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ORIGINAL ARTICLE

The impact of combining educational program with the improving of infrastructure to diagnose on early detection of primary immunodeficiencies in children

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Abstract

Early detection of primary immunodeficiency diseases (PID) is vital for adequate prevention and management of PID infectious complications. The objective of this study was to evaluate the impact of a model combining physician education and public awareness with the infrastructure to diagnose PID to improve its early detection in children. Three approaches were combined and the results were followed from February 2017 to February 2019 in Ternopil region, Ukraine: the education of primary care physicians and other specialists on early PID detection using workshops, trainings, and targeted publications; organization of public events and media appearances to raise PID awareness; performing immunological testing for patients with suspected PID. Among the 150 individuals that were screened, PID was diagnosed in 19 patients (12.7%). The majority of diagnosed PID cases were combined immunodeficiency with associated or syndromic features, followed by antibody deficiencies. Patients referred by the specialist doctors had the highest percentage of confirmed PID compared with those referred by primary care physicians ($p = 0.0273$) and risk group patients ($p = 0.0447$). Among warning signs in patients with PID, two or more pneumonias within 1 year occurred most often (26.3%), followed by failure of an infant to gain weight or grow normally (21.1%). Among other signs of PID, dysmorphic features and microcephaly were the most prevalent (31.6%). In conclusion, a program combining physician education and public awareness with infrastructure needed to diagnose primary immunodeficiency diseases is an effective tool for early PID diagnosis. Physician education was a more effective tool compared with rising public awareness.

Keywords Primary immunodeficiencies · Diagnostic · Physician education · Public awareness

Introduction

Primary immunodeficiency diseases (PID) is a group of more than 350 rare, chronic disorders in which a part of the body's immune system is missing or functions improperly. Detection of early symptoms is very important for adequate prevention and management of infectious complications associated with PID [1].

The rapid development of genetic testing allows discovering new PID every year [2]. In most countries, including Ukraine, the delayed and poor diagnosis of PID is an important problem [3].

The incidence of PID in Europe is varying from 1 to 10 per 100,000 inhabitants [4]. Currently in Ukraine, there are about 1500 children diagnosed with PID and receiving specialized immunological care, which suggests that a significant number of PID cases had not been detected and diagnosed [5]. The rate of PID diagnosis in Ternopil region of Ukraine is similar or

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even worse. The prevalence of PID in Ternopil region is lower than that in Ukraine (1.03/100,000 in Ternopil region vs. 1.23/100,000 in Ukraine). Therefore, the issue of early diagnosis, timely and effective treatment, prevention of complications, and improved prognosis remains an extremely relevant one.

Our previous studies have demonstrated that physicians, postgraduate medical students of different specialties, and medical students all have insufficient knowledge about PID [3, 6, 7].

The other reason of poor PID diagnostic in Ukraine is insufficient access to diagnostic testing, as the state-run clinics and hospitals cannot afford to offer the costly immunological testing; often, it is left to the patients to seek commercial testing options.

In 2003, Jeffrey Modell Foundation (JMF) has launched a campaign of physician education and raising public awareness about PID. The goal of the campaign is to raise awareness, ensure early diagnosis and proper treatment of these diseases [1, 8]. The results of this campaign were evaluated in many countries of the world and have shown a significant improvement in the diagnosis and treatment of children with PID.

In eastern and central Europe since 2004, the implementation of J PROJECT under the leadership of Professor Laszlo Marodi (Hungary) has resulted in significant increase of PID diagnoses. The main objective of the project was to raise physician awareness through education [9]. After implementation of the program from 2004 to 2007, the improved diagnostic approach resulted in the number of patients with confirmed PID diagnosis increasing from tens to thousands. Today, the J PROJECT program geography has considerably expanded and it covers now 29 countries, including Egypt, Kazakhstan, Azerbaijan, Tajikistan, and Iran [10].

To improve PID detection in our area, we have launched a project "Implementation of the Model of Combining Physician Education and Public Awareness with the Infrastructure to Diagnose Primary Immunodeficiency Diseases in Children in Western Ukraine". The project received support from the Jeffrey Modell Foundation.

