Research Square

Preprints are preliminary reports that have not undergone peer review.
They should not be considered conclusive, used to inform clinical practice,
or referenced by the media as validated information.

Neurologic Status of Patients With Purine Nucleoside Phosphorylase Deficiency Before and After Hematopoetic Stem Cell Transplantation

Betul Gemici Karaaslan
Isılay Turan
Sezin Aydemir
Zeynep Meric
Didem Atay
Arzu Akcay
Aysun Ayaz Sari
Michael Hershfield
Funda Cipe
Basak Adakli Aksoy
Gizem Zengin Ersoy
Ceyhun Bozkurt
Yasemin Kendir Demirkol
Gulyuz Ozturk
Cigdem Aydogmus
Ayca Kiykim (ayca.kiykim@iuc.edu.tr)
Istanbul University-Cerrahpasa, Cerrahpasa School of Medicine https://orcid.org/0000-0001-5821-3963
Haluk Cokugras

Research Article

Keywords: purine nucleoside phosphorylase deficiency, neuromotor development, hematopoietic stem cell transplantation, combined immune deficiency

Posted Date: July 18th, 2023

DOI: https://doi.org/10.21203/rs.3.rs-3153599/v1

License: This work is licensed under a Creative Commons Attribution 4.0 International License. Read Full License

Version of Record: A version of this preprint was published at Journal of Clinical Immunology on September 20th, 2023. See the published version at https://doi.org/10.1007/s10875-023-01585-6.

Abstract

Background

Purine nucleoside phosphorylase (PNP) deficiency is a rare autosomal recessive combined immunodeficiency. The phenotype is profound T-cell deficiency with variable B- and NK-cell functions and results in recurrent and persistent infections that typically begin in the first year of life. Neurologic findings occur in approximately two-thirds of patients. The mechanism of neurologic abnormalities is unclear. Hematopoietic stem cell transplantation (HSCT) is the only curative treatment for PNP deficiency.

Methods

We report here six patients from five unrelated families with PNP deficiency treated in two centers in Turkey. We evaluated the neurological status of patients and compared to post-transplantation period if available. Then we performed PubMed, Google Scholar, and Researchgate searches using the terms "PNP" and "hematopoietic stem cell transplantation" to find all reported cases of PNP transplantation and compared to our cohort.

Results

Six patients were treated in two centers in Turkey. One patient died from post-transplant complications. The other four patients underwent successful HSCT with good immune reconstitution after transplantation (follow-up 21–48 months) and good neurological outcomes. The other patient with a new mutation is still waiting for a matching HLA donor.

Discussion

In PNP deficiency, clinical manifestations are variable, and this disease should be considered in the presence of many different clinical findings. Despite the comorbidities that occurred before transplantation, HSCT currently appears to be the only treatment option for this disease. HSCT not only cures immunologic disorders, but probably also improves or at least stabilizes the neurologic status of patients.

Introduction

Purine nucleoside phosphorylase (PNP) is an enzyme of the purine salvage pathway. In purine metabolism, PNP catalyzes the phosphorylation of inosine, guanosine, and deoxy nucleosides to hypoxanthine and guanine, respectively. The metabolites are detoxified or converted back to the nucleotide. In different tissues, the PNP enzyme is expressed to varying degrees; the highest expression is found in lymphoid tissue. The accumulation of toxic metabolites (especially deoxyguanosine to deoxyguanosine triphosphate (dGTP)) is toxic to T cells in lymphoid tissue. Accumulation of dGTP leads to inhibition of ribonucleotide reductase and blocks DNA synthesis, proliferation, and repair mechanisms[1,2]. In addition, double-positive thymocytes are more susceptible to activation-induced apoptosis because these accumulations disrupt mitochondrial membrane potential[3].

PNP deficiency is a rare autosomal recessive combined immunodeficiency. The phenotype is profound T-cell deficiency with variable B- and NK-cell functions and results in recurrent and persistent infections that typically begin in the first year of life. Patients account for approximately 4% of all patients with severe combined immunodeficiency (SCID)[4]. Patients present with autoimmune manifestations, neutropenia, neurologic abnormalities, or infections resembling those of SCID or combined immunodeficiency[5–13]. Neurologic manifestations may precede infectious and autoimmune complications[14].

Approximately two-thirds of patients present with neurologic findings; in addition, one-third of patients have autoimmune manifestations.

The mechanism of neurologic abnormalities such as ataxia, hyper-/hypotonia, tremors, spasticity, and varying degrees of developmental delay is unclear. Although toxic purines impair neurological functions, neurotoxicity cannot explain every abnormality. GTP is essential for neurotransmission in the central nervous system; low GTP levels may be the main reason for the neurologic findings[14,15].

Autoimmune manifestations often include autoimmune hemolytic anemia (AIHA) and immune thrombocytopenia (ITP)[4,13,14,16]. Hematologic malignancies have also been reported in PNP patients[17,18].

