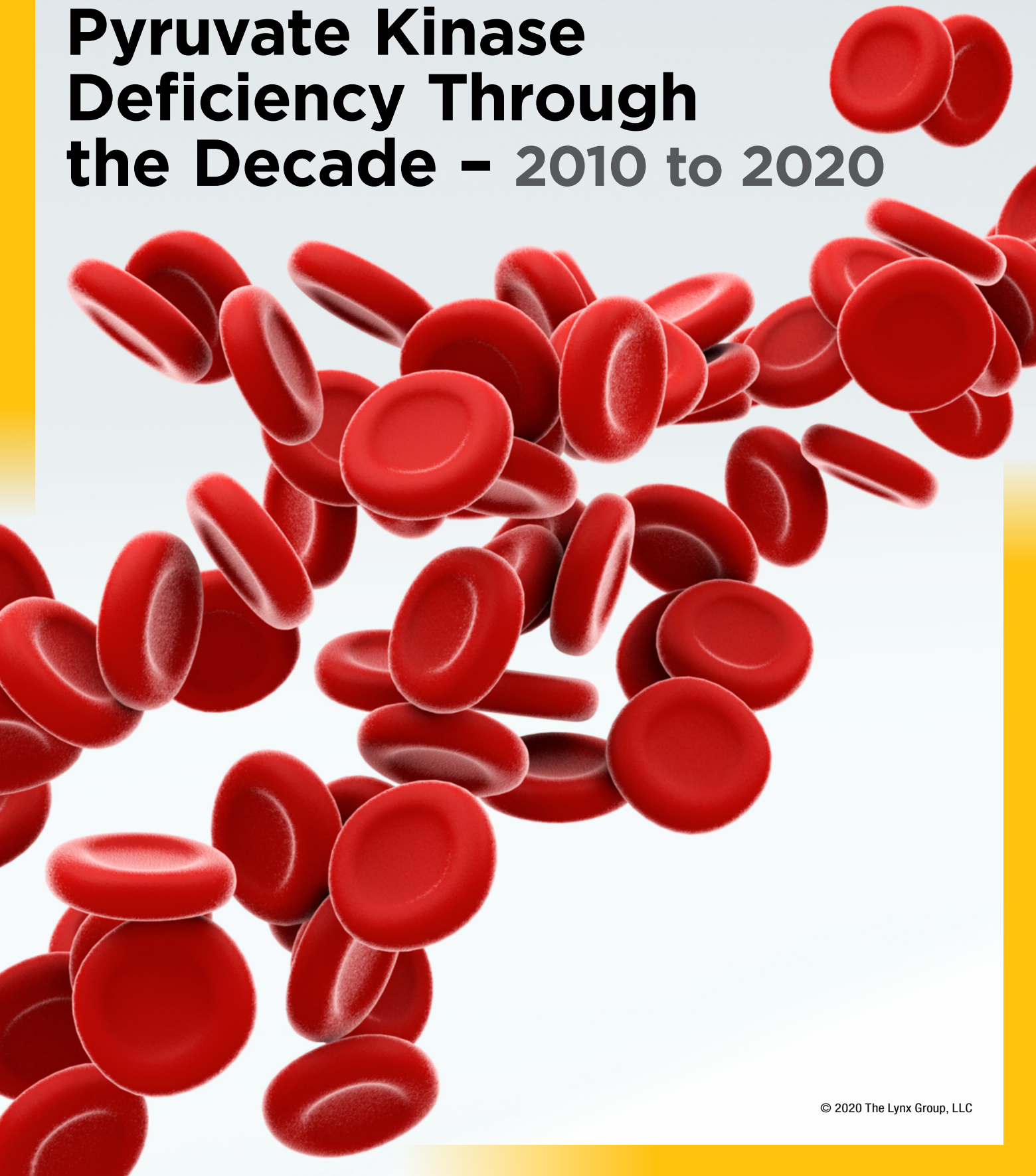
OCTOBER 2020 | SPECIAL FEATURE

This special feature was funded by

agios

Pyruvate Kinase Deficiency Through the Decade – 2010 to 2020



© 2020 The Lynx Group, LLC

Pyruvate Kinase Deficiency Through the Decade – 2010 to 2020

table of contents

3 **Part 1: Review of Pyruvate Kinase Deficiency**

12 **Part 2: Patient Perspectives on Living with PK Deficiency**

15 **Part 3: Summaries of PK Deficiency Literature (2010-2020)**

19 **Glossary**

thelynxgroup

President/CEO
Brian Tyburski

Executive Vice President
Nicholas Englezos
nenglezos@thelynxgroup.com

Executive Vice President
John W. Hennessy
jhennessy2@thelynxgroup.com

Executive Vice President
Russell Hennessy
rhennessy@thelynxgroup.com

Executive Vice President
Philip Pawelko
ppawelko@thelynxgroup.com

Executive Vice President
Shannon Sweeney
ssweeney@thelynxgroup.com

Senior Vice President, Finance
Andrea Kelly

Senior Director, Human Resources
Mara Castellano

Senior Medical Director
John Welz

AONN+ Program Director
Sharon S. Gentry, MSN, RN, HON-ONN-CG, AOCN, CBCN

Senior Director, Education and Program Development & Co-Director, Certification
Emily Gentry, BSN, RN, HON-ONN-CG, OCN

Director, Patient Navigation Program Development & Co-Director, Certification
Monica Dean, HON-OPN-CG

Custom Publishing Editorial Director
Todd Kunkler

Senior Director, Quality Control
Barbara Marino

Senior Director, Production & Manufacturing
Alaina Pede

Senior Director, Creative & Design
Robyn Jacobs

Senior Director, Digital Marketing
Samantha Weissman

Director, Association Project Management
Rachael Baranoski

STRATEGIX
a division of the lynx group

Managing Director
John Vassiliou

Vice President, Account Group Supervisor
Alexandra Charles
Deanna Martinez

Account Group Supervisors
Debora Burke
Pamela Intile

Published by The Lynx Group, LLC, 1249 South River Rd, Suite 202A, Cranbury, NJ 08512. Copyright © 2020 by The Lynx Group, LLC. All rights reserved. No part of this publication may be reproduced or transmitted in any form or by any means now or hereafter known, electronic or mechanical, including photocopy, recording, or any informational storage and retrieval system, without written permission from the Publisher. Printed in the United States of America.

The ideas and opinions expressed in this issue do not necessarily reflect those of the Editorial Board, the Editors, or the Publisher. Publication of an advertisement or other product mentioned in this issue should not be construed as an endorsement of the product or the manufacturer's claims. Readers are encouraged to contact the manufacturers about any features or limitations of products mentioned. Neither the Editors nor the Publisher assume any responsibility for any injury and/or damage to persons or property arising out of or related to any use of the material mentioned in this publication.

EDITORIAL CORRESPONDENCE should be addressed to **EDITORIAL DIRECTOR**, The Lynx Group, LLC, 1249 South River Rd, Suite 202A, Cranbury, NJ 08512. Phone: **732-992-1880**. Correspondence regarding permission to reprint all or part of the articles published in this supplement should be addressed to **REPRINT PERMISSIONS DEPARTMENT**, The Lynx Group, LLC, 1249 South River Rd, Suite 202A, Cranbury, NJ 08512.

TLG1646-2

Pyruvate Kinase Deficiency Through the Decade – 2010 to 2020

Advancements over the past decade have given individuals diagnosed with pyruvate kinase (PK) deficiency more reasons to be optimistic. As of May 2020, PK deficiency remains a rare condition with no approved medical therapies available. But things appear to be changing. Translational and clinical researchers are now working on the development of treatments that might address the underlying problem in PK deficiency—that is, a deficiency in the PK enzyme. What are these new developments? Could future therapies change the ways in which patients with PK deficiency live with their condition?

The goal of this article is to help individuals who have been diagnosed with PK deficiency, along with their caregivers and family members, understand more about the disorder and the latest breakthroughs in research. The article is divided into 3 parts:

- The first section reviews the basics of PK deficiency: its causes and symptoms, the steps that physicians take to diagnose the condition, and management approaches that can be used for patients with the disorder.
- The second section of the article highlights studies that have been undertaken to learn patients' perspectives about living with PK deficiency and how the condition affects their day-to-day lives.
- The final section of the article reviews some of the important clinical research on PK deficiency that has been published over the past decade. Researchers and clinicians who specialize in PK deficiency have made significant strides in understanding this disease. Reading through these summaries will help patients, caregivers, and family members have a clearer appreciation of the main findings from the PK deficiency–focused literature that their physicians read.

Please note that this article is not intended to serve as medical advice, to substitute for a doctor's appointment, or to be used for diagnosing or treating a disease. Readers are advised to consult with their physicians before making any decisions about their health.

Part 1: Review of Pyruvate Kinase Deficiency

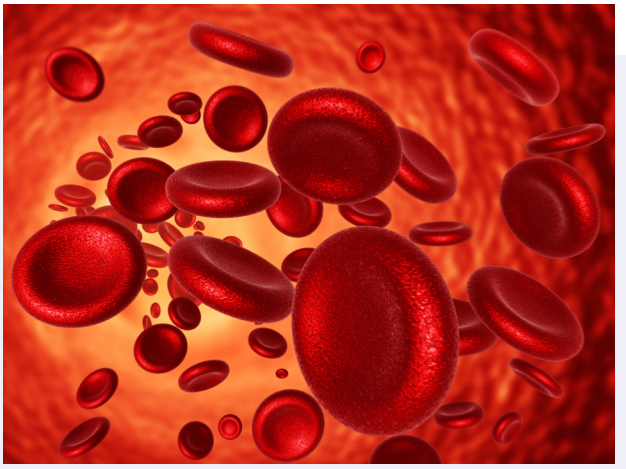
What Is PK Deficiency?

Pyruvate kinase (PK) deficiency is a rare genetic disease that affects red blood cells (RBCs).[1] Individuals with the condition are born with low levels of the PK enzyme in their bodies.[1]

Normally, RBCs use PK to break down glucose (a sugar) for energy.[2] Because individuals with PK deficiency have low amounts of the PK enzyme, their RBCs do not generate enough energy.[2] Instead, their RBCs hemolyze—meaning that they burst or rupture.[2] Rather than lasting for 120 days, RBCs in patients with PK deficiency last for only a few days to weeks.[2] (See **Sidebar 1**: What Are Red Blood Cells?)

When the number of RBCs in the blood becomes too low, this results in a condition known as anemia.[2] Patients with PK deficiency have lifelong hemolytic anemia.[1] Essentially, their bone marrow cannot produce RBCs fast enough to replace the RBCs that are being destroyed "too early."[2]

PK deficiency is a rare genetic condition that affects between 3.2 and 8.5 individuals of every 1 million, according to a report published by Storm and



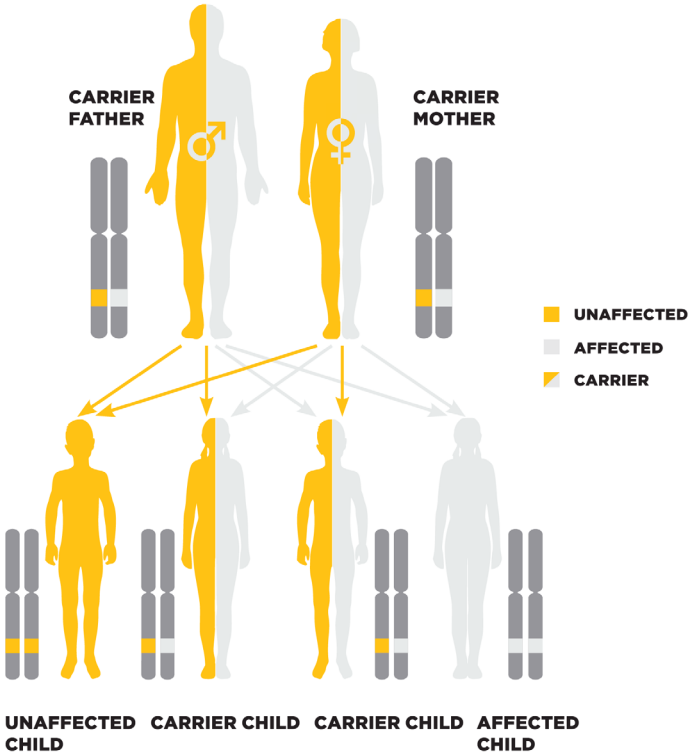
Sidebar 1: What Are Red Blood Cells?

RBCs, which are also called erythrocytes, move the oxygen that we breathe in through our lungs to all of the cells of our bodies. They also move carbon dioxide from those same cells to the lungs, to be exhaled.[2]

RBCs make energy by converting glucose into pyruvate—an important molecule in metabolism—and a high-energy molecule called adenosine triphosphate in a multistep process known as glycolysis.[2] The energy that is generated by glycolysis helps healthy RBCs keep their normal shape, stay flexible, and protect themselves from injury. In individuals with a normal amount of PK, RBCs can generate sufficient energy to last for an average of 120 days.[2]

RBCs that do not have enough PK cannot generate sufficient energy to hold their shape, and thus they break apart more easily than healthy RBCs. Instead of lasting for 120 days, PK-deficient RBCs last for only a few days to weeks.[2]

FIGURE 1. Inheritance of *PKLR* Gene and PK Deficiency from Parents Who Are Carriers of *PKLR* Gene Mutations



Source: Agios Pharmaceuticals, Inc.
PK indicates pyruvate kinase, *PKLR*; pyruvate kinase liver/red cell.

colleagues in 2019.[3] This suggests that of the approximately 784 million individuals in Europe, approximately 2500 to 6700 have PK deficiency. Some researchers believe that the actual prevalence of PK deficiency is higher, however—up to 51 individuals per million—because the condition is often undiagnosed.[3] As such, the number of undiagnosed cases of PK deficiency in Europe could be more than 40,000.

PK deficiency affects males and females in equal numbers.[1] Although many patients with PK deficiency reside in Europe and other areas of the world,[1,4] the condition is diagnosed more often in specific ethnic communities, such as the Pennsylvania Amish.[5,6]

What Causes PK Deficiency?

PK deficiency is an inherited condition, meaning that it can

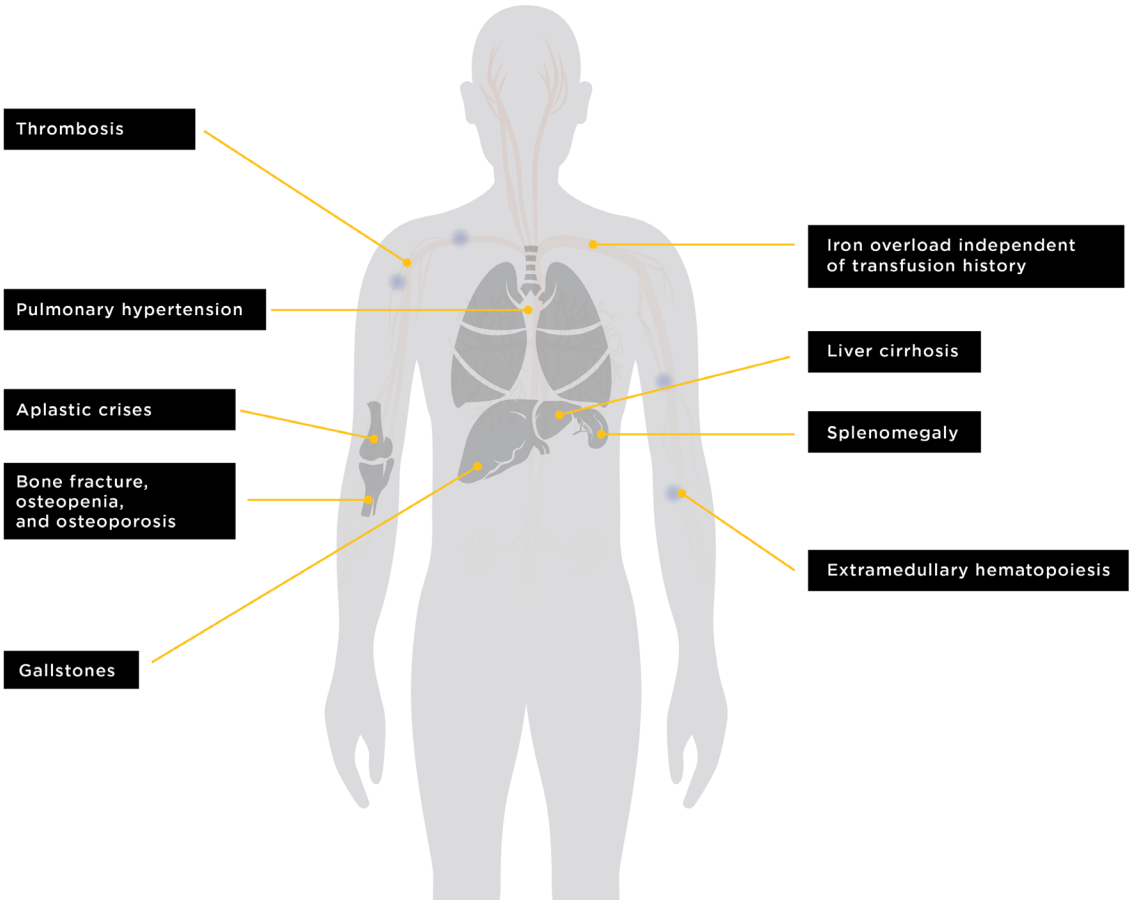
"My own parents...they were very anxious [about my PK deficiency]. As a child, I was constantly being looked over for listlessness, having my fingernails pressed down to see how quickly they would lighten up again with the blood circulation, and always going in for blood tests. Some of their anxiety probably transferred to me; I certainly have my own anxieties, even about medical issues. I'm comfortable in a hospital, but I'm also anxious there."—JS (patient)

be passed on from parents to their children.[2] PK deficiency is caused by permanent changes (or mutations) in a portion of DNA (or gene) called *PKLR*[2]—which stands for pyruvate kinase liver/red blood cells. The *PKLR* gene tells the body how to make pyruvate kinase.[2]

Genetic conditions occur when specific combinations of genetic material (ie, chromosomes) are received from each parent. "Recessive" genetic disorders occur when a person inherits the same mutated gene from each parent (see **Figure 1**).[2] A person who receives a normal gene from one parent and a mutated gene from the other parent is known as a disease carrier; these individuals usually do not have any symptoms of disease.[1] As shown in Figure 1, parents can be carriers but have no symptoms of PK deficiency.[2]

Recent studies of PK deficiency

FIGURE 2. Signs and Symptoms Associated with PK Deficiency



Source: Agios Pharmaceuticals, Inc.
PK indicates pyruvate kinase.

among individuals who live in the Middle East and sub-Saharan Africa suggest that another factor may be involved. People in these areas of the world are at a higher risk for malaria—a parasitic infection. They also have been shown to have a higher rate of PK deficiency.[7] Researchers have learned that the *PKLR* gene and the proteins that are made by the body in response to instructions by that gene may be important in helping individuals in those parts of the world resist the development of malaria.[8] In these patients, PK enzyme deficiency may be an "unintended consequence" of their exposure to malaria.[9]

"Patients with PK deficiency are jaundiced because they are destroying blood rapidly. Having jaundice and yellow eyes is not clinically problematic, but it is a psychological problem. People think that these patients have hepatitis."—BG (PK deficiency specialist)

Symptoms Associated with PK Deficiency

The symptoms and complications that people with PK deficiency experience can vary widely. **Figure 2** lists the signs and symptoms that are associated with PK deficiency.

- The anemia associated with PK deficiency can range from mild to severe.[2] According to recently published research, between 90% and 95% of patients with PK deficiency are anemic, ranging from mild anemia to RBC transfusion dependence[9]
- Enlargement of the spleen, or "splenomegaly," is also very common, affecting 80% to 85% of people with PK deficiency[9] (the spleen is an organ in the body that acts primarily as a blood filter)
- Yellowing of the skin and eyes, known as jaundice, is observed among 40% to 70% of individuals with PK deficiency[9]
- Gallstones are reported in 30% to 45% of patients[9]

PK deficiency is a lifelong condition. As such, symptoms of the disorder can be associated with long-term complications. The most common include iron overload and gallstones, but infection and other problems can occur as well.

- Iron overload
  - Iron overload can occur in individuals with PK deficiency even if they are not receiving regular blood transfusions[10]
  - Patients who receive blood transfusions should realize that every unit of transfused blood contains 200 to 250 mg of iron.[11] When patients with PK deficiency receive repeated blood transfusions, iron can accumulate in their body, causing iron overload.[2] Iron overload can damage the liver (abdominal pain), the heart (irregular heartbeat, heart failure), and the endocrine system (loss of sex drive)[2]
  - Because the human body is not able to eliminate excess iron, patients with PK deficiency who require chronic blood transfusions may need to receive iron chelation therapy as well.[2] It is important to understand that even patients who receive infrequent transfusions or who have never received a transfusion may require iron chelation treatment for high iron levels.[2] Several types of iron chelating agents are available, including some that are administered orally (ie, by mouth)
  - **Screening and monitoring of iron levels are important in all patients with PK deficiency, beginning in childhood.** Regular monitoring for iron overload and treatment, if needed, are key to the prevention of heart damage, liver problems, and other possible complications[2,9]
- Gallstones
  - Gallstones, also known as cholelithiasis, can occur in any person with PK deficiency and at any age[9]
  - Children, teenagers, and adults with gallstones often have higher levels of bilirubin—a waste product of RBCs[9]
  - Surgical removal of the gallbladder, a procedure known as cholecystectomy, is often recommended to treat gallstones and their complications[2]
  - Patients with PK deficiency should undergo regular

monitoring for gallbladder disease[9]

Other complications of PK deficiency can include severe infection, aplastic crisis (abrupt and severe anemia usually related to infection with parvovirus), hemolytic crisis (severe anemia associated with infection and pregnancy), and osteopenia (loss of bone mass).[9]

Other symptoms and potentially life-threatening adverse events associated with PK deficiency include the following[2,3,5,9]:

- Thrombosis (formation of blood clots)
- Pulmonary hypertension
- Dyspnea (shortness of breath)
- Fatigue
- Cognitive impairment

How Is PK Deficiency Diagnosed?

Please note that common signs and symptoms of PK deficiency are similar to those of other conditions. A comprehensive physical examination, laboratory tests, and imaging may be required to establish a definitive diagnosis.

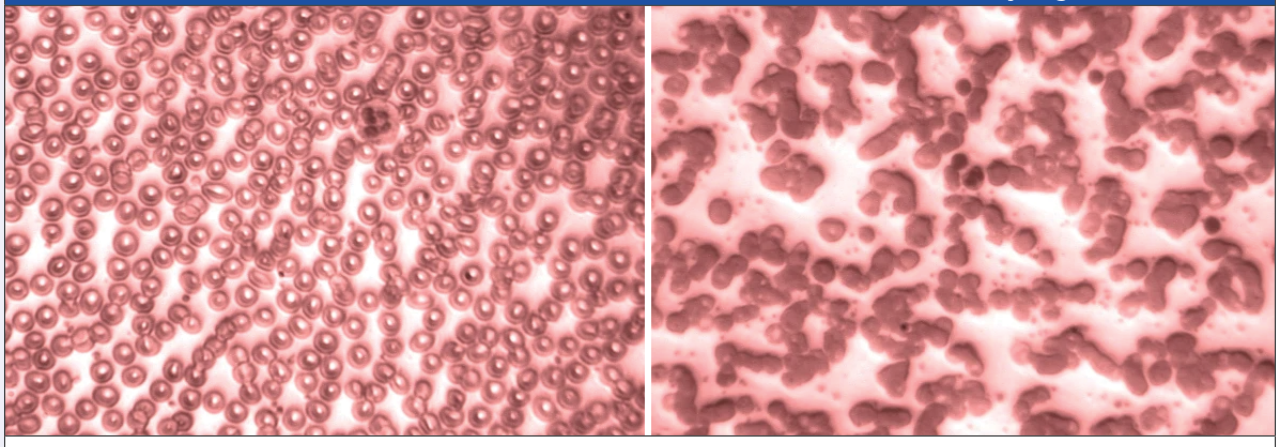
The degree of anemia that occurs among individuals with PK deficiency varies widely, ranging from very mild anemia with no symptoms to life-threatening anemia in newborns.[2] This means that some patients with

Sidebar 2: Red Blood Cell Testing

One test that physicians who diagnose and treat patients with PK deficiency use is called a complete blood count, or CBC. This test measures the cells that comprise your blood, including RBCs, white blood cells, and platelets. CBC testing is important, because it also reveals a patient's level of hemoglobin—the protein in the blood that holds oxygen.[12] People with PK deficiency usually have low hemoglobin levels (6-12 g/dL), which is attributable to the early destruction of their RBCs.[2] This leads to high levels of bilirubin—a substance released by RBCs as they break down.[2] The body offsets this RBC destruction by producing more RBCs, which means a patient's CBC will reveal a higher number of young, immature RBCs, or reticulocytes.[2]

The image on the left side of **Figure 3**[13] illustrates healthy RBCs, whereas the image on the right is from a patient with PK deficiency. In the right-hand image, fewer normal RBCs and more irregularly shaped dark cells can be seen. These darker cells are immature RBCs, or reticulocytes.

FIGURE 3. Normal RBCs (left) and Abnormal RBCs from a Patient with PK Deficiency (right)



RBC indicates red blood cells, PK; pyruvate kinase.

"The important thing for hematologists is to think about PK deficiency during the differential diagnosis. We believe that PK deficiency is more common than what we actually see in the clinics, because there is a lot of mild disease out there."
—BG (PK deficiency specialist)

PK deficiency are diagnosed as infants, whereas others are diagnosed later in life.[4]

In adults with anemia, healthcare providers typically perform a physical examination and ask about possible symptoms of PK deficiency, including fatigue, jaundice, and an enlarged spleen. When PK deficiency is suspected, screening includes bloodwork and genetic testing.[9] (See **Sidebar 2**: Red Blood Cell Testing.)

To determine whether a patient has PK deficiency, rather than another condition that causes hemolytic anemia (such as thalassemia, sickle cell anemia, or glucose-6-phosphate dehydrogenase [G6PD] deficiency), physicians look to observe 2 things:

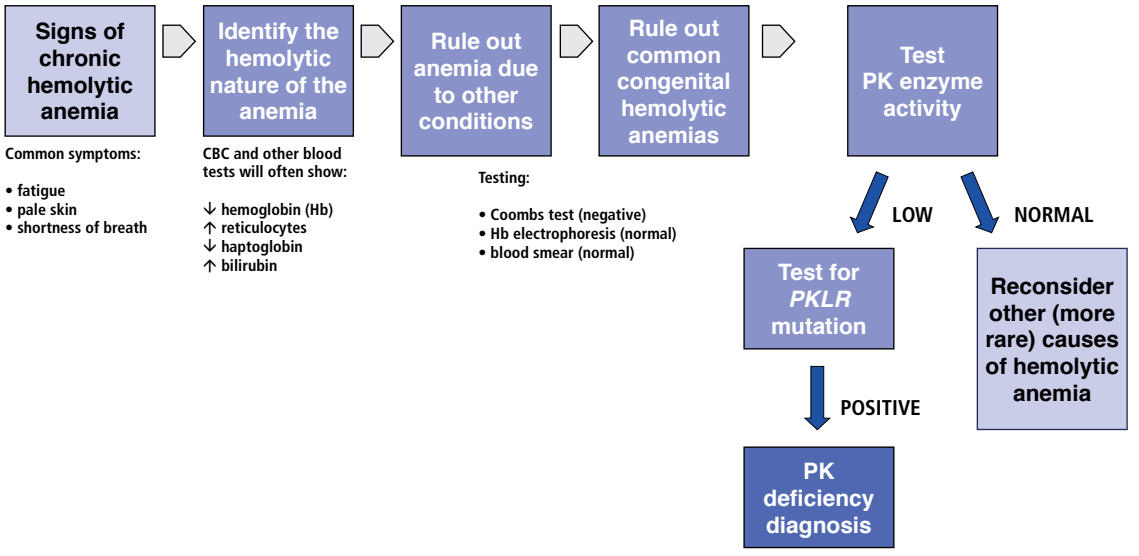
(1) Evidence of reduced PK enzyme activity

(2) Changes (ie, mutations) in the *PKLR* gene that are either compound heterozygous or homozygous[9] (see **Sidebar 3** for more information on this terminology).

When both reduced PK enzyme

"You can go to any hematologist and be lucky if they've seen anyone with PK deficiency. I've had a couple of stress fractures and bone fractures. This is not uncommon in PK deficiency; bone structure is weakened due to the larger bone marrow. But my regular doctors don't get that....I feel like I am still out there on my own, even though I live in a metropolitan area. There's still this lack of knowledge or lack of time for hematologists to really research PK deficiency."—JS (patient)

FIGURE 4. PK Deficiency Diagnosis Steps



Adapted from Grace RF, Layton DM, Barcellini W. How we manage patients with pyruvate kinase deficiency. *Brit J Haematol*. 2019;184(5):721-734.
CBC indicates complete blood count; PK, pyruvate kinase; *PKLR*, pyruvate kinase liver/red cell.

activity and a mutation in the *PKLR* gene are found, physicians can confirm the diagnosis of PK deficiency (see **Figure 4**).[9]

To date, more than 300 mutations in the *PKLR* gene have been associated with PK deficiency.[9] As new therapies are being developed for the treatment of patients with PK deficiency, accurate diagnosis becomes even more important.[4] Not only does the accurate diagnosis of PK deficiency help patients to understand what they are experiencing, but it also allows physicians who treat PK deficiency to determine the best therapy and monitoring approach.

Any patient without a genetically confirmed diagnosis of PK deficiency should ask his or her physician about molecular testing for the disorder. Patients with PK deficiency might also want to speak with their undiagnosed family members about genetic counseling to learn whether future offspring could be affected by the condition.[4]

How Is PK Deficiency Managed?

Physicians—typically, hematologists—who care for patients with PK deficiency focus on managing anemia and other symptoms related to the disorder. None of the currently available therapies for PK deficiency affects the underlying cause of the enzyme deficiency.[2]

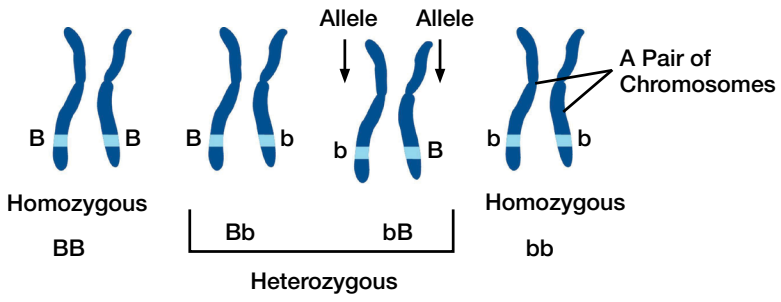
Patients with anemia associated with PK deficiency may require blood transfusions. Recent literature estimates that 84% of individuals with PK deficiency have received a transfusion at least once in their lifetime.[9] Hematologists offer RBC transfusions based on each patient's symptoms and ability to tolerate anemia, rather than on a particular

Sidebar 3: Heterozygous and Homozygous Gene Mutations

Every person receives 2 forms of each gene—or alleles—one from each of their biological parents.[14] **Figure 5** illustrates heterozygous and homozygous genes.[15]

This figure depicts **chromosomes**—the threadlike structures of DNA and protein that are found in the nucleus of most living cells. Chromosomes carry genetic information in the form of genes. An **allele** is any of the possible forms in which a gene for a specific trait can occur. In almost all human cells, 2 alleles for each gene are inherited—1 from each parent. B and b represent 2 different alleles of a single gene. Homozygous refers to a person who has inherited the same allele from each parent (eg, BB), and heterozygous refers to a person who has inherited different alleles from each parent (eg, Bb).

FIGURE 5. Homozygous and Heterozygous Genes



- Heterozygous gene mutation: The mutation affects only 1 form of a gene (1 allele).[16] This means that an individual has inherited 2 different forms of the gene—1 from each parent[15]
- Compound heterozygous mutation: Each allele has a different mutation[16]
- Homozygous mutation: Identical mutations are found on both alleles. This means that an individual has inherited the same DNA sequence for a particular gene from both parents[15]

Most patients with PK deficiency have compound heterozygous mutations, which means that the symptoms of PK deficiency that they experience can vary widely.[9]

"In adolescence, I knew what it was like to be transfused to a hemoglobin of 10 [g/dL] and how you felt like Superman. The feeling between 8.5 and 10 [g/dL] is an incredible difference. You feel like you can run. You feel like you don't require as much sleep. You feel really good for a few days...maybe for a couple of weeks."—JS (patient)

hemoglobin level. That means that when and how often a patient with PK deficiency should receive an RBC transfusion is personalized, based on the patient's symptoms, as well as the impact that anemia has on his or her day-to-day activities and quality of life (QOL).[9] **Table 1** outlines the pros and cons associated with blood transfusions.

Patients who receive regular RBC transfusions should be closely monitored for iron overload. Regardless of whether they receive transfusions, patients with PK deficiency should avoid taking iron supplements, including multivitamins with iron and foods that are high in iron (eg, liver, red meat).[2] Other vitamins and supplements, however, may be beneficial for patients with PK deficiency. Depending on the amount of folic acid in their diet, patients with PK deficiency may need to take folic acid supplements. Folic acid is a B vitamin that is needed to make RBCs. Additionally, vitamin D and calcium supplements may promote bone health in these patients.[2]

Surgical removal of the spleen (ie, splenectomy) can decrease RBC transfusion requirements in children and adults with PK deficiency. Most older children and

TABLE 1. Benefits and Risks of Blood Transfusions in Patients with PK Deficiency

Benefits of Blood Transfusions	Drawbacks of Blood Transfusions
In PK deficiency, the body may not have enough RBCs to carry oxygen through the body to the heart and brain Replacing RBCs through transfusions: • ensures that oxygen gets to vital organs • helps with symptoms of anemia: the person feels better and has more energy Blood products are tested according to national guidelines to ensure their safety	Transfusions require time and a visit to a hospital or clinic, which can be burdensome for some patients and families Infection risk: HIV, hepatitis B, hepatitis C Additional risks and reactions: • Allergic reactions, including fever, chills, and rash • Breathing problems due to fluid overload • Immune reaction in which the body makes antibodies that destroy the transfused RBCs • Errors leading to administration of the wrong product Iron overload in regularly transfused patients (6 or more transfusions each year)

Adapted from Cleveland Clinic. A patient's guide to blood transfusions. Accessed June 20, 2020. https://my.clevelandclinic.org/health/treatments/14755-a-patients-guide-to-blood-transfusions.
HIV indicates human immunodeficiency virus; PK, pyruvate kinase; RBCs, red blood cells.

TABLE 2. Benefits and Risks of Splenectomy in Patients with PK Deficiency

Benefits of Splenectomy	Risks of Splenectomy
• Anemia or hemolysis improves* • The need for transfusions ceases or decreases (in most patients) • Hemoglobin levels rise by an average of 1.5-2 g/dL (in most patients) • Reticulocytes survive and numbers increase (from 5%-15% prior to splenectomy to 50%-70% in some instances) • No further risk for splenic injury or rupture (if spleen was enlarged) *Improvement in anemia or hemolysis associated with PK deficiency almost always follows splenectomy, but hemolysis persists in nearly all cases, with a rise in reticulocyte and bilirubin levels.	• General risks associated with anesthesia and surgery; these should be discussed with the surgeon prior to the splenectomy • 10% risk for blood clots, which can lead to stroke, pulmonary embolism, and other life-threatening complications • Higher lifetime risk for serious, even life-threatening, infections – Long-term antibiotic treatment required – Long-term fever management protocol required • Rarely, no significant effect on hemoglobin levels; transfusions still required

Adapted from Grace R. *Fast Facts for Patients and Supporters.* Pyruvate Kinase Deficiency: A rare genetic disease that affects red blood cells. Basel, Switzerland: S. Karger Publishers Limited; 2018. Accessed May 25, 2020. www.karger.com/Book/Home/278171.
PK indicates pyruvate kinase.

adults with PK deficiency have undergone a splenectomy.[9] Because a high risk for infection exists among young children, whenever possible, this surgery is typically delayed until after 5 years of age.[9] **Table 2** outlines the pros and cons associated with splenectomy.[2]

> "The reason we take out the spleen is basically because someone has a high transfusion requirement. But there are risks with not having a spleen. If you start treating PK deficiency with a new drug earlier, in childhood, could you avoid the need for splenectomy? That would make a difference."—BG (PK deficiency specialist)

In 2018, Grace and colleagues reported the results of an international, multicenter registry—the Pyruvate Kinase Deficiency Natural History Study—that collected clinical data on patients with PK deficiency. In this study, splenectomy was performed in 150 of 254 patients (59%) with PK deficiency.[18] Patients' median age at the time of splenectomy was 4.1 years (range, 0.4-37.8 years).[18]

The most common reasons for splenectomy in these patients included the following[18]:

- Improve baseline anemia (88%)
- Decrease transfusion burden (85%)
- Improve patient QOL (83%)
- Reduce jaