did not specify the etiology. Genetic sequencing for

Table 2 Clinical findings and genetic analysis of 13 patients with CDDs[a].

Patient no	Age/Gender	Main clinical findings in addition to CDDs	Targeted gene/whole genome	Gene and mutation	Diagnosis
#1	2 months/M	Diffuse cytomegalovirus infection (liver, pulmonary and gastrointestinal involvement)	Immune deficiency panel	*RFXANK* gene/c.634C > T (p.R212X) homozygous mutation	MHC Class II deficiency
#2	4 months/F	Early onset enterocolitis and recurrent fever	Congenital diarrhea panel	*MEFV* gene/both homozygous mutation of M694V and R202Q	FMF
#3	1 month/M	Severe secretory IDI, death in siblings with congenital diarrhea, PAS positive granules	Targeted gene for MVID	*MYO5B* gene/c.2014A > T (p.K672) homozygous mutation	MVID
#4	2 days/F	Severe mixt type IDI, hypogammaglobulinemia, dolichocephaly	Congenital diarrhea panel	*TTC37* gene/c.2122C > T (p.Gln708*) homozygous mutation	THES
#5	8 days/F	Severe secretory IDI, metabolic acidosis	Congenital diarrhea panel	*MYO5B* gene/c.1323-2A > G homozygous mutation	MVID
#6	2 months/F	Osmotic diarrhea, hypernatremic dehydration and metabolic acidosis, unresponsive to fructose-based formula, polydipsia and polyuria, low serum levels of c-peptide and insulin, and increased levels of pro-insulin	Targeted gene for PC1/3D	*PCSK1* gene/c.544−2A > G homozygous splice-site mutation	PC1/3D
#7	1 month/F	Osmotic diarrhea, hypernatremic dehydration, metabolic acidosis, sibling of patient #6	Targeted gene for PC1/3D	PCSK1 gene/c.544−2A > G homozygous splice-site mutation	PC1/3D
#8	22 months/M	Fatty diarrhea, dysmorphic features, anal atresia, fatty pancreas on tomography	Targeted gene for JBS	*UBR1* gene/c.497A > G p.(H166R) homozygous mutation	JBS
#9	15 months/M	Fatty diarrhea, A, D, E and K vitamin deficiency, typical white coating on the small intestinal mucosa and vacuolated enterocytes positively with Oil Red O	Targeted gene for CRD	*SAR1B* gene/(c.142delG) homozygous mutation	CRD
#10	5 months/M	Fatty diarrhea, positive sweat chloride test, cardiomegaly, severe lactic acidosis, hypoglycemia, neutropenia, history of BS in close relatives	Targeted gene for CF and BS	• *CFTR* gene/V754 M/c.1408G > A (p.Val470Met) SNP/c.4389G > A (p.Gln1463) SNP compound heterozygote mutation • *TAZ* gene/exon 1 mutation c51.G > C (p.Trp17X)	CF and BS

(*continued on next page*)

Table 2 (*continued*)

Patient no	Age/Gender	Main clinical findings in addition to CDDs	Targeted gene/whole genome	Gene and mutation	Diagnosis
#11	21 days/M	Carbohydrate malabsorption and response to lactose-free diet	Targeted gene for CLD	*LCT* gene/c.4173+1G > T heterozygote mutation	CLD
#12	6 months/F	Osmotic diarrhea, hyponatremia, hypokalemia, metabolic alkalosis, neurological involvement, pancytopenia	Whole-exome sequencing	*STXBP2* gene/c.902+5G > A (IVS10+5G > A) homozygous mutation	FHL-5
#13	6 months/M	Osmotic diarrhea, pancytopenia, neurological involvement, hyponatremia, hemophagocytosis in bone marrow	Targeted gene for HLH	*STXBP2* gene/c.56T > C (p.Ile19Thr)/c.704G > C (p.Arg235Pro) compound heterozygote mutation	FHL-5

[a] BS: Barth syndrome, CDDs: congenital diarrheal disorders, CRD: chylomicron retention disease, CLD: congenital lactase deficiency, CF: cystic fibrosis, FMF: familial Mediterranean fever, FHL-5: familial hemophagocytic lymphohistiocytosis type 5, JBS: Johanson-Blizzard syndrome, MHC: major histocompatibility complex, MVID: microvillus inclusion diseases, PC1/3D: proprotein convertase 1/3 deficiency, SNP: single nucleotide polymorphism, THES: trichohepatoenteric syndrome.

the CDDs has been increasing in recent years. Ye at al studied 137 children with congenital diarrhea and confirmed the genetic diagnosis in 64.2% of the patients.[9] The diagnostic rate was 68%, 48.1% and 71.4% in the bloody, watery, and fatty diarrhea, respectively. The diagnostic rate was higher in the neonatal group. According to CDDs network data, the disease causative mutations were able to be established in 61 of 93 patients (65.5%).[5] In our study, genetic diagnosis was achieved in 87.5% of the patients. On the other hand, genetic examination is also important for the planning the treatment. Apart from TPN or specific diet eliminations some patients may benefit from HSCT or novel therapies. Ye at al diagnosed immune deficiency in some patients in their patient group that underwent HSCT.[9] Similarly, we performed HSCT on two patients with FHL-5 and planned to on one patient with MHC Class II deficiency. Additionally, increased numbers of genetic testing will help us to make a correlation between the genotype-phenotype. This is especially important for some patients with ES because studies showed that some patients with tufting enteropathy may have enteral autonomy in adult life, and early intestinal transplantation should be avoided in these patients.[22] Early genetic testing may give some clues about future enteral autonomy. In addition, LeBlanc et al. reported that early diagnosis of the patients with congenital diarrhea by genetic analysis is also cost-effective.[23]

Extraintestinal involvement may be seen in patients with CDDs especially those with the defects in ES and ECD. Neurological involvement was reported in 55% of the patients with FHL-5 as hyperintense symmetric lesions. Skin and hair abnormalities, cardiac involvement and dysmorphic features were reported in patient with THES. Renal Fanconi syndrome or cholestasis was reported in patients with MVID. Endocrinopathies are common in ECD. Extraintestinal anomalies such as absent corpus callosum, dysplastic kidneys or "tissue-paper like" skin were defined in the patient group of Larcher et al.[24] Neurological and renal involvement and endocrinopathies were common in our patient group. Interestingly, postaxial polydactyly in association with MVID has not been reported previously. These extraintestinal findings may represent a clue for the clinicians in the differential diagnosis of patients with CDDs.

Mortality of CDD varied between 3.7% and 31.5% in previous studies, depending on the patient characteristics.[21,25] Mortality is common in patients with defects in ES. Long-term TPN or bowel transplantation is essential in this patient group. Death is generally related with infectious complications. In our study, mortality rate was 25%, and it was common in patients with defects in ES.

In conclusion, we share our experience about the etiology, clinical feature, and outcome of the patients with CDD. Many diagnostic algorithms were developed for the diagnosis of CDDs, but we think that genetic examinations must be included at the top of diagnostic algorithms not only for specific diagnoses but also for treatment.

Declaration of competing interest

None.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.pedneo.2021.05.024.