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Common variable immunodeficiency due to a novel variant in *NFKB1* with a neonatal presentation of parechovirus meningitis and sepsis-like illness

Aleksandra Szczawińska-Popłonyk[a]*, Natalia Popłonyk[b], Katarzyna Bernat-Sitarz[c], Katarzyna Jończyk-Potoczna[d], Agnieszka Pollak[e]

[a]*Poznań University of Medical Sciences, Institute of Pediatrics, Department of Pediatric Pneumonology, Allergy and Clinical Immunology, Szpitalna Poznań, Poland*
[b]*District Hospital In Poznań, Poznań, Poland*
[c]*Children's Memorial Health Institute, Department of Immunology, Aleja Dzieci Polskich Warsaw, Poland*
[d]*Poznań University Of Medical Sciences, Institute of Pediatrics, Department of Pediatric Radiology, Szpitalna Poznań, Poland*
[e]*Medical University of Warsaw, Department of Medical Genetics, Pawińskiego Warsaw, Poland*

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Abstract
Monoallelic variants in the nuclear factor kappa B 1 (*NFKB1)* gene reinforce the functional haploinsufficiency causing immunodeficiency with marked individual and intrafamilial variability in genotype-phenotype correlations. The leading clinical manifestations of NF-κB1 deficiency are recurrent infections and immune dysregulation disorders with autoinflammatory and lymphoproliferative disease. The reported patient initially presented with a neonatal sepsis-like illness with meningitis because of a parechovirus infection. The diagnosis of an inborn error of immunity, common variable immunodeficiency (CVID), was established based on hypogammaglobulinemia, impaired antibody response to vaccines, IgG subclass deficiency, and low numbers of switched memory B cells. Molecular genetic testing using trio whole exome sequencing was done to define the background of the presenting phenotype, and it revealed a novel heterozygous variant of *NFKB1*. Viral meningitis and sepsis-like illness are unusual, previously unreported, infectious complications in NF-κB1 deficiency. The transcriptional NF-κB1 regulatory effect on different target gene repertoires and numerous processes including immune and inflammatory responses may indiacte the vulnerability of deficient patients to severe viral infections. This case report exemplifies the advancement of immunogenetics paving the way for the transition from the initial era of clinical recognition to the era of molecular diagnosis of the pediatric CVID.

***Corresponding author**: Aleksandra Szczawińska-Popłonyk, Poznań University of Medical Sciences, Institute of Pediatrics Department of Pediatric Pneumonology, Allergy and Clinical Immunology, Szpitalna 27/33, 60-572 Poznan, Poland. *Email address*: aszczawinska@ump.edu.pl

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Introduction

The nuclear factor of kappa light chain gene enhancer in B-cells 1 (NF-κB1) plays a pivotal role in the regulation of immune responses and inflammation.[1,2] The NF-κB pathway regulates numerous cellular processes including proliferation, apoptosis, immune response, and inflammation. In the inactive state, NF-κB dimers are sequestrated in the cytoplasm and bound by a set of inhibitory proteins of the IκB family. Upon stimulation, these proteins undergo phosphorylation, ubiquitination, and proteasomal degradation. Subsequently, activated NF-κB complexes translocate to the nucleus where they bind to DNA at κB sites and elicit their effector functions by promoting or repressing the target gene expression.[2-5] The activation of NF-κB dimers occurs through either the classical (canonical) or the alternative (noncanonical) pathways. The classical pathway is initiated by cytokines, such as interleukin (IL)-1 and tumor necrosis factor alpha (TNFα), as well as bacterial products, e.g., lipopolysaccharide (LPS). The alternative pathway is stimulated by binding B-cell activating factor (BAFF, aka tumor necrosis factor ligand superfamily member 13B, TNFSF13B), lymphotoxin, or CD40 costimulatory molecule on antigen-presenting cells.[4,6]

As a key regulator of inducible target gene expression, NF-κB plays an essential role in the development and function of the immune system. Lymphoid organogenesis and homeostasis of primary (bone marrow and thymus), as well as secondary lymphoid tissues (lymph nodes, Peyer patches, mucosa-associated lymphoid tissue (MALT), and the spleen) are reulated by NF-kB.[7,8] In the case of bacterial and viral infections, the canonical NF-κB signaling pathway is stimulated through the recognition of microbial pathogen-associated molecular patterns (PAMPs) by receptors, such as toll-like receptors (TLRs), that elicit subsequent stages of B and T lymphocyte activation, thereby linking the innate and adaptive immune responses.[9,10] The canonical NF-κB pathway, mediated by NF-κB1, has been a focus of interest because of its role in the pathogenesis of several inborn errors of immunity (IEI), such as caspase recruitment domain-containing protein 9 and 11 (CARD9 and CARD11, respectively), or NF-κB essential modulator (NEMO) deficiencies.[11,12] Monoallelic variants in the *NFKB1* gene have been considered as the most common cause of antibody deficiency in the European cohort.13,14). Patients with identifiable variants in *NFKB1* resulting in functional haploinsufficiency may develop common variable immunodeficiency (CVID). Also, a marked clinical individual heterogeneity and intrafamilial variability in genotype–phenotype correlations have been reported.[13-16] The leading clinical manifestations of the NF-κB1 deficiency are recurrent infections of the airways and gastrointestinal tract, with occasionally observable Epstein-Barr virus (EBV)-driven lymphoproliferative disease.[9,17] Since the expression of NF-κB1 is not confined to the immune system, and is an integral part of the homeostasis in the cellular skin components, inborn errors of NF-κB1 signaling may also be characterized by mucosal and cutaneous involvement.[18]

Herein, we report the case of a pediatric patient with an early onset antibody deficiency in whom the CVID diagnosis was established, according to current diagnostic guidelines. In this patient, the CVID diagnosis was supported by molecular genetic analysis revealing a novel heterozygous variant in the *NFKB1* gene.[19] In this report, we expand the clinical characteristics by adding viral meningitis and sepsis-like illness to the spectrum of the NF-κB1 deficiency. This clinical symptomatology is an unusual infectious complication, previously unreported in NF-κB1-deficient individuals.

Case Report

The patient is a 6-year-old girl who is the only child of a nonconsanguinous Caucasian couple. The family history is positive toward chronic diseases: maternal uncle and his son suffer from asthma, and galactosemia was diagnosed in two maternal cousins; furthermore, an infant had died the mother's family with a syndromic disorder. Both parents occasionallysuffer from respiratory infections, but they do not present with autoimmune, allergic, or malignant diseases. During the pregnancy, the mother was treated for a genital herpes simplex (HSV) infection with acyclovir for 4 weeks before childbirth. The child was born at the 38th week gestational age, with an Apgar score of 9, and a birth weight 2980 g.