The main aim of this project was to improve detection of PID in children on its early stages through the implementation of a methodology combining education of the physicians, public awareness, immunological testing, and effective medical assistance at various stages of these diseases.

Methods

From February 2017 to February 2019, a combination of three approaches and its results were followed in Ternopil region, Ukraine. The first approach involved education of primary care physicians (pediatricians, general practice (GP) doctors) and other specialists (rheumatologists, otolaryngologists, pulmonologists, surgeons, etc.) on early PID symptoms and their detection using workshops, trainings, and targeted publications. The second approach was organization of public events and media appearances to raise PID awareness among the public. The third approach was to perform immunological testing for patients with suspected PID.

Physician education

Lectures, workshops, and trainings for primary care physicians (pediatricians and GPs) on early PID detection were organized in every district of Ternopil region (in total, 15 districts were covered) within the first year of the project. Overall, 540 physicians and nurses attended the lectures and workshops, including primary care physicians: pediatricians, general practitioners, and other specialists. During the visits to district hospitals, the main topics of lectures were the clinical symptoms, warning signs of PID, and the main stages of laboratory diagnostics. We also spoke about the problems of early diagnosis of primary immunodeficiencies in children. We carried out workshops based on the analysis of clinical cases, such as: "Who should be referred to an immunologist," "Diagnosis of antibody deficiency," and "Recurrent fever in children". Particular attention was paid to the most common PIDs found in our region, namely, Nijmegen breakage syndrome (NBS), Di George syndrome (DGS), and ataxia-telangiectasia (AT). Certain educational materials on PID warning signs and testing stages were translated into Ukrainian. Others were created anew by the members of our team. All these educational materials were published and disseminated among the physicians.

We also visited outpatient clinics and units in Ternopil city, talking to doctors, as well as the patients and their parents about the PID warning signs.

During the second year of our project, we had focused our efforts on training the specialists (such as rheumatologists, otolaryngologists, pulmonologists, allergists, dentists, endocrinologists, and surgeons) on early PID symptoms and detection.

We organized workshops for the specialist doctors targeting specific disease symptoms that they could face in their practice. For example, the workshop "Arthritis in children with primary immunodeficiencies" was presented to pediatric rheumatologists, "Changes in periodontal tissues and oral mucosa in children with primary immunodeficiency" to dentists, and "Primary immunodeficiency in the practice of a neonatologist" to neonatologists. We also presented at specialist conferences with relevant reports on PID diagnostics. In total, 820 specialists (pulmonologists, pediatric neurologists, otolaryngologists, cardiologists, gastroenterologists, pediatric rheumatologists, allergists, neonatologists, dentists, pediatric hematologists, surgeons, orthopedists, pediatric infectionists) attended the lectures and presentations.

We also disseminated the knowledge about PID among medical students, involving them in preparing reports for student meetings, and in discussing clinical cases.

Public awareness

A number of public events were organized during the same period to raise awareness of PID. We publicized and joined the events of World Primary Immunodeficiencies Week (WPIW). In one of the events, medical students, physicians, and patients have launched the "Burst the Bubble" balloons to raise public awareness of PID. Under the supervision of their professors, medical students visited the outpatient departments of Ternopil Regional and City Children's hospitals, talking to the patients and their parents about the warning signs of PID. Together with their teachers, medical students organized an information campaign at Ternopil's main pedestrian square, talking to the public about PID and distributing information materials.

Program participants have visited a regional television studio to discuss the issue of primary immunodeficiency on air. The interview touched on defining PID, their warning signs, and treatment options.

Infrastructure to diagnose PID

The examination of the children at risk for PID was conducted during the visits to district hospitals. For the children with suspected PID, we introduced examination record cards. Those children that had one or more warning signs, or had other PID symptoms, were referred to the Regional Children's hospital for further examination.

Primary care physicians and other specialists referred children with suspected PID to an immunologist for an in-depth exam clinical manifestations, history, and physical examination.

A targeted screening of children at risk was also carried out: children with microcephaly were examined for possible NBS; children with autoimmune diseases received an immunological especially in the cases of an early onset, or with atypical course; children suffering from diabetes were examined to detect selective IgA deficiency.

We offered free immunological testing for the patients with suspected PID: complete blood count (CBC) and differential, immunoglobulin levels IgG, IgM, IgA; IgG subclass analysis; lymphocyte surface markers CD3/CD4/CD8/CD19/CD56; neutrophil oxidation burst.

Currently, Ukrainian labs only offer genetic testing for NBS and AT, and FISH test for DGS. Working together with laboratories in Europe and the USA, we ordered genetic testing for all other diseases.

Prior written informed consent was obtained from all individual participants of the study cohort. The experimental protocol was carried out in accordance with the guidelines of the Helsinki Declaration of 1975, as revised in 2000.

Statistical analysis

The statistical analysis of the results was carried out using a statistical package STATISTICA 10.0 and table editor Microsoft Excel 2003. The comparison of frequency indices in the observation groups was performed using the χ^2 criterion. The differences between the values were considered significant at $p < 0.05$.

Results

In total, 150 patients were selected for a follow-up in-depth exam: 44 in the first year of the program and 106 in the second year (Table 1).

In the first year, an equal number of patients was referred by primary care physicians and specialist doctors, while in the second year, the number of referrals by specialist doctors was three times as high as those from primary care physicians (38.7% versus 12.3%, $p = 0.0011$).

In the second year, there were five patients from the public.

We screened 47 risk group patients in the second year: 44 with diabetes mellitus, 2 with an atypical course of autoimmune diseases, and 1 with microcephaly.

Mean patient's age was 5.65 years (range 2 months–20 years). There was an equal number of boys and girls, 75 each (50%).

Table 1 Breakdown of referred patients with suspected PID for further examination

Who referred	1st year *n*/%	2nd year *n*/%	Total *n*/%
Primary care physicians	21/47.7	13/12.3	34/22.7
Public	0	5/4.7	5/3.3
Pediatric specialists:	22/50.0	41/38.7	63/42.0
Pulmonologists	5/11.4	8/7.5	13/8.7
Infectious diseases	4/9.1	9/8.5	13/8.7
Gastroenterologists	6/13.6	8/7.5	14/9.3
Neurologists	1/2.3	7/6.6	8/5.3
Hematologists	3/6.8	1/0.9	4/2.7
Surgeons	1/2.3	2/1.9	3/2.0
Cardiologists	1/2.3	1/0.9	2/1.3
Rheumatologists	1/2.3	1/0.9	2/1.3
Otolaryngologists	0	2/1.9	2/1.3
Dermatologists	0	1/0.9	1/0.7
Endocrinologists	0	1/0.9	1/0.7
Risk groups patients	1/2.3	47/44.3	48/32.0
Total	44/29.3	106/70.7	150/100

Of the 150 examined individuals, PID was diagnosed in 19 patients (12.7%) (Table 2).

Among the diagnosed PIDs, the most common ones were combined immunodeficiencies (CID) with associated or syndromic features, followed by antibody deficiencies (Table 3). Phagocytosis defects were detected less often. We did not detect any cases of severe combined immunodeficiencies (SCID), diseases of immune dysregulation, defects in intrinsic and innate immunity, auto-inflammatory disorders, and complement deficiencies.

Five PIDs (26.3% of the total) were diagnosed in the first year of the project, representing 11.4% of the examined patients. The second year of the project, we diagnosed 14 PIDs (73.7% of the total), which constituted 13.2% of the examined patients (Table 4). We are still following up 12 patients with suspected PIDs that require monitoring their disorders and repeat examinations to finalize the diagnosis.

The highest percentage of PID diagnoses was among the patients referred by specialist doctors ($p = 0.0273$ compared with primary care physicians, and $p = 0.0447$ compared with risk group patients).

The mean age of patients with diagnosed PID was 6.29 years (range 2 months–16 years). This group had more boys than girls (11 or 57.9%). The mean delay from initial symptoms to diagnosis was 35.8 months, ranged from 1 to 156 months.