Low plasma uric acid level has been suggested as a marker for diagnosis, but Walker et all. found normal uric acid levels in five patients. Therefore, this marker is deceptive[19]. Diagnosis can be made by measuring PNP enzyme activity or by genetic analysis of the PNP gene.

Hematopoietic stem cell transplantation (HSCT) is the only curative treatment for PNP deficiency. HSCT not only improves immunologic impairment but also stabilizes or even improves neurologic findings[7,10,12,20,21].

We report here six patients from five unrelated families with PNP deficiency treated in two centers in Turkey.

Methods

Patient data

We reviewed the medical records of five patients with PNP deficiency and prospectively screened them using the Denver Developmental Screening Test (DDST), which suggests age-appropriate milestones. Patient data included medical history, transplant history, posttransplant clinical course, and immunologic parameters collected at baseline until the patients' last review.

Literature Review

We performed PubMed, Google Scholar, and Researchgate searches using the terms "PNP" and "hematopoietic stem cell transplantation" to find all reported cases of PNP transplantation.

Results

Patients

Patient 1 (P1) was born to consanguineous parents. He had a sister who died of a seizure of unknown cause at the age of 3 years. His medical history showed treatment-resistant oral candidiasis, recurrent upper respiratory tract infections, lymphopenia, neutropenia, thrombocytopenia, and autoimmune hemolytic anemia (AIHA), which was treated with intravenous immunoglobulin (IVIG) and systemic steroids. After his first year of life, he required multiple hospitalizations for sepsis and upper respiratory tract infections. The thorax computer tomography (CT) showed a mosaic pattern and ground-glass infiltrates. Bone marrow biopsy revealed erythroid hyperplasia, normoblastic maturation, and a relative decrease in myeloid series. At 18 months of age, he underwent immunologic evaluation, which revealed failure to thrive and global developmental delays. Computed tomography of the skull (CT) showed only a cavum septum pellucidum and a vergae variant. On examination, growth parameters were below the third percentile. He had hepatosplenomegaly and lymphadenopathy. A neurological examination revealed global developmental delay and mild truncal hypotonia. On

immunologic examination, immunoglobulin levels were normal except for a low IgA level (but the patient had been treated with IVIG several times); his lymphocyte subset analyses revealed a T-B-NK + phenotype, and uric acid levels were very low (1.3 mg/dl), suggesting PNP deficiency (Table 1). Autoantibodies, including ANA, anti- GAD, and direct Coombs were positive. Homozygous mutation c.349G > A (p.Ala117Thr) was detected by genetic sequencing of the PNP gene. While awaiting a suitable HLA donor, the patient received prophylactic trimethoprim-sulfamethoxazole (TMP-SMZ), fluconazole, acyclovir, isoniazid (due to BCG vaccination before diagnosis), and immunoglobulin replacement therapy. During the follow-up period, the patient developed vision loss in both eyes. CMV retinitis was diagnosed, and the left eye was treated with intravitreal ganciclovir injection; unfortunately, treatment could not be given because of complete vision loss in the right eye. CMV viremia also developed, which was treated with systemic ganciclovir. Liver biopsy was performed to explain the elevated liver function test, and the result was normal. At the time of HSCT, he was 38 months old and could not yet speak or walk but could sit without support. The patient underwent HSCT from a matched family donor (healthy sister). His conditioning regimen consisted of treosulfan, fludarabine. Mesenchymal stem cells were given at day − 1 to provide engraftment. The patient was transplanted with an unmanipulated bone marrow graft. For prophylaxis of graft-versus-host disease (GVHD), he received cyclosporine A and mycophenolate mofetil (MMF). Neutrophil engrafment was achieved at day + 15 and thrombocyte engraftment was achieved at day + 34. At the first, sixth, and twelfth months after HSCT, he had complete donor chimerism; he was also free of GVHD. After HSCT, cranial examination CT showed cerebrocerebellar atrophy and minimal ventriculomegaly. Currently, he is 6 years and 2 months old and has low IgM and normal IgG levels without infections; leukopenia and anemia have also improved (Table 1). Neurological assessment with DDST is delayed, but improvement is noted (Table 2).

Table 1
Pre-transplantation and Post-transplantation Immune Work-up

	P1		P2		P3	P4		P5		P6
	Pre HSCT	Post HSCT	Pre HSCT	Post HSCT	Pre HSCT	Pre HSCT	Post HSCT	Pre HSCT	Post HSCT	Pre HSCT
Age(months)	22	71	14	69	43	6	35	6	40	12
Neutrophils/mm3	500	2620	2390	2210	2900	2560	2500	1910	2700	1500
Lymphocytes/mm3	600	3330	330	5090	1100	1400	4470	600	2800	500
% CD3+	N/A	83	23,7	75,12	23	41,9	83.04	54	70	45,8
% CD3 + CD4+	N/A	40,1	7,5	44,21	58	11,6	34.83	78	36	34,3
% CD3 + CD8+	N/A	42,8	16	49,14	23	21,8	58.55	10	21	10,4
% CD19+	N/A	11,4	6,2	12,66	47	30,5	14.63	6	12,2	16,8
% CD3-CD16 + 56+	N/A	N/A	70,1	32,62	21	N/A	4.68	N/A	16	36,2
Recent Thymic Emigrant (%)	N/A	N/A	N/A	59,3	46	N/A	64.15	53	58,2	1
IgM (mg/dl)	101	47	105	98	N/A	48	< 0,01	34	53	48,1
IgA (mg/dl)	44,4	55	313	124	155,4	17	9	14	116	15,4
IgG (mg/dl)	492	748	1869	766	N/A	438	742	340	985	346
Total IgE (kU/L)	2.67	< 0,2	1,78	N/A	< 16,5	375,6	2,35	6	379	6,09
Uric Acid (mg/dL)	1.3	3,6	2,4	5,1	1,7	1,7	3,09	0.1	4	0,1
PNP analyze	N/A		N/A		N/A	14,8		58,5	N/A	6,9
Autoantibody Status	ANA(+) D. Coombs(+) AntiGAD (+)		Anti-dsDNA(-) Anti TTG(-) D.Coombs(-)		N/A	D.Coombs(-)		???		D.Coombs(-)

Table 2
Pretransplantation and Posttransplantation Neurological Development

	Neurological Development	
	Pretransplantation	**Posttransplantation**
P1	**18 months:** **• Global developmental delay** **• mild truncal hypotonia** **38 months:** **• unable to speak** **• unable to walk** **• sit without support**	**74 months:** **• speak with articulation problems** **• walk and run with mild ataxia.** **DDST:** **• Social and Emotional: 36 months** **• Language/Communication:51 months** **• Gross motor skills: 33 months** **• Minor motor skills: 51 months**
P2	**12 months**: • gross motor delay (only hold head) • gross delay in communication and language (no speech)	**73 months**: • walk with mild ataxia from 30 months • speech is appropriate 45 months of age. **DDST**: • Social and Emotional: 30 months • Language/Communication: 45 months • Gross motor skills: 18 months • Minor motor skills: 51 months
P4	**6 months**: • mild hypotonia • hold head • sit with support	**37 months** • Walk without support • Speak several single words **DDST** • Social and Emotional: 24 months • Language/Communication: 24 months • Gross motor skills: 18 months • Minor motor skills: 22 months

P5	**3 months**: • hold head steady, unsupported **6 months**: • roll over from tummy to back	**60 months** • Walk without support • Speak several single words **DDST** • Social and Emotional: 36 months • Language/Communication: 15 months • Gross motor skills: 27 months • Minor motor skills: 16 months

Patient 2 (P2) was born to consanguineous parents The family history was significant for another sibling who had died of sepsis at 18 months of age. He presented at 12 months of age with severe gastroenteritis requiring hospitalization and was referred to immunology for treatment of resistant diarrhea. Neurologically, he had significant motor delay and delayed speech development. Immunologic examination revealed normal immunoglobulin levels, positive vaccine-specific antibodies, and T- and B-cell lymphopenia (T- B- NK + phenotype). Uric acid levels were low (2.5 mg/dl). Autoantibodies included antibodies to gliadin and tissue transglutaminase, and the direct Coombs test was negative. Clinical exome sequencing (CES) revealed a homozygous mutation of the PNP gene (deletion of exon 3–5). Before HSCT, he received prophylaxis with TMP-SMZ, fluconazole, isoniazid, and immunoglobulin replacement therapy. CMV retinitis developed and was treated with systemic ganciclovir. The patient underwent HSCT from a HLA full matched sibling donor at 18 months of age using treosulfan, fludarabine. Mesenchymal stem cells were given at day − 1 to provide engraftment. Cyclosporine A and MMF were used for GVHD prophylaxis. Neutrophil engrafment was achieved at day + 13 and thrombocyte engraftment was achieved at day + 27. Immediately after transplantation, the patient suffered from ileus, which was successfully treated. The patient's chimerism on days 40, 42, 61, and 65 showed mixed chimerism (72% at day 40 and 53% at day 65). On day 79, the patient received an infusion of donor lymphocytes. On day 99, he had complete donor chimerism. After HSCT, he underwent bone osteotomy and fixation for a slipped femoral epiphysis. Today, 6 years and 1 month after transplantation, he is doing well, has complete donor chimerism, is free of infection, and has normal immunoglobulin levels (Table 1). In terms of development, he has not been able to catch up with his delay but began to walk unassisted at 30 months of age, although he has mild ataxia. His speech is at the developmental level of 3 years and 9 months. However, his development improves after HSCT (Table 2).

Patient 3 (P3) was born of consanguineous parents. The patient presented with recurrent respiratory infections including tuberculosis and severe developmental delays in all areas at the age of 4 years. Neurological examination revealed severe mental retardation, ataxia, dysarthria, dystonia, and gross motor delay. Head holding, sitting without support, and walking without support were delayed at 6, 12, and 24 months, respectively. Cranial examination CT was normal. Thoracic examination CT revealed bilateral acino-nodular infiltration and lymphadenopathy. On immunologic examination, lymphocyte subset analysis showed T- and B-cell lymphopenia. Genetic analysis revealed a homozygous missense mutation of the PNP gene. The patient underwent bone marrow transplantation with a full matched unrelated donor at the age of 60 months but unfortunately died on day 14 due to respiratory infection.

Patient 4 (P4) was born to consanguineous parents with meconium aspiration syndrome that required hospitalization for the first 7 days. The patient's brother (P3) had died after a diagnosis of PNP deficiency. At 6 months of age, he presented with upper extremity cramps, wheezing, eczema, food allergy, and vomiting. Physical examination was normal except for mild hypotension. Immunologic examination revealed normal IgG, IgM, and IgA levels, high IgE, T-cell, and B-cell lymphopenia, and a low uric acid level (1.7 mg/dl). Skin prick tests with cow's milk, egg white, and egg yolk were also positive (Table 1). PNP enzyme assay revealed low PNP levels (14.8 nmol/h/mg). Genetic testing revealed a homozygous missense mutation of the PNP gene (p.R67P(c.200G > C)). Prophylaxis was started with TMP-SMZ, fluconazole,

ganciclovir, and immunoglobulin replacement therapy. The patient underwent bone marrow transplantation at 16 months of age with a fully matched male unrelated donor. He received a conditioning regimen of treosulfan and fludarabine. GVHD prophylaxis consisted of cyclosporine A and MMF. Neutrophil engrafment was achieved at day + 13 and thrombocyte engraftment was achieved at day + 21. Chimerism analysis revealed 100% donor chimerism in all cells (at days 28 and 99). 5 months after HSCT, the patient developed cervical and mesenteric multiple lymphadenopathies and EBV viremia (copy number: 68521). A lymph node biopsy was consistent with posttransplant lymphoproliferative disease. The patient was treated successfully with acyclovir and rituximab (3 doses every week). During follow-up, chimerism had decreased to 32% 12 months after HSCT. The PNP enzyme test was re-evaluated, and PNP activity was less than 10% of the mean of controls, although the patient was infection-free. In addition, immunologic examination revealed a normal IgG level, low IgA and IgM levels, and a normal lymphocyte count despite an impaired CD4/CD8 ratio (Table 1). His neurologic development showed no further deterioration. At 37 months of age, his gross and fine motor skills were equivalent to what would be expected at 18 and 22 months, respectively. His cognitive, social-emotional, and language abilities were similar to those of a 2-year-old child. The assessment concluded that he had clearly reached new developmental milestones.

Patient 5 (P5) was born to non-consanguineous parents with a normal perinatal period. At 4 months of age, she presented with failure to thrive, mild neutropenia, and severe lymphopenia without infection. Neuromotor development was largely normal (holding her head without support at 3 months and rolling from her abdomen to her back). Because leukopenia persisted, a bone marrow aspiration was performed, which was normal. At 9 months of age, a very low uric acid level was noted (0.1 mg/dl), and a PNP enzyme test was performed; enzyme levels were low and dAXP was not detected. Genetic analysis revealed that the patient was compound heterozygous for a novel mutation and a described mutation in the PNP gene (c.389T > G (p.Met130Arg) and c.265G > A (p.Glu89Lys)). While she was waiting for a suitable HLA donor, DDST results at 8 months of age were adequate for her age, except for gross motor skills, which were at 6 months. At 12 months of age, she received a bone marrow transplant with 5/10 HLA loci from an haploidentical related donor. GVHD prophylaxis consisted of cyclosporine A. At day 28, chimerism was 100%. Within a few months, low donor chimerism (63%) and low PNP activity were detected. At month 6 after transplantation, the patient received donor lymphocyte infusion (DLI) three times due to low chimerism and PNP activity. Following DLI, diarrhea and elevated liver enzymes were evaluated as GVHD and treated with steroids. at 18 months after HSCT, chimerism was 96%. Today, she is 5 years old and has a normal immunologic workup, including immunoglobulin and lymphocyte counts, subgroup analyses, and proliferation (Table 1). Neurologic assessment with DDST is delayed, and her gross and fine motor skills are what would be expected at 27 and 16 months, respectively. Their cognitive, social, and emotional abilities are equivalent to those of a 3-year-old child. Language skills are appropriately identified at 15 months. The assessment concluded that she has clearly reached new milestones in her development.

Patient 6 (P6) was born to consanguineous parents. She was healthy until she was 12 months old, when she was referred to a pediatric unit for cough and severe lymphopenia and was found to have decreased IgG levels. Neurologically, she had significant motor delay but normal language development: at 10 months, she was sitting with support, and at 1 year, she was speaking two words. The immunological examination revealed T-B-NK + lymphopenia, low IgG, and low uric acid levels (0.1 mg/dl). PNP enzyme assay was evaluated, and PNP enzyme levels were low, ADA activity was normal, and dAXP was undetectable (Table 1). Genetic analysis revealed that the patient was homozygous for a novel mutation in the PNP gene ((c.59_60delACinsCT (p.His20Pro). The disruption of this protein causing PNP deficiency has been reported previously.11 Unfortunately, there was no suitable HLA donor. While waiting for HSCT, the patient received prophylactic TMP-SMZ, fluconazole, ganciclovir, and immunoglobulin replacement therapy. She is currently 21 months old and waiting for a matching HLA donor. On neurological examination, she speaks two-word sentences, and her motor skills are developing in accordance with her age, although she does not walk without assistance.

Literature Review

In the literature review, the 20 case reports with HSCT for PNP deficiency were described in Table 3. A total of 3/20 patients had neutropenia, malignancy and autoimmunity, especially autoimmune cytopenia. The median age at transplantation was 40 months (range 6.5–168 months). Survival was 85%, with two patients dying immediately after transplantation from infectious complications and the other 14 days after HSCT from veno-occlusive disease. All surviving patients are doing well; median follow-up after transplantation is 25 months (8-108 months). Two patients still required IVIG treatment. 4,7 Most patients have chimerism of 90% or greater, and one patient with mixed chimerism was 60% clinically and immunologically adequate[10].

Neurologic sequelae are relatively mild; two patients were reported to have no improvement in development[16,22], whereas in the others, it was reported that " assessment at this age concluded that the patient had reached new developmental milestones" Therefore, neurologic improvement in development was noted in most patients after transplantation.

Discussion

We described six patients with PNP deficiency from four different families in Turkey. The mutations found in our five patients were described in previous reports. P5 was compound heterozygous, one is novel mutation (c.389T > G (p.Met130Arg)). P6 was homozygous for a novel mutation in the PNP gene (c.59_60delACinsCT (p.His20Pro)), and her phenotype was mild; without severe infections and neurological development was almost normal. (Fig. 1) Despite their identical genotype, P3 and P4 had different presentations; P3 presented later and was diagnosed at 5 years of age. At the time of diagnosis, he showed recurrent respiratory infections, including M. tuberculosis, gross motor developmental delay, and was dystonic. Unfortunately, he died on day 14 after transplantation due to respiratory infection. P4 presented earlier and was diagnosed after his brother's diagnosis. Probably, the early diagnosis was an advantage for him. He underwent HSCT after 12 months. Despite the decline in donor chimerism, the patient is infection-free and has developed new developmental skills. The two cases presented here may provide evidence that neurologic involvement may increase over the years and that early HSCT may prevent this. Although there are no systematic studies of genotype/phenotype correlation in PNP deficiency, we hypothesize that the clinical spectrum of PNP deficiency is variable and not related to genotype. Clinical variability ranges from SCID phenotype to the presence of neurologic symptoms only. Typically, patients with PNP deficiency have severe T-cell defects causing bacterial, viral, and fungal infections, but also variable B and NK defects with/without decreased serum immunoglobulin levels[17]. In our cohort, three patients (P4, P5, and P6) were diagnosed in the first year of life and were infection-free or had mild infections. The others had severe bacterial, viral, and fungal infections, such as CMV viremia, recurrent respiratory infections, and oral candidiasis in later life. However, immunological examination revealed T and B cell deficiency with variable serum immunoglobulin levels in all our patients. Although immunoglobulin levels could be normal, in three patients both B and T cells were affected by similar dysfunction as in previous studies[14].

Purine metabolism is essential for all cells and the cellular function can be ensured by a proper functioning purine-salvage pathway. PNP is a ubiquitous enzyme that provides purine homeostasis[1,2]. In addition to immunological dysfunction, the occurrence of diseases of other tissues in PNP deficiency is not surprising. Besides recurrent infections autoimmune manifestations or malignancy (especially EBV-related malignancy) may be seen[13–18]. Autoimmune cytopenia such as AIHA and ITP are the most common autoimmune manifestations[4,13,14,16]. Autoimmune mechanisms can also be a cause of neutropenia[6]. In our cohort, only one patient (P1) had autoimmune manifestations (AIHA, ITP), and neutropenia was documented in P1 and P6.

Neurologic dysfunction, which occurs in two-thirds of patients (66%), may be helpful in diagnosing the disease[26]. The mechanism of neurologic disease in PNP deficiency is not well understood; PNP deficiency results in an accumulation of deoxyguanosine. This accumulation is catalyzed by deoxyguanosine kinase (mitochondrial enzyme) or deoxycytidine kinase (nuclear enzyme) to dGTP. dGTP inhibits DNA synthesis, proliferation, and repair mechanisms[15]. In addition, this

molecule induces apoptosis and causes cellular toxicity.27 Even if immunologic disorders are explained by lymphotoxicity, neurotoxicity cannot explain all neurologic abnormalities. GTP is essential for neurotransmission in the central nervous system, and low GTP levels may be the main reason for the neurologic findings[14,15]. Moreover, de novo synthesis of GTP in the brain is limited, supporting this mechanism[20]. In a previous study, PNP-deficient mice were found to have a small cerebellum, corpus callosum, and thalamus[28]. Following this, the recent studies reported abnormal MRI results; one patient had significant neurologic impairment with atrophy of the cerebellum and cerebral cortex with loss of white matter myelination[29], and two other patients had brain atrophy[5,30]. However, Torun et al described two patients with normal MRI images[14]. In all our patients, neurologic symptoms were mild or marked. In particular, late presentation (P1, P2, P3) was associated with more advanced neurological dysfunction. In addition, P1 suffered a cardiopulmonary arrest requiring resuscitation during HSCT, and a cranial CT examination revealed cerebrocerebellar atrophy and minimal ventriculomegaly. Cranial MRI examination of P4 after transplantation revealed normal findings.

A low plasma uric acid level has been suggested as a marker for diagnosis, but Walker et all found normal uric acid levels in five patients; therefore, this marker is deceptive[19,26]. PNP enzyme activity or genetic analysis of the PNP gene are the gold standard diagnostic tests[4].

In PNP deficiency, the neonatal screening program fails in early diagnosis because TREC and KREC levels may be normal in infancy. Immunologic injury is caused by the gradual accumulation of toxic metabolites. La Marca et al described a spectrometric method for early diagnosis of dried blood spots to identify PNP metabolites[31].

Almost all patients with PNP deficiency died in the second decade of life from infectious complications. Only one patient reported surviving without HSCT at the age of 21 years. The baby of the PNP carrier was born after her pregnancy under intensive observation[32]. Despite the comorbidities that occurred before transplantation, HSCT currently appears to be the only treatment option for this disease. Moreover, the success of HSCT was reported to be 85% in this study. Another disease is deficiency of another enzyme in the purine rescue pathway, adenosine deaminase, and the survival rate is 80% after transplantation[33]. In our cohort, P3 died after HSCT on day 14 because of respiratory infection. The other four patients survived after transplantation and were free of infection despite posttransplant complications.

Neurological improvement after transplantation is difficult to assess. Because new developmental milestones are usually achieved over time and patients are relatively young at the time of transplantation, it is difficult to determine what neurologic progress they would have made if they had not been transplanted[14,20]. It is unclear whether HSCT is the cause of neurologic improvement; however, it appears that most patients do not have new neurologic impairments after transplantation. After transplantation, neurologic status may improve in some patients, whereas in others, further neurologic impairment may be arrested. Carpenter et al and Hallet et al reported that "no new developmental milestones were achieved"[16,22]. However, our review found that 19 of the posttransplant patients who reported their neurologic status after HSCT achieved new developmental milestones with good immune reconstitution (Table 3). A previous study also reported that Purkinje cell layer can be corrected by early treatment in mouse models[28].

In the review, some researchers reported that early treatment better prevents neurological complications. Early diagnosis and treatment play a critical role in preventing neurologic damage and other complications[14,28]. In our cohort, late-diagnosed patients had severe neurologic findings, whereas early-diagnosed patients had mild findings. Nevertheless, some neurologic deficits remain after both groups reached new developmental milestones after HSCT. In addition, some studies reported that patients' neurological status did not improve in the first year after transplantation, but they clearly reached new developmental milestones at repeated follow-up[13,21]. Consequently, HSCT probably improves or at least stabilizes patients' neurological status.

Declarations

Funding

This work was supported by the Research Fund of Istanbul University-Cerrahpasa (Project number: TOA-2021-35251).

Conflict of Interest

None of the authors has any potential financial conflict of interest related to this manuscript.

Availability of data and material

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

Authorship Contributions

All authors contributed to the study conception and design. BGK, IT, SA, ZM, DA, AA, AAS, YKD, MH, FC, BAA, GZE, CB, GO, CA, AK, HC followed patients, collected samples and provided clinical data. Material preparation, and data analysis were performed by BGK, IT and AK. The first draft of the manuscript was written by BGK and AK all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

Ethics approval

This study was performed in line with the principles of the Declaration of Helsinki. Approval was granted by the Ethics Committee of Istanbul University-Cerrahpasa (06.07.2023-727977).

Consent To Participate

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

Consent for Publication

The authors affirm that human research participants provided informed consent for publication of the patients' data.

References

1. Carson DA, Lakow E, Wasson DB, Kamatani N. Lymphocyte dys- function caused by deficiencies in purine metabolism. Immunol Today. 1981;2:234–8.
2. Giblett, E.R., Ammann, A.J., Wara, D.W., Sandman, R., Diamond, L.K., 1975. Nucleoside- phosphorylase deficiency in a child with severely defective T-cell immunity and normal B-cell immunity. Lancet 1, 1010–1013.
3. Papinazath T, Min W, Sujiththa S, Cohen A. Effects of purine nu- cleoside phosphorylase deficiency on thymocyte development. J Allergy Clin Immunol. 2011;128:854-63.
4. Schejter YD, Even-Or E, Shadur B, NaserEddin A, Stepensky P, Zaidman I. The Broad Clinical Spectrum and Transplant Results of PNP Deficiency. J Clin Immunol. 2020 Jan;40(1):123-130.
5. Madkaikar MR, Kulkarni S, Utage P, Fairbanks L, Ghosh K, Marinaki A, et al. Purine nucleoside phosphorylase deficiency with a novel PNP gene mutation: a first case report from India. BMJ Case Rep. 2011;2011:bcr0920114804 bcr0920114804.
6. Alangari A, Al-Harbi A, Al-Ghonaium A, Santisteban I, Hershfield M. Purine nucleoside phosphorylase deficiency in two unrelated Saudi patients. Ann Saudi Med 2019;29:309–12.
7. Aytekin C, Yuksek M, Dogu F, Yagmurlu A, Yildiran A, Fitoz S, et al. An unconditioned bone marrow transplantation in a child with purine nucleoside phosphorylase deficiency and its unique compli- cation. Pediatr Transplant.

2008;12:479–82.

8. Broome CB, Graham ML, Saulsbury FT, Hershfield MS, Buckley RH. Correction of purine nucleoside phosphorylase deficiency by transplantation of allogeneic bone marrow from a sibling. J Pediatr. 1996;128:373–6.
9. Celmeli F, Turkkahraman D, Uygun V, la Marca G, Hershfield M, Yesilipek A. A successful unrelated peripheral blood stem cell transplantation with reduced intensity-conditioning regimen in a patient with late-onset purine nucleoside phosphorylase deficiency. Pediatr Transplant.
10. Singh V. Cross correction following haemopoietic stem cell trans- plant for purine nucleoside phosphorylase deficiency: engrafted donor-derived white blood cells provide enzyme to residual enzyme-deficient recipient cells. JIMD Rep. 2012. p. 39–42.
11. Yeates L, Slatter MA, Gennery AR. Infusion of sibling marrow in a patient with purine nucleoside phosphorylase deficiency leads to split mixed donor chimerism and normal immunity. Front Pediatr. 2017;5:143.
12. Myers LA, Hershfield MS, Neale WT, Escolar M, Kurtzberg J. Purine nucleoside phosphorylase deficiency (PNP-def) presenting with lymphopenia and developmental delay: successful correction with umbilical cord blood transplantation. J Pediatr. 2004;145:710–2.
13. Delicou S, Kitra-Roussou V, Peristeri J, Goussetis E, Vessalas G, Rigatou E, et al. Successful HLA-identical hematopoietic stem cell transplantation in a patient with purine nucleoside phosphorylase deficiency. Pediatr Transplant. 2007;11:799–803
14. Torun B, Bilgin A, Orhan D, Gocmen R, Kılıc SS, Kuskonmaz B, Cetinkaya D, Tezcan I, Cagdas D. Combined immunodeficiency due to purine nucleoside phosphorylase deficiency: Outcome of three patients. Eur J Med Genet. 2022 Mar;65(3):104428.
15. Simmonds HA, Fairbanks LD, Morris GS, Morgan G, Watson AR, Timms P, et al. Central nervous system dysfunction and erythrocyte guanosine triphosphate depletion in purine nucleoside phosphory- lase deficiency. Arch Dis Child. 1987;62:385–91.
16. Carpenter PA, Ziegler JB, Vowels MR. Late diagnosis and correction of purine nucleoside phosphorylase deficiency with allogeneic bone marrow transplantation. Bone Marrow Transplant. 1996 Jan;17(1):121-4.
17. Markert, M.L., Finkel, B.D., McLaughlin, T.M., et al., 1997. Mutations in purine nucleoside phosphorylase deficiency. Hum. Mutat. 9 (2), 118–121.
18. Pannicke U, Tuchschmid P, Friedrich W, Bartram CR, \. Two novel missense and frameshift mutations in exons 5 and 6 of the purine nucleoside phosphorylase (PNP) gene in a severe combined immu- nodeficiency (SCID) patient. Hum Genet [Internet]. 1996;98:706– 9.
19. Walker, P.L., Corrigan, A., Arenas, M., Escuredo, E., Fairbanks, L., Marinaki, A., 2011. Purine nucleoside phosphorylase deficiency: a mutation update. Nucleos Nucleot. Nucleic Acids 30, 1243–1247.
20. Brodszki N, Svensson M, van Kuilenburg ABP, Meijer J, Zoetekouw L, Truedsson L, et al. Novel genetic mutations in the first Swedish patient with purine nucleoside phosphorylase defi- ciency and clinical outcome after hematopoietic stem cell trans- plantation with HLA-matched unrelated donor. JIMD Rep [Internet]. 2015;24:83–9.
21. Classen CF, Schulz AS, Sigl-Kraetzig M, Hoffmann GF, Simmonds HA, Fairbanks L, et al. Successful HLA-identical bone marrow transplantation in a patient with PNP deficiency using busulfan and fludarabine for conditioning. Bone Marrow Transplant [Internet]. 2001;28:93–6.
22. Hallett RJ, Gaspar B, Duley JA, et al. Allogeneic bone marrow transplantation corrects the immunodeficiency in PNP deficiency but does not reverse the neurological abnormalities. Cell Mol Biol Lett. 1999;4:374.
23. Baguette C, Vermylen C, Brichard B, Louis J, Dahan K, Vincent MF, et al. Persistent developmental delay despite successful bone marrow transplantation for purine nucleoside phosphorylase defi- ciency. J Pediatr Hematol Oncol. 2002;24:69–71.

24. Isoyama K, Ohnuma K, Ikuta K, Toyoda Y, Nakajima F, Yamada K, et al. Unrelated cord blood transplantation for second hemopoietic stem cell transplantation. Pediatr Int. 2003;45:268–74.
25. Kütükçüler N, Bölük E, Tökmeci N, Karaca NE, Azarsız E, Aksu G, Aykut A. Recurrent infections, neurologic signs, low serum uric acid levels, and lymphopenia in childhood: Purine nucleoside phosphorylase deficiency, an emergency for infants. Turk Pediatri Ars. 2020 Sep 23;55(3):320-327. doi: 10.14744/TurkPediatriArs.2019.83788.
26. Al-saud B, Alsmadi O, Al-muhsen S, Al-ghonaium A, Al-dhekri H. Case Report A novel mutation in purine nucleoside phosphorylase in a child with normal uric acid levels. Clin Biochem. The Canadian Society of Clinical Chemists; 2009;42:1725–7.
27. Camici, M., Micheli, V., Ipata, P.L., Tozzi, M.G., 2009. Pediatric neurological syndromes and inborn errors of purine metabolism. Neurochem. Int. 56, 367–378.
28. Mansouri, A., Min, W., Cole, C.J., et al., 2012. Cerebellar abnormalities in purine nucleoside phosphorylase deficient mice. Neurobiol. Dis. 47, 201–209.
29. Ozkinay F, Pehlivan S, Onay H et al (2007) Purine nucleoside phosphorylase deficiency in a patient with spastic paraplegia and recurrent infections. J Child Neurol 22:741–743
30. Somech R, Lev A, Grisaru-Soen G, Shiran SI, Simon AJ, Grunebaum E (2013) Purine nucleoside phosphorylase deficiency presenting as severe combined immune deficiency. Immunol Res 56: 150–154
31. la Marca G, Canessa C, Giocaliere E et al (2014) Diagnosis of immunodeficiency caused by a purine nucleoside phosphorylase defect by using tandem mass spectrometry on dried blood spots. J Allergy Clin Immunol 134:155–159
32. Martin, Jessica, Sharma, Richa, Nelson, Robert P., Schubert, Frank, Weida, Jennifer, 2016. The first report of a pregnancy in a patient with purine nucleoside phosphorylase deficiency. Fetal Pediatr. Pathol. 35 (2), 120–123.
33. Cohen A, Grunebaum E, Arpaia E, Roifman CM (2000) Immunodeficiency caused by purine nucleoside phosphorylase deficiency. Immunol Allergy Clin North Am 20(1):143–159.

Table 3

Table 3 is available in Supplementary Files section.

Figures

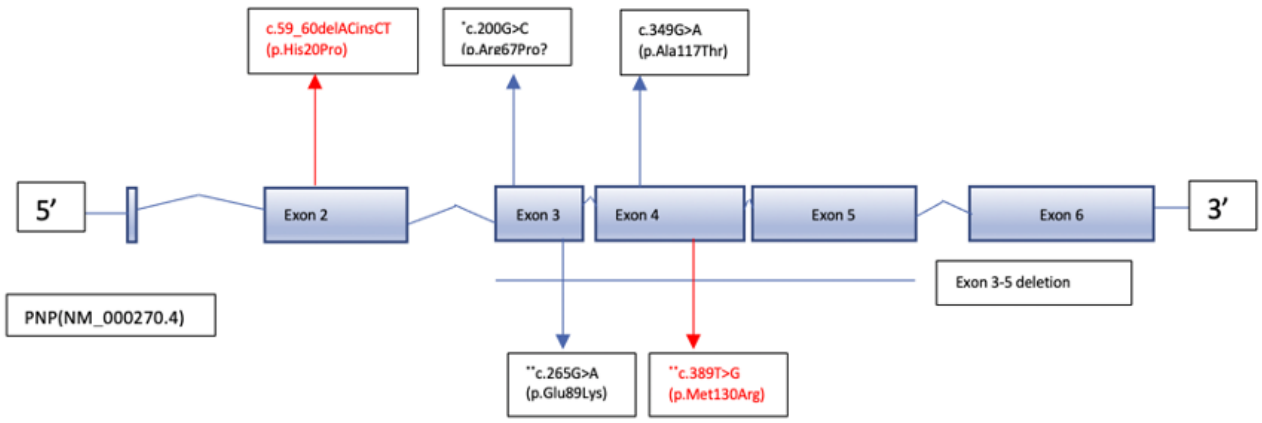


Fig 1.

Figure 1

The schematic representation of patients variants. *Shown in 2 patients. ** compound heterozygous

Novel mutations are shown with red color.

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- Table32.docx