The child was first referred to the infectious diseases unit of our tertiary care university pediatric hospital at the age of 5 weeks because of fever of 39 degrees Celsius, apathy, and loss of appetite with a preliminary diagnosis of the neonatal sepsis. On admission, she presented with weakness, dehydration, pharyngitis, and stomatitis. However, there were no signs of abdominal organomegaly, lymphadenopathy, or meningismus during physical examination, and the lungs and heart were clear on auscultation. The infectious workup including blood cultures, tracheal aspirate, and blood real time-polymerase chain reaction (RT-PCR) viral and bacterial panels did not show any pathogens. In the cerebrospinal fluid, the parechovirus (HPeV)-RNA was detected thereby supporting the diagnosis of the neonatal sepsis-like illness with central nervous system (CNS) infection. The summary of an infectious workup performed in the patient is displayed in Table S1. The magnetic resonance imaging (MRI) examination with a contrast medium of the brain revealed abnormally increased enhancement of the leptomeninges (pia mater and arachnoid mater). The results of cerebral MRI consistent with meningitis are shown in Supplemental Figure 1. Because of hypogammaglobulinemia regarding all isotypes, IgG 230 mg/dL (N:270-780), IgA 2 mg/dL (N:6-58), and IgM 11 mg/dL (N:12-87), and anemia, hemoglobin 8.8 g/dL and red blood cell count $2.7x10^6$/mcL, transfusions of intravenous immunoglobulins and packed red blood cells were indicated. She recovered completely and her further psychomotor development was age-appropriate. She was vaccinated according to the national vaccination program with the live Bacille Calmette-Guerin (BCG) and anti-hepatitis B vaccines after birth, and then she continued the vaccination schedule with anti-tetanus, anti-diphtheria, inactivated poliomyelitis vaccine (IPV), and as a 13-month-old girl, she was vaccinated with a trivalent measles-mumps-rubella live vaccine without adverse effects following immunization (AEFI). At the age of 8 months,

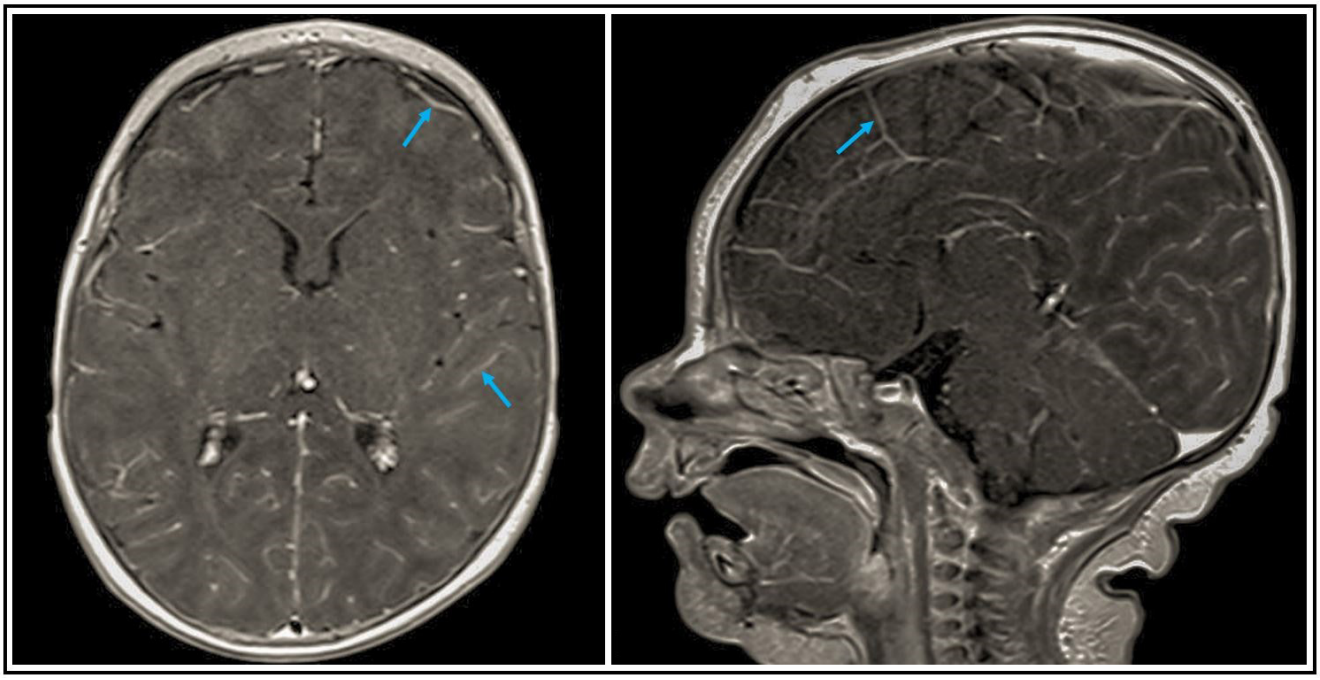

Figure 1 Cerebral MRI examination with a contrast medium showing abnormal increased enhancement of leptomeninges, pia, and arachnoid maters (marked with arrows).

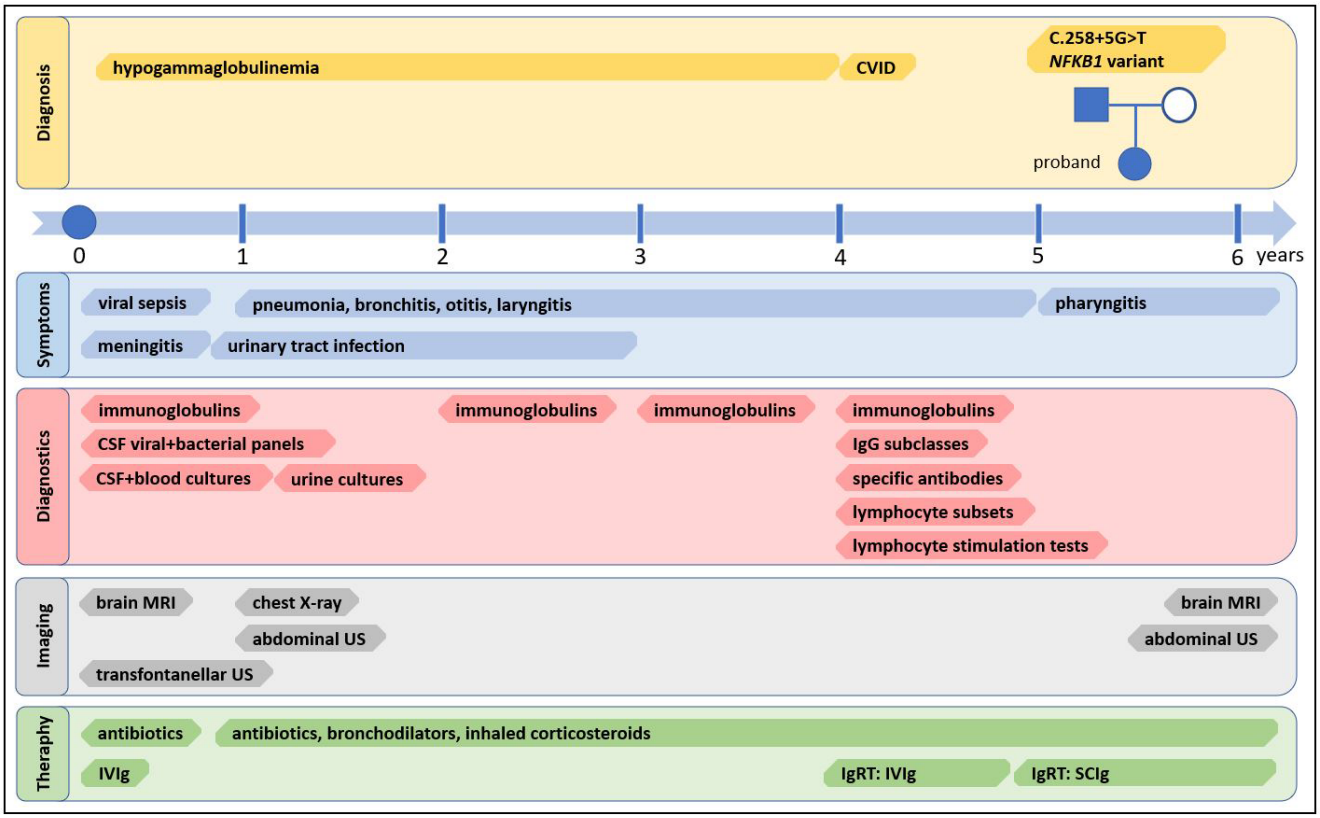

Figure 2 Timeline showcasing the symptomatology, immunodiagnostics, imaging examinations, therapy, and diagnosis in the reported CVID-affected patient with a *NFKB1* variant.

the girl was hospitalized in the department of pediatric nephrology for *Proteus mirabilis* urosepsis. Thereafter, by the age of 3 years, she suffered from recurrent urinary tract infections. Since the end of the first year of life, the girl also presented with recurrent respiratory tract infections: pneumonia, bronchitis, otitis media, tonsillitis, and laryngitis, requiring antibiotic therapy 6–8 times every year.

Referring to the patient's history of very early onset of infections and hypogammaglobulinemia, a suspicion of an inborn IEI was raised. According to the European Society for Immunodeficiencies (ESID) definition,[20] major immunoglobulin class deficiency persisting beyond the age of 4 years, low IgG subclass levels, specific anti-diphtheria and anti-tetanus toxoids antibody deficiency, accompanied by maturational disorders within the B cell compartment prompted us to diagnose CVID. Table 1 summarizes the immunodiagnostics performed in the patient at the CVID diagnosis. To identify the genetic background of the IEI, whole exome sequencing (WES) was performed in the patient and her parents. A heterozygous c.258+5G>T variant in the *NFKB1* gene was found in our patient affected with CVID. Since monoallelic alterations in *NFKB1* are a well-known genetic cause of an early onset CVID,[19,21,22] the *NFKB1* variant was corresponding with the infectious symptomatology and the IEI diagnosis.

Regular intravenous immunoglobulin (IVIg) replacement therapy (Ig-RT) has been implemented in the patient, intermittently accompanied by antibiotic prophylaxis with azithromycin and inhaled therapy with corticosteroids and bronchodilators to better control the respiratory tract infections and symptoms of bronchial obstruction, respectively.

To follow the patient's history, including signs and symptoms; immunodiagnostics; imaging examinations; therapeutic interventions; and diagnosis, see timeline displayed in Figure 1.

Because of the rarity of the reported case with the initial inflammatory manifestations and consequent clinical and immunogenetic IEI diagnosis, an informed consent for publication was obtained from the patient's parent.

Methods

Flow cytometric peripheral blood lymph cell immunophenotyping

Cells were labeled with the following murine fluorochrome-stained monoclonal antibodies: anti-CD45 FITC (fluorescein isothiocyanate), anti-CD14 PE (phycoerythrin), anti-CD19 PE, anti-CD19 PerCP (peridinin chlorophyll protein), anti-IgM FITC, anti-IgD FITC, anti-CD38 APC (allophycocyanin), anti-CD27 PE, anti-CD21 FITC, as well as anti-CD3 FITC, anti-CD4 FITC, CD45RA FITC, CD127 FITC, CD185 FITC, anti-CD8 PE, anti-CD16+CD56 PE, CD25 PE, CD31 PE, CD45RO PE, anti-CD3 PerCP, CD197 PerCP, anti-CD4 APC, and anti-CD8 APC (all Beckton-Dickinson Biosciences, USA). The acquisition of cells and analysis was carried out with the use of the flow cytometer FACSCanto and FACSDiva software (Beckton-Dickinson, USA). With sequential gating on biparametric scattering CD45+CD14- lymphocytes, the following lymphocyte subpopulations were identified:

- CD19+ B cells, immature CD19+CD21lo, immature activated CD19+CD38loCD21lo, transitional CD19+CD38hisIgMhi, non-switched memory CD19+CD27+sIgD+, switched memory CD19+CD27+IgD- B cells, and CD19+CD38hisIgM-plasmablasts
- CD3+ T cells, CD3+CD4+ T helper cells, CD3+CD4+CD31+CD45RA+ recent thymic emigrants, naïve CD3+CD4+CD27+CD45RA+, regulatory CD3+CD4+CD25++CD27-, central memory CD3+CD4+CD27+CD45RO+, effector memory CD3+CD4+CD27-CD45RO+, terminally differentiated CD3+CD4+CD27-CD45RA+, follicular CD3+CD4+CD185+CD45RO+, and regulatory CD3+CD4+CD45RO+CD127-CD25++ T helper cells. Among CD3+CD8+ cytotoxic T cells, the following subsets were distinguished: naïve CD3+CD8+CD197+CD27+CD45RA+, central memory CD3+CD8+CD197+CD27+CD45RO+, effector memory CD3+CD8+CD197-CD27-CD45RO+, and terminally differentiated CD3+CD8+CD197-CD27-CD45RA+ cells.
- CD3-CD16+CD56+ NK cells.

The relative values of PB lymphocytes, the B, T, and NK cells of the total lymphocyte population as well as B and T cell subsets were calculated. The absolute counts of all cell subsets were calculated from the PB leukocyte counts. A comparative analysis was done with the reference cutoff values of B and T cell subsets for pediatric populations of different age groups.

Molecular genetic testing

DNA from the proband and her parents was obtained from peripheral blood and extracted using standard protocols. Library preparation for the WES was performed on the proband's DNA sample with Twist Human Core Exome spiked-in with: Twist mtDNA Panel, Twist RefSeq Panel, and Custom Panel covering variants located in noncoding regions that have been linked to clinical phenotypes according to the ClinVar database (Twist Bioscience, San Francisco, CA, USA). Enriched library was paired-end sequenced (2×100 bp) on NovaSeq 6000 (Illumina, San Diego, CA, USA) to obtain 117 476 696 reads, resulting in mean depth of 133.82× (99.3% of target bases were covered at a minimum of 20×, whereas 99.4% had coverage of min. 10×). Bioinformatic analysis of raw WES data and variants prioritization were performed as previously described (PMID: 34448338**)**. Reads were aligned to the hg38 reference genome sequence and visualized by an Integrative Genomic Viewer.

Presence of the selected variant was subsequently studied by amplicon deep sequencing (ADS) in the proband and her parents. The heterozygous variant in the *NFKB1* gene (LRG_1316_t1:c.258+5G>T) has been indicated for further verification. The *NFKB1* c.258+5G>T variant causes a noncoding transcript, intron, splice donor 5th base change. The variant was absent in null control chromosomes in the GnomAD project and in an in-house database of >10 000 WES of Polish individuals. No clinical diagnostic laboratories have submitted clinically significant assessments for this variant to the ClinVar database. ADS confirmed the presence of the heterozygous c.258+5G>T *NFKB1* variant

Table 1 The immunological workup with antibody-mediated response and peripheral blood flow cytometric immunophenotyping in the patient with NFκB1 deficiency at the CVID diagnosis (aged 4 years).

Immunological workup		
Antibody response	Results	Reference values
Immunoglobulins		
IgG	305 mg/dL	570-1410 mg/dL
IgA	5 mg/dL	65-240 mg/dL
IgM	10 mg/dL	55-210 mg/dL
IgE	<2 kU/L	>2 kU/L
IgG subclasses		
IgG1	195 mg/dL	306-945 mg/dL
IgG2	41 mg/dL	60-345 mg/dL
IgG3	19 mg/dL	99-122 mg/dL
IgG4	2 mg/dL	20-112 mg/dL
Antigen-specific antibodies		
Anti-Diphtheria toxoid	<0.1 IU/mL	>1.0 IU/mL
Anti-Tetanus toxoid	<0.1 IU/mL	>1.0 IU/mL
PB lymphocyte immunophenotyping		
Full PB count		
WBC	6700cc	29.6-49.8%, 1700-3600cc
Lymphocytes CD45+/SSC low	35.0%, 23326cc	
B cell compartment		
B CD19+	13.0%, 315cc	9.7-23.7%, 300-600cc
Transitional B CD19+CD38+IgM++	10.7%, 34cc	4.6-8.3%, 10-40cc
Mature naïve B CD19+CD27-IgD+	83,3%, 262cc	47.3-77.0%, 130-460cc
Non-switched memory B (MZL) CD19+CD27+IgD+	9.7%, 31cc	5.2-20.4%, 20-100cc
Switched memory B CD19+CD27+IgD-	4.7%, 15cc	10.9-30.4%, 40-140cc
Immature B CD19+CD21lo	8.2%, 26cc	5.9-25.8%, 20-120cc
Activated B CD19+CD38loCD21lo	5.4%, 17cc	2.3-10.0%, 10-40cc
Plasmablasts CD19+CD38++IgM-	0.5%, 2cc	0.6-5.3%, 0-3cc
T cell compartment		
T CD3+	77.0%, 1818cc	55.0-97.0%, 850-4300cc
T helper CD3+CD4+	42.0%, 1001cc	26.0-61.0%, 500-2700cc
T suppressor/cytotoxic CD3+CD8+	29.0%, 684cc	13.0-47.0%, 200-1800cc
CD4+/CD8+	1.46	1.5-2.5
CD45RA+/CD45RO+	2.57	>1.0
Recent thymic emigrants CD3+CD4+CD45RA+CD31+	74.2%, 906cc	41.0-81.0%, 170-2600cc
Naïve T helper CD3+CD4+CD45RA+CD27+	78.0%, 952cc	46.0-99.0%, 300-2300cc
Central memory T helper CD3+CD4+CD45RA-CD27+	21.0%, 257cc	0.35-100%, 160-660cc
Effector memory T helper CD3+CD4+CD45RA-CD27-	0.8%, 9cc	0.3-18.0%, 3-89cc
Terminally differentiated memory T helper CD3+CD4+CD45RA+CD27-	0.2%, 3cc	0.0-1.8%, 0-16cc
Follicular CXCR5+ T helper CD3+CD4+CD45RO+CD185+	22.5%, 64cc	7.0-85.0%, 13-170cc
Regulatory T helper CD3+CD4+CD25++CD127-	0.9%, 11cc	4.0-14.0%, 39-150cc
Naïve T suppressor/cytotoxic CD3+CD8+CD27+CD197+	78.4%, 667cc	16.0-100%, 53-1100cc
Central memory T suppressor/cytotoxic CD3+CD8+CD45RA-CD27+CD197+	7.2%, 61cc	1.0-6.0%, 4-64cc
Effector memory T suppressor/cytotoxic CD3+CD8+CD45RA-CD27-CD197-	0.4%, 3cc	5.0-100%, 24-590cc
Terminally differentiated T suppressor/cytotoxic CD3+CD8+CD45RA+CD27-CD197-	1.6%, 13cc	15.0-41.0%, 25-530cc
NK cells		
NK CD3-CD45+CD16+CD56+	6.0%, 184cc	2.0-25.0%, 61-510cc
Lymphocyte stimulation tests		
PHA	37261 cpm	>160000 cpm
PHA stimulation index	142.2	>65.0
Anti-CD3	66818 cpm	>15000 cpm
Anti-CD3 stimulation index	255.0	>60.0
Pansorbin cells	4399 cpm	>2000 cpm
Pansorbin cells stimulation index	16.8	>5.0
Complement		
C3	135 mg/dL	90-180 mg/dL
C4	29 mg/dL	10-40 mg/dL
CH50	100 EqU/mL	70-187 EqU/mL

within the DNA of the proband and that of her father. In silico analysis of the pathogenicity using the dbscSNV engine (PMID: 25416802) indicated that the c.258+5G>T *NFKB1* variant is deleterious (ADA score =0,9989) via impact on pre-mRNA splicing. Selected variant in the *NFKB1* gene has not been described to date in The Human Gene Mutation Database (HGMD Professional 2021.4), although other deleterious variants within the *NFKB1* gene are related to immunodeficiency, common variable, 12 (MIM# 616576), with an autosomal dominant model of inheritance.

Discussion

Herein, authors provide a comprehensive report of a pediatric patient with an ususual initial symptomatology and an early onset antibody deficiency in whom the CVID diagnosis was established and supported by molecular genetic analysis revealing a novel heterozygous variant in the *NFKB1* gene. In light of the ever-increasing progress of insights into molecular genetics underpinning the pediatric CVID, we aimed to show the links between complex genetics and neonatal parechovirus meningitis and sepsis-like illness as a warning clinical symptomatology of an IEI. We also expand the phenotype-genotype characteristics by adding viral meningitis and sepsis-like illness to the spectrum of the NF-κB1 deficiency.

Individuals bearing heterozygous *NFKB1* variants show a marked heterogeneity of clinical phenotypic expression, ranging from severe immunodeficiency with multiorgan autoinflammatory and EBV-mediated lymphoproliferative disease, through antibody deficiency with predisposition to infections, to asymptomatic course in variant-proven relatives.[21,23,24] In a Finnish cohort of carriers of distinct heterozygous variants in *NFKB1*, the clinical penetrance was not complete and with advancing age of affected patients, hypogammaglobulinemia in CVID was less prominent, whereas autoinflammatory disorders were more frequent.[25] However, despite considerable individual and intrafamilial variability of clinical manifestations in patients with CVID and variants in *NFKB1,* severe infectious and noninfectious complications have been reported. These include recurrent sinopulmonary and skin infections, and severe immune dysregulation disorders, such as autoimmune hemolytic anemia (AIH), autoimmune neutropenia,[26] an immune dysregulation, polyendocrinopathy, and enteropathy (IPEX)-like disease,[27] as well as malignant and nonmalignant proliferation.[28] The symptomatology of *NFKB1*-related disease-causing variants remarkably expand the immunogenetic landscape of this IEI.[29-31] These observations correspond with the phenotypic expression of the monoallelic c.258+5G>T *NFKB1* variant in our CVID-affected proband and her mildly affected father, reflecting the incomplete penetrance of this *NFKB1* variant and the resulting autosomal-dominant disease. Importantly, like other gene defects with impact on the immune system control, *NFKB1* variants are characterized by highly variable penetrance (PMID 27365489, 29477724), thereby explaining the subtle symptoms in the proband's father and absence of serious infections, and autoimmune, autoinflammatory, and lymphoproliferative disorders.

Noteworthy, while the neonatal and early childhood onset of clinical symptoms has been reported in *NFKB1*-haploinsufficient patients, age-dependent severity of clinical phenotypes has also been observed.[20,23] Indeed, our patient manifested viral sepsis-like illness and meningitis along with hypogammaglobulinemia as a 5-week-old young infant, yet presently, aged 6 years, she fulfills clinical and immunological criteria of the pediatric CVID. The diagnosis has been made because of recurrent respiratory tract infections, low serum immunoglobulin levels, IgG subclass deficiency, poor response to vaccines, and defective B cell development with low switched-memory B cells, without evidence of profound deficiency in the T cell pool.[19,23] However, because of the variant in *NFKB1*, she is at risk of age-related progressive depletion of B cells and hence, impaired class switch recombination, somatic hypermutation, antibody affinity maturation, abrogation of T cell functions, and severe infectious episodes, as well as autoimmune, autoinflammatory, lymphoproliferative, and malignant complications.[8,20,23,28,30,32-36]

In this report, we expand the clinical phenotypic features of the *NFKB1* gene-related disease by adding a neonatal viral sepsis and meningitis because of an HPeV infection to the symptomatology of the NF-κB1 deficiency. HPeV, in particular HPeV3 types are the major cause of meningitis in neonates and young infants. The HPeV3 strains are the most pathogenic among HPeV types. They replicate rapidly in neuronal cells, suggesting their relationship with neuropathogenicity.[37,38] In neonates, HPeV elicits a specifically modulated innate immune response profile, with several inflammatory mediator elevated levels, such as interleukin (IL)-15, a proinflammatory cytokine important in eradication of viral pathogens, fractalkine CXCL1, interferon (IFN)-α2, and IFN-γ-induced protein 10 (IP) CXCL10. Simultaneously, no significant increase in IFN-γ levels or downstream type I interferon pathway effectors were noted.

This phenomenon of immune response to HPeV may be associated with its tissue tropism and limitation of the CNS infection to smooth muscle cells of leptomeningeal blood vessels.[39] This apparently deficient innate immune response may be assumed to be a contributory factor to prolonged HPeV infection or long-term viral persistence despite good short-term clinical neurological outcomes. In children in whom CNS parechoviral infection and sepsis-like symptoms do not require intensive care, the disease is often not notifiable. However, the study on a large cohort of HPeV-infected infants indicate the need for long-term clinical and imaging monitoring as short-term evaluation does not specifically predict the neurodevelopmental outcomes.[40] Whereas NF-κB1 canonical pathway activates the components of the innate immune response, such as TLRs, important questions are then raised about the role of the monoallelic *NFKB1* alterations and susceptibility to HPeV infection in our patient, and whether NF-κB deficiency is underpinning the less robust TLR sensitization, impaired adaptive immune response, and proneness to HPeV infection.

Importantly, regulation of NF-κB signaling pathways and its activation and controlling antiviral host defense is a ubiquitination-related process.[41,42] It may therefore be hypothesized that the ever-advancing knowledge on the regulatory mechanisms of NF-κB activity in virus-triggering inflammation will contribute to a better understanding

of the risk of viral evasion in CVID patients with *NFKB1* variants.

The patient's perspective refers to CVID-related infectious and noninfectious complications. Infections of the respiratory tract, e.g., pneumonia and bronchitis, and upper airway infections, such as otitis media, sinusitis, and tonsillitis, are the leading CVID sequelae in pediatric patients, followed by gastrointestinal infections. Affected individuals are also at risk of developing severe infectious episodes, such as sepsis, osteomyelitis, septic arthritis, or CNS infections.[23] Furthermore, the spectrum of immunophenotypes also include CVID-related immune dysregulation, and autoimmune, autoinflammatory, and lymphoproliferative disorders. Among them autoimmune cytopenias, such as AIH, autoimmune thrombocytopenia, and autoimmune neutropenia (AIN), are the most frequently observable complications associated with specific *NFKB1* gene variants.[43–46] It was also shown that the type of variants shapes the clinical phenotype in CVID, as more pronounced autoimmune and inflammatory disorders, such as enteropathy or bronchiectasis, occurred in patients bearing frameshift or nonsense *NFKB1* variants.[47] Regular Ig-RT supplying specific antibodies and providing both antimicrobial and immunomodulatory activities, reinforced by antibiotic prophylaxis on as needed basis, are the mainstay of the management strategy. Vaccination strategy should be elaborated individually. Live attenuated vaccines are not recommended in children with CVID and in patients receiving IgRT as they may become inactivated. Inactivated vaccines, including intramuscular influenza vaccine, can be safely administered in CVID but may not elicit a protective antigen-specific immune response.[48–50] The lifelong risk of CVID-related infectious and immune dysregulation disorders requires a multidisciplinary care of specialists in immunology, infectious diseases, neurology, pulmonology, oncology, and hematology.

Conclusion

By including viral meningitis and sepsis-like illness in a child with antibody deficiency, this comprehensive clinical-molecular study expands the spectrum of warning signs of IEI. The manuscript is therefore meant not only for specialists in clinical immunology and medical genetics but also has an educational aspect for clinicians facing diagnostic challenges. Hopefully, it will contribute to increased awareness and recognition of IEI in children with hypogammaglobulinemia affected with viral meningitis.

Along with the ever-increasing role of immunogenetics in disease definition and delineation, this case report shows that pediatric patients with infectious symptomatology and antibody deficiency should be offered genetic testing. It might be helpful in anticipating the prognosis and making the space for novel patient-tailored therapeutic approaches.

Ethics Approval

NA. This research was conducted retrospectively from data obtained for clinical purposes. Ethical approval was waived by the Bioethical Committee of Poznań University of Medical Sciences. In view of the case report and retrospective nature of the study, all the procedures being performed were part of the routine care and did not need ethical approval.

Informed Consent to Participate/Publication

An informed consent for publication of clinical data, genetic testing, and imaging was obtained from the patient's parent.

Availability of Data and Material/ Data Transparency

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

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Authors Contributions

Aleksandra Szczawińska-Popłonyk was responsible for the intellectual content of the mauscript, coordinated and supervised data collection, and drafted and critically revised the final manuscript. Natalia Popłonyk was responsible for writing the first draft of the manuscript. Katarzyna Bernat-Sitarz and Katarzyna Jończyk-Potoczna were responsible for the collection of clinical data and participated in writing the first draft of the manuscript. Agnieszka Pollak was responsible for genetic work-up and helped in critical revision of the manuscript. All authors read and approved the final manuscript.

Conflict of Interest

All authors declare no existing financial and nonfinancial conflict of interest regarding this manuscript.

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References

1. Barnabei L, Laplantine E, Mbongo W, Rieux-Laucat F, Weil R. NF-kappaB: At the borders of autoimmunity and inflammation. Front. Immunol. 2021 Aug;12:716469. https://doi.org/10.3389/fimmu.2021.716469

2. Schnappauf O and Aksentijevich I. Mendelian diseases of dysregulated canonical NF-κB signaling: From immunodeficiency to inflammation. J. Leukoc. Biol. 2020 Aug;108(2):573–589. https://doi.org/10.1002/JLB.2MR0520-166R
3. Liu T, Zhang L, Joo D, Sun SC. NF-κB signaling in inflammation. Signal Transd. Target Ther. 2017 Jul;2:17023. https://doi.org/10.1038/sigtrans.2017.23
4. Sun SC. The non-canonical NF-κB pathway in immunity and inflammation. Nat. Rev. Immunol. 2017 Sep;17(9):545–558. https://doi.org/10.1038/nri.2017.52
5. Napetschnig J and Wu H. Molecular basis of NF-κB signaling. Ann. Rev. Biophys. 2013 Mar;42:443–468. https://doi.org/10.1146/annurev-biophys-083012-130338
6. Gaspar Pereira S and Oakley F. Nuclear factor-kappaB1: Regulation and function. Int. J. Biochem. Cell Biol. 2008 May;40(8):1425–1430. https://doi.org/10.1016/j.biocel.2007.05.004
7. Liu P, Li Y, Wang W, Bai Y, Jia H, Yuan Z, et al. Role and mechanisms of the NF-κB signaling pathway in various developmental processes. Biomed. Biopharm. 2022 Sep;153:113513. https://doi.org/10.1016/j.biopha.2022.113513
8. Hayden MS and Ghosh S. NF-kappaB in immunobiology. Cell Res. 2011 Feb;21(2):223–244. https://doi.org/10.1038/cr.2011.13
9. Boztug H, Hirschmugl T, Holter W, Lakatos K, Kager L, Trapin D, et al. NF-κB1 haploinsufficiency causing immunodeficiency and EBV-driven lymphoproliferation. J. Clin. Immunol. 2016 Aug;36(6):533–540. https://doi.org/10.1007/s10875-016-0306-1
10. Guldenpfennig C, Teixeiro E, Daniels M. NF-kB's contribution to B cell fate decisions. Front Immunol. 2023 Jul;14:1214095. https://doi.org/10.3389/fimmu.2023.1214095
11. Durkee-Shock J, Zhang A, Liang H, Wright H, Magnusson J, Garabedian E, et al. Morbidity, mortality, and therapeutics in combined immunodeficiency: data from the USIDNET Registry. J. Allergy Clin. Immunol. Pract. 2022 May;10(5):1334–1341. https://doi.org/10.1016/j.jaip.2022.01.042
12. Dabbah-Krancher G and Snow AL. Mistuned NF-κB signaling in lymphocytes: Lessons from relevant inborn errors of immunity. Clin. Exp. Immunol. 2023 Apr;212(2):117–128. https://doi.org/10.1093/cei/uxad006
13. Tuijnenburg P, Lango Allen H, Burns SO, Greene D, Jansen MH, Staples E, et al. Loss-of-function nuclear factor κB subunit 1 (NFKB1) variants are the most common monogenic cause of common variable immunodeficiency in Europeans. J. Allergy Clin. Immunol. 2018 Oct;142(4):1285–1296. https://doi.org/10.1016/j.jaci.2018.01.039
14. Schroder C, Sogkas G, Fliegauf M, Dork T, Liu D, Hanitsch LG, et al. Late-onset antibody deficiency due to monoallelic alterations in *NFKB1*. Front. Immunol. 2019 Nov;10:2618. https://doi.org/10.3389/fimmu.2019.02618
15. Li J, Lei WT, Zhang P, Rapaport F, Seeleuthner Y, Lyu B, et al. Biochemically deleterious human *NFKB1* variants underlie an autosomal dominant form of common variable immunodeficiency. J. Exp. Med. 2021 Nov;218(11):20210566. https://doi.org/10.1084/jem.20210566
16. Fliegauf M, Kruger R, Steiner S, Hanitsch LG, Buchel S, Wahn V, et al. A pathogenic missense variant in NFKB1 causes common variable immunodeficiency due to detrimental protein damage. Front. Immunol. 2021 Apr;12:621503. https://doi.org/10.3389/fimmu.2021.621503
17. Kaustio M, Haapaniemi E, Goos H, Hautala T, Park G, Syrjanen J, et al. Damaging heterozygous mutations in NFKB1 lead to diverse clinical phenotypes. J. Allergy Clin. Immunol. 2017 Sep;140(3):782–796. https://doi.org/10.1016/j.jaci.2016.10.054
18. Shen Y, Boulton APR, Yellon RL, Cook MC. Skin manifestations of inborn errors of NF-κB. Front. Pediatr. 2023 Jan;10:1098426. https://doi.org/10.3389/fped.2022.1098426
19. Ameratunga R, Lehnert K, Woon ST. All patients with common variable immunodeficiency disorders (CVID) should be routinely offered diagnostic genetic testing. Front. Immunol. 2019 Nov;10:2678. https://doi.org/10.3389/fimmu.2019.02678
20. Seidel MG, Kindle G, Gathmann B, Quinti I, Buckland M, van Montfrans J, et al. The European Society for Immunodeficiencies (ESID) Registry working definitions for the clinical diagnosis of inborn errors of immunity. J. Allergy Immunol. Pract. 2019 Jul-Aug;7(6):1763–1770. https://doi.org/10.1016/j.jaip.2019.02.004
21. Bogaert DJA, Dullaers M, Lambrecht BN, Vermaelen KY, De Baere E, Haerynck F. Genes associated with common variable immunodeficiency: One diagnosis to rule them all? J. Med. Genet. 2016 Sep;53(9):575–590. https://doi.org/10.1136/jmedgenet-2015-103690
22. Cunningham-Rundles C, Casanova JL, Boisson B. Genetics and clinical phenotypes in common variable immunodeficiency. Front. Genet. 2024 Jan;14:1272912. https://doi.org/10.3389/fgene.2023.1272912
23. Lorenzini T, Fliegauf M, Klammer N, Frede N, Proietti M, Bulashevska A, et al. Characterization of the clinical and immunologic phenotype and management of 157 individuals with 56 distinct heterozygous NFKB1 mutations. J. Allergy Clin. Immunol. 2020 Oct;146(4):901–911. https://doi.org/10.1016/j.jaci.2019.11.051
24. Szczawińska-Popłonyk A, Schwartzmann E, Bukowska-Olech E, Biernat M, Gattner S, Korobacz S, et al. The pediatric common variable immunodeficiency - from genetics to therapy: A review. Eur. J. Pediatr. 2022 Apr;181(4):1371–1383. https://doi.org/10.1007/s00431-021-04287-6
25. Tuovinen EA, Kuismin O, Soikkonen L, Martelius T, Kaustio M, Hamalainen S, et al. Long-term follow up of families with pathogenic NFKB1 variants reveals incomplete penetrance and frequent inflammatory sequelae. Clin. Immunol. 2023 Jan;246:109181. https://doi.org/10.1016/j.clim.2022.109181
26. Fathi N, Nirouei M, Rizi ZS, Fekrvand S, Abolhassani H, Salami F, et al. Clinical, immunological, and genetic features in patients wit *NFKB1* and *NFKB2* mutations: A systematic review. J. Clin. Immunol. 2024 Jul;44(7):160. https://doi.org/10.1007/s10875-024-01763-0
27. Baxter SK, Walsh T, Casadei S, Eckert MM, Allenspach EJ, Hagin D, et al. Molecular diagnosis of childhood immune dysregulation, polyendocrinopathy, and enteropathy, and implications for clinical management. J. Allergy Clin. Immunol. 2022 Jan;149(1):327–339. https://doi.org/10.1016/j.jaci.2021.04.005
28. Staels F, De Keukeleere K, Kinnunen M, Keskitalo S, Lorenzetti F, Vanmeert M, et al. Common variable immunodeficiency in two kindreds with heterogeneous phenotypes caused by novel heterozygous NFKB1 mutations. Front. Immunol. 2022 Sep;13:973543. https://doi.org/10.3389/fimmu.2022.973543
29. Tangye SG, Al-Herz W, Bousfiha A, Cunningham-Rundles C, Franco JL, Holland SM, et al. Human inborn errors of immunity: 2022 Update on the classification from the International Union of Immunological Societies Expert Committee. J. Clin. Immunol. 2022 Oct;42(7):1473–1507. https://doi.org/10.1007/s10875-022-01289-3
30. Ramirez NJ, Posadas-Cantera S, Caballero-Oteyza A, Camacho-Ordonez N, Grimbacher B. There is no gene for CVID – novel monogenetic causes for primary antibody deficiency. Curr. Opin. Immunol. 2021 Oct;72:176–185. https://doi.org/10.1016/j.coi.2021.05.010
31. Szczawińska-Popłonyk A, Ciesielska W, Konarczak M, Opanowski J, Orska A, Julia Wróblewska, et al. Immunogenetic landscape in pediatric common variable immunodeficiency. Int J Mol Sci. 2024 Sep;25(18):9999. https://doi.org/10.3390/ijms25189999

32. Fliegauf M, Bryant VL, Frede N, Slade C, Woon ST, Lehnert K, et al. Haploinsufficiency of the NF-κB1 subunit p50 in common variable immunodeficiency. Am. J. Hum. Genet. 2015 Sep;97(3):389–403. https://doi.org/10.1016/j.ajhg.2015.07.008
33. Fekrvand S, Khanmohammadi S, Abolhassani H, Yazdani R. B- and T-cell subset abnormalities in monogenic common variable immunodeficiency. Front. Immunol. 2022 Jun;13:912826. https://doi.org/10.3389/fimmu.2022.912826
34. Rojas-Restrepo J, Caballero-Oteyza A, Huebscher K, Haberstroh H, Fliegauf M, Keller B, et al. Establishing the molecular diagnoses in a cohort of 291 patients with predominantly antibody deficiency by targeted next-generation sequencing: experience from a monocentric study. Front. Immunol. 2021 Dec;12:786516. https://doi.org/10.3389/fimmu.2021.786516
35. Mauracher AA, Gujer E, Bachmann LM, Gusewell S, Pachlopnik Schmid J. Patterns of immune dysregulation in primary immunodeficiencies: A systematic review. J. Allergy Clin. Immunol. Pract. 2021 Feb;9(2):792–802. https://doi.org/10.1016/j.jaip.2020.10.057
36. Fathi N, Mojtahedi H, Nasiri M, Abolhassani H, Marzbali MY, Esmaeili M, et al. How do nuclear factor kappa B (NF-κB)1 and NF-κB2 defects lead to the incidence of clinical and immunological manifestations of inborn errors of immunity? Expert Rev. Clin. Immunol. 2023 Mar;19(3):329–339. https://doi.org/10.1080/1744666X.2023.2174105
37. De Jong EP, van den Beuken MGA, van Elzakker EPM, Wolthers KC, Sprij AJ, Lopriore E, et al. Epidemiology of sepsis-like illness in young infants: Major role of enterovirus and human parechovirus. Pediatr. Infect. Dis. 2018 Feb;37(2):113–118. https://doi.org/10.1097/INF.0000000000001718
38. Autore G, Bernardi L, Perrone S, Esposito S. Update on viral infections involving the central nervous system in pediatric patients. Children. 2021 Sep;8(9):782. https://doi.org/10.3390/children8090782
39. Fortuna D, Cardenas AM, Graf EH, Harshyne LA, Hooper DC, Prosniak M, et al. Human parechovirus initiate distinct CNS innate immune responses: Pathogenic and diagnostic implications. J. Clin. Virol. 2017 Jan;86:39–45. https://doi.org/10.1016/j.jcv.2016.11.007
40. Joseph L, May M, Thomas M, Smerdon C, Tozen S, Bialasiewicz S, et al. Human parechovirus 3 in infants: Expanding our knowledge of adverse outcomes. Pediatr. Infect. Dis. 2019 Jan;38(1):1–5. https://doi.org/10.1097/INF.0000000000002136
41. Song K and Li S. The role of ubiquitination in NF-κB signaling during virus infection. Viruses. 2021 Jan;13(2):145. https://doi.org/10.3390/v13020145
42. Zheng Y and Gao C. Fine tuning of antiviral innate immunity by ubiquitination. Adv. Immunol. 2020 Dec;145:95–128. https://doi.org/10.1016/bs.ai.2019.11.004
43. Chawla S, Barman P, Tyagi R, Jindal AK, Sharma S, Rawat A, et al. Autoimmune cytopenias in common variable immunodeficiency are a diagnostic and therapeutic conundrum: an update. Front Immunol. 2022 Jun;13:869466. https://doi.org/10.3389/fimmu.2022.869466
44. Danandeh K, Jabbari P, Rayzan E, Zoghi S, Shahkarami S, Heredia RJ, et al. Case report of a novel NFκB mutation in a lymphoproliferative disorder patient. Endocr. Metab. Immune. Disord. Drug Targets. 2022;22(10):1040–1046. https://doi.org/10.2174/1871530322666220407091356
45. Szczawińska-Popłonyk A, Tąpolska-Jóźwiak K, Schwartzmann E, Popłonyk N. Immune dysregulation in pediatric common variable immunodeficiency: Implications for the diagnostic approach. Front Pediatr. 2022 Mar;10:855200. https://doi.org/10.3389/fped.2022.855200
46. Walter JE, Ayala IA, Milojevic D. Autoimmunity as a continuum in primary immunodeficiency. Curr Opin Pediatr. 2019 Dec;31(6):851–862. Doi: 10.1097/MOP.0000000000000833
47. Yin J, Hayes KM, Ong M, Mizgerd JP, Cunningham-Rundles C, Dominguez I, et al. Common variable immunodeficiency clinical manifestations are shaped by presence and type of heterozygous NFKB1 variants. J Allergy Clin Immunol Pract. 2024 Dec;s2213-2198(24)01242-X. https://doi.org/10.1016/j.jaip.2024.12.002
48. Principi N and Esposito S. Vaccine use in primary immunodeficiency disorders. Vaccine. 2014 Jun;32(30):3725–3731. https://doi.org/10.1016/j.vaccine.2014.05.022
49. Milito C, Soccodato V, Collalti G, Lanciarotta A, Bertozzi I, Rattazzi M, et al. Vaccination in PADs. Vaccines. 2021 Jun;9(6):626. https://doi.org/10.3390/vaccines9060626
50. Szczawińska-Popłonyk A, Bręborowicz A, Samara M, Ossowska L, Dworacki G. Impaired antigen-specific immune response to vaccines in children with antibody production defects. Clin. Vaccine Immunol. 2015;22:875–882. https://doi.org/10.1128/CVI.00148-15

Supplementry

Table S1 The infectious workup in the patient with NFκB1 deficiency at the diagnosis of the neonatal meningitis and sepsis-like illness.

Infectious workup							
Cerebrospinal fluid	Results	Blood	Results	Airways	Results	Urine	Results
Viral panel		Viral panel		Airway aspirate viral / bacterial panel			
CMV-DNA	(-)	CMV-DNA	(-)	Influenza virus A, A H1N1, B-RNA	(-)		
EBV-DNA	(-)	EBV-DNA	(-)	Rhinovirus-RNA	(-)		
ADV-DNA	(-)	ADV-DNA	(-)	Coronavirus NL63, 229E, OC43, HKU1-RNA	(-)		
HSV1-DNA	(-)	HSV1-DNA	(-)	Parainfluenza virus 1,2,3,4-RNA	(-)		
HSV2-DNA	(-)	HSV2-DNA	(-)	Human metapneumovirus A, B-RNA	(-)		
HHV6-DNA	(-)	HHV6-DNA	(-)	Respiratory syncytial virus A, B-RNA	(-)		
HHV7-DNA	(-)	HHV7-DNA	(-)	Enterovirus-RNA	(-)		
EV-RNA	(-)	EV-RNA	(-)	Parechovirus-RNA	(-)		
HPeV-RNA	(+)	HPeV-RNA	(-)	Bocavirus-DNA	(-)		
VZV-DNA	(-)	VZV-DNA	(-)	Mycoplasma pneumoniae-DNA	(-)		
HPVB19-DNA	(-)	HPVB19-DNA	(-)	Chlamydophila pneumoniae-DNA	(-)		
		HBV-DNA	(-)	Staphylococcus aureus-DNA	(-)		
		HCV-DNA	(-)	Streptococcus pneumoniae-DNA	(-)		
				Hemophilus influenzae-DNA	(-)		
Bacterial panel		Bacterial panel					
Streptococcus pneumoniae-DNA	(-)	Streptococcus pneumoniae-DNA	(-)				
Neisseria meningitidis-DNA	(-)	Neisseria meningitidis-DNA	(-)				
Hemophilus influenzae-DNA	(-)	Hemophilus influenzae-DNA	(-)				
CSF culture	(-)	Blood culture	(-)			Urine culture Proteus mirabilis	(+)