Among the warning signs, as defined by the JMF Medical Advisory Board, two or more pneumonias within 1 year were the most common indicator (26.3%), followed by the failure of an infant to gain weight or grow normally (21.1%) (Table 5). Recurrent sinusitis and the need for intravenous antibiotics were observed in 15.8% of the cases.

Among other signs of PID, dysmorphic features and microcephaly were the most frequent (31.6%). Chronic diarrhea with malabsorbtion, lymphopenia, and lymphadenopathy were observed in 3 children (15.8%), while the other signs—less frequent. All patients presented at least one and up to five signs. Two or more signs were observed in 15 PID patients (78.9%).

Table 2 PID diagnosed in the examined children

Diagnosis	$N = 19$
Nijmegen breakage syndrome	3
22q11.2 deletion syndrome (Di George syndrome)	3
Ataxia-telangiectasia	1
Common variable immunodeficiency (CVID)	2
Selective IgA deficiency	3
IgG subclasses deficiency	2
Chronic granulomatous disease	1
Congenital neutropenia, unspecified	2
Cartilage-hair hypoplasia	1
FINCA syndrome	1

Table 3 PID groups diagnosed

Group	*N*/%
Antibody deficiencies	7/36.8
CID with associated or syndromic features	8/42.2
Congenital defects of phagocyte number, function, or both	3/15.8
New unclassified (FINCA syndrome)	1/5.2

NBS was diagnosed in three patients: in two of them at the age of 2 months, and in one patient at the age of 9 years. Their weight at birth ranged from 1320 to 3050 g, all had microcephaly (27–29.5 sm), dysmorphic facial features, and lymphopenia (700–1800 cells/mm^3). One child diagnosed with NBS had a pronounced hypogammaglobulinemia since the early age (IgG 94.2 mg/dL, IgA < 2.6 mg/dL), as well as reduced subpopulations of lymphocytes (CD3–46%/506 cells/mm^3; CD4–26%/286 cells/mm^3; CD8–16%/176 cells/mm^3; CD19–10%/110 cells/mm^3), even though, after following up the child for a year, we did not detect an infectious syndrome. Another child with early diagnosed NBS did not have hypogammaglobulinemia, and the deviations in subpopulations of lymphocytes were insignificant; however, the child had recurrent urinary tract infections. A boy diagnosed with NBS at the age of nine has a delayed physical development and suffers from malabsorption syndrome and recurrent upper respiratory tract infections, while possessing normal immunoglobulin levels and subpopulations of lymphocytes, as well as normal mental development.

The Di George (22q11.2 deletion) syndrome was diagnosed in three patients, with the age ranging from 6 months to 1.5 years. All these patients had congenital heart disease (CHD): tetralogy of Fallot in one patient, and ventriculoseptal defect in the rest that was associated with pulmonary stenosis in one child and pulmonary atresia in the other. The severity of their condition was primarily due to the CHD. In all these cases, children underwent surgical interventions. Thymic aplasia was revealed in all of the cases, T cell lymphopenia in one patient, and recurrent infections in one patient. Characteristic facial features were found in two cases. Developmental disability was presented as motor delays in two cases. Hypocalcemia was not revealed, and seizures were absent. One patient subsequently died at home from heart failure.

Ataxia-telangiectasia was diagnosed in a 16-year-old boy. He was hospitalized to the neurological department for progressive neurodegenerative disease with ataxia and cognitive impairment. AT was initially suspected at the age of six; however, a genetic test at the time failed to detect ATM gene mutation, leading to delayed diagnosis of the disease. At the time of hospitalization, the boy had a pronounced ataxia,

Table 4 The number of diagnosed PIDs depending on the referring specialist

Who referred	1st year $N = 5$	2nd year $N = 14$	% diagnosed/ referred	% diagnosed/ total
Primary care physicians	1	0	2.9	5.3
Pediatric specialists	4	10	22.2	73.7
Public	-	1	20.0	5.3
Risk group patients	-	3	6.3	15.7

ocular telangiectasia, increased level of alpha-fetoprotein 393.3 IU/ml (reference < 9.96 IU/ml), lymphopenia (1200–1500 cells/mm^3), T cell lymphopenia (CD3–400 cells/mm^3), and dysglobulinemia (low level of IgA < 2 mg/dL and IgE – 0.532 IU/ml, high level of IgM – 407 mg/dL, while IgG was normal – 1469.6 mg/dL). MRI revealed cerebellar atrophy. He has no history of severe infections.

Two girls, 4- and 7-year-old, were diagnosed with neutropenia. The 7-year-old had the onset of the symptoms at 10 months of age, during the first teeth eruption. The symptoms included fever, stomatitis, gingivitis, enlarged cervical lymph nodes, and fatigue. The patient's mother reported that ever since every 2 to 4 weeks, the girl has been having ulcerative stomatitis episodes, which are accompanied by fever, enlarged and painful cervical lymph nodes, increased symptoms of gingivitis, and sometimes angular cheilitis. We have been following up the girl for the last 1.5 years, and during this time recorded exacerbation episodes every 3–4 weeks, with each lasting 4–6 days. A number of complete blood counts revealed neutropenia (260–960/mm^3), anemia, and compensatory monocytosis. Cyclic neutropenia was suspected, but a direct sequencing of all exons of the ELANE gene did not reveal any pathogenic variants.

The second girl was also diagnosed with grave neutropenia, but without severe infectious syndrome. In both children, hematological pathology was excluded. Currently, WES is being performed to clarify the diagnosis.

Chronic granulomatous disease (CGD) was diagnosed in a 2-year-old boy who lives in a neighboring region. The patient's mother reported he had toxic erythema and pneumonia in the neonatal period; then, tumor-like lymphadenitis was observed twice in different places at the age of 4 months, and since the age of 6 months, he produces liquid feces with blood 4–5 times a day. Colonoscopy with biopsy was conducted, and eosinophilic colitis was diagnosed. The child presented chronic anemia and delayed physical development. Neutrophil oxidative burst test was questionable; however, WES revealed a splice site mutation in CYBB, allowing to diagnose X-linked CGD.

Selective IgA deficiency was diagnosed in 3 patients: 1 patient with autism, cholelithiasis, and recurrent tonsillofaringitis, and 2 asymptomatic patients with diabetes mellitus.

Cartilage-hair hypoplasia was revealed in a 15-year-old girl with severe, poorly controlled diabetes mellitus. At that age, her height was 129 cm (− 4, 8σ), and weight 36 kg. Diabetes mellitus was diagnosed at the age of 18 months. She presented with diabetes mellitus complications such as Mauriac syndrome, nephropathy, and neuropathy. In addition, a girl has some skeletal abnormalities, teeth abnormalities, periodontitis, gingivitis, fine, sparse hair, seborrheic dermatitis, a distended belly since the first year of life, chronic anemia, periodic lymphopenia, and increased erythrocyte sedimentation rate (ESR). Genetic testing revealed pathogenic variant in PMRP.

A patient with transient hypogammaglobulinemia, lymphopenia, recurrent respiratory symptoms, malabsorbtion, and neurological disorders was diagnosed with FINCA syndrome (fibrosis, neurodegeneration, and cerebral angiomatosis). It is a new syndrome, first described in 2018.

Discussion

Our study demonstrates that children with suspected PIDs were most common referred by specialist doctors (42%), while primary care physicians were referred 22.7% of the children. Only five patients (3.3%) referred after public events. While in the first year of the study, there was no significant difference between the referrals of children with suspected PID by primary care physicians and specialist doctors, in contrast, in the second year, the proportion of children referred by specialist doctors did not change significantly, while the proportion of children referred by primary care physicians has decreased. The highest percentage of diagnosed PIDs was in children referred by specialist doctors (73.7%). Examining risk group children had a slightly lower effectiveness and even a smaller proportion of children were diagnosed with PID after being referred by primary care physicians or public. These results correlate with a previous study, which also showed that the largest proportion of children subsequently diagnosed with PIDs were referred by hospital specialists [11]. This study also suggested that improving physician's education rather than raising general awareness is a more effective way to improve PID diagnosis, a conclusion that also correlates with our results.

Table 5 Clinical manifestations in patients with PID

Sign	$N = 19$	%
Warning signs (as developed by the JMF Medical Advisory Board)		
Four or more new ear infections within 1 year	1	5.2
Two or more serious sinus infections within 1 year	3	15.8
Two or more pneumonias within 1 year	5	26.3
Failure of an infant to gain weight or grow normally	4	21.1
Recurrent, deep skin or organ abscesses	1	5.2
Persistent thrush in mouth or fungal infection on skin	1	5.2
Need for intravenous antibiotics to clear infections	3	15.8
Two or more months on antibiotics with little effect	1	5.2
Two or more deep-seated infections including septicemia	1	5.2
A family history of PID	0	-
Other signs		
Bronchiectasis	0	-
Delayed separation of the umbilical cord	0	-
Complicated course of BCG vaccination	0	-
Chronic diarrhea (over 1 month) with malabsorbtion	3	15.8
Thrombocytopenia and/or small their size	1	5.2
Neutropenia	2	10.5
Lymphopenia	3	15.8
Recurrent unexplained fevers of than 38C with inflammatory changes of blood	0	-
Recurrent, non-causative, non-inflammatory edema of the skin or mucous membranes	0	-
Ataxia	1	5.2
Telangiectasia	1	5.2
Microcephaly, dysmorphic features	6	31.6
Congenital heart disease with/or without seizures on the background of hypocalciemia	2	10.5
Arthritis in the early age	0	-
Late teeth eruption	2	10.5
Difficulties in the treatment of respiratory tract obstruction	2	10.5
Unusual autoimmunity and/or lymphoproliferative disorders	1	5.2
Partial albinism, abnormal hair structure	0	
Gingivitis, aphthous stomatitis	1	5.2
Lymphadenopathy or absence of lymphoid organs	3	15.8
Hyperplasia of the organs	2	10.5
Vasculitis	0	-
Infectious, causes by atypical pathogens or atypical localization	1	5.2
Recurrent infectious, causes by the same pathogens	1	5.2
Family child death due to infectious or oncological diseases	2	10.5

Our results point out to the need for ongoing professional development of primary care physicians and specialist doctors by reiterating the main diagnostic approaches and covering new aspects of these diseases, since even a few cases of PID diagnosis after a referral by primary care physicians can prevent complications and improve quality of life.

In this study, the proportion of confirmed PID diagnoses among the children we have examined was 12.7%, which corresponds to the results of other studies [12].

Among the diagnosed PIDs, the most prevalent was CID with associated or syndromic features (42.2%), followed by an antibody deficiency (36.8%). This is in contrast to the data of existing PID patient registries [1, 4, 5, 13], in which the highest proportion of PIDs falls within antibody deficiencies. However, other similar studies also showed the prevalence of T lymphocyte PID [11], as well-defined syndromes [12].

In our study, the following warning signs of PID, as defined by the JMF Medical Advisory Board, had the highest diagnostic value: recurrent pneumonia, the failure of an infant to gain weight or grow normally, need for intravenous antibiotics, and recurrent sinusitis. Subbarayan A. et al. (2011) similarly highlighted the failure of an infant to gain weight and need for intravenous antibiotics as the most important signs in identifying children with PID [11]. However, they suggest the

key sign for diagnostic PID is the family history of PID. None of our patients with PID diagnosed had family history of PID. In our study, only two children had a family history of child death from infectious and oncological diseases. This relatively small number can be due to the ethnic and regional variation in the prevalence of PID in different countries [11]. In addition, PID started being diagnosed in Ukraine only recently; therefore, when taking family history, more attention should be paid to early child deaths from infections and oncological diseases and to the occurrence of serious but undetermined illnesses in the family.

We also evaluated other signs that had been suggested by Ukrainian Association of Pediatric Immunology as significant for the diagnosis of PID. The most meaningful for the diagnosis of PID were dysmorphic features and microcephaly (occurred in 31.6% of patients), chronic diarrhea with malabsorbtion, lymphopenia, lymphadenopathy, or absence of lymphoid organs (each of them occurred in 15.8% of patients). The most common PIDs diagnosed in infants (NBS, 22q11.2 deletion syndrome, cartilage-hair hypoplasia) explain the high frequency of dysmorphic features and microcephaly.

Thus, we suggest that additional signs such as dysmorphic features and microcephaly, lymphopenia, chronic diarrhea with malabsorbtion, and lymphadenopathy or absence of lymphoid organs should also be considered in suspected PID cases.

The presence of 2 or more signs increases the probability of PID diagnosis.

Nosological forms of PID reflect certain patterns characteristic of the region, since most of our NBS patients are of Slav ancestry and carry a major founder mutation 657del5 in the exon 6 of the NBS1 gene [14]. High carrier frequency of the 657del5 mutation (1/177) was found in three Slav-dominated countries of Poland, Czech Republic, and Ukraine.

22q11.2 deletion syndrome is also relatively common in our area and is characterized by variability of its clinical manifestations [15].

This study underscored regional problems of PID diagnostics. A proportion of diagnosed PIDs caused by antibody deficiency remain low, indicating the need for further implementation of training programs as well as screening to determine IgG, IgM, and IgA levels in children with recurrent infections, autoimmune diseases, and children in other risk groups.

We did not detect any cases of SCIDs, diseases of immune dysregulation, defects in intrinsic and innate immunity, autoinflammatory disorders, and complement deficiencies. Improved early diagnosis of SCID is especially needed. Since neonatal screening has not yet been implemented in Ukraine, there is a need to detect and monitor children with lymphopenia, and to determine lymphocyte subpopulations in children with severe infections in the first 3 months of life. Targeted screening for PID in the neonatal period and early infancy significantly improves the early diagnosis of combined immunodeficiencies [16].

Risk group children, such as those with microcephaly, conotrunk heart disease, dysmorphia, and absence of thymus on the X-ray, require follow-up examination and referral to an immunologist. This will improve the effectiveness of the diagnosis of CID with associated or syndromic features.

One of the challenges of PID diagnosis in Ukraine is the lack of genetic testing facilities. Today, patients in Ukraine have access to genetic tests confirming only 3 PID forms: Nijmegen syndrome, ataxia-telangiectasia, and Di George syndrome. Thus, further collaboration between J-Project countries and other counties in the field of molecular testing is vital [8]. At the same time, developing on-site molecular genetic labs and updating existing laboratories in Ukraine is very important.

Despite these challenges, in the first 2 years of the program, we have increased the number of detected PIDs almost threefold, and in comparison to 2 years before awareness campaign (2015–2016) almost fivefold (2 cases per year before versus 9.5 cases per year during awareness campaign).

Conclusions

Early PID detection remains an important issue in Ukraine and Ternopil region.

Our program of combining physician education and public awareness with developing an infrastructure to diagnose primary immunodeficiency diseases proved an effective approach for early diagnosis of PID. Of these components, physician education was more effective compared with raising public awareness. To strengthen this approach and its effectiveness, we suggest holding regular professional development seminars.

Warning signs of PID are useful for early its detection. Our study showed the most significant signs for PID diagnostics were recurrent pneumonia, failure of an infant to gain weight, the need for intravenous antibiotics, and recurrent sinusitis. Other signs also can be used to improve PID detection: dysmorphic features and/or microcephaly, chronic diarrhea with malabsorbtion, lymphopenia, lymphadenopathy, or absence of lymphoid organs. The probability of positive diagnosis increases if there are two or more signs.

Detection and follow-up of children with lymphopenia, as well as determining the lymphocyte subpopulations in children with severe infections during the first 3 months of life, will allow for timely diagnosis of CID in countries where neonatal screening has not been implemented.

Author contributions All authors contributed to the study conception and design. Material preparation, data collection, and analysis were performed by Oksana Boyarchuk, Tetyana Hariyan, and Maria Kinash.

The first draft of the manuscript was written by Oksana Boyarchuk and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

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Compliance with ethical standards

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee (I.Horbachevsky Ternopil State Medical University ethical committee) and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Informed consent Informed consent was obtained from all individual participants included in the study.

Conflict of interest The authors declare that there is no conflict of interest.

References

1. Modell V, Orange JS, Quinn J, Modell F. Global report on primary immunodeficiencies: 2018 update from the Jeffrey Modell Centers Network on disease classification, regional trends, treatment modalities, and physician reported outcomes. Immunol Res. 2018;66:367–80. https://doi.org/10.1007/s12026-018-8996-5.
2. Pessach I, Walter J, Notarangelo LD. Recent advances in primary immunodeficiencies: identification of novel genetic defects and unanticipated phenotypes. Pediatr Res. 2009;6:3–12.
3. Boyarchuk O, Lewandowicz-Uszyńska A, Kinash M, Haliyash N, Sahal I, Kovalchuk T. Physicians' awareness concerning primary immunodeficiencies in the Ternopil Region of Ukraine. Pediatria Polska - Polish J Paediatr. 2018;93(3):221–8. https://doi.org/10.5114/polp.2018.77435.
4. Eades-Perner A, Gathmann B, Knerr V, Guzman D, Veit D, Kindle G, et al. The European internet-based patient and research database for primary immunodeficiencies: results 2004–06. Clin Exp Immunol. 2007;147:306–12. https://doi.org/10.1111/j.1365-2249.2006.03292.x.
5. Chernyshova LI, Bondarenko AV, Kostyuchenko LV, Savvo OM, Volokha AP, Rabosh OV. Epidemiologiya pervynnych imunodeficytiv v Ukrayini za danymy reyestru paciyentiv. Zdorov'e rebenka. 2015;7:16–23.
6. Boyarchuk O, Volyanska L, Kosovska T, Lewandowicz-Uszynska A, Kinash M. Awareness about primary immunodeficiency diseases among medical students. Georgian Med News. 2018;12(285):124–30.
7. Boyarchuk O, Kinash M, Hariyan T, Bakalyuk T. Evaluation of knowledge about primary immunodeficiencies among postgraduate medical students. Arch Balkan Med Union. 2019;54(1):11–9.
8. Modell F, Puente D, Modell V. From genotype to phenotype. Further studies measuring the impact of a Physician Education and Public Awareness Campaign on early diagnosis and management of primary immunodeficiencies. Immunol Res. 2009;44(1):132–49.
9. Marodi L, Casanova JL. Primary immunodeficiency diseases: the J project. Lancet. 2009;373(9682):2179–81.
10. Maródi L, the J Project study group. Fifteen years of the J Project. J Clin Immunol. 2019;39:363.
11. Subbarayan A, Colarusso G, Hughes SM, Gennery AR, Slatter M, Cant AJ, et al. Clinical features that identify children with primary immunodeficiency diseases. Pediatrics. 2011;127(5):810–6.
12. Alvarez-Cardona A, Espinosa-Padilla SE, Reyes SOL, et al. Primary immunodeficiency diseases in Aguascalientes, Mexico: results from an educational program. J Clin Immunol. 2016;36:173–8.
13. Pac M, Bernatowska E. Comprehensive activities to increase recognition of primary immunodeficiency and access to immunoglobulin replacement therapy in Poland. E Eur J Pediatr. 2016;175:1099.
14. Varon R, Seemanova E, Chrzanowska K, Hnateyko O, Piekutowska-Abramczuk D, Krajewska-Walasek M, et al. Clinical ascertainment of Nijmegen breakage syndrome (NBS) and prevalence of the major mutation, 657del5, in three Slav populations. Eur J Hum Genet. 2000;8(11):900–2.
15. Boyarchuk O, Volyanska L, Dmytrash L. Clinical variability of chromosome 22q11.2 deletion syndrome. Central Eur J Immunol. 2017;42(4):412–7.
16. Galal N, Ohida M, Meshaal S, Abd Elaziz D. Targeted screening for primary immunodeficiency disorders in the neonatal period and early infancy. Afr Health Sci. 2019;19(1):1449–59.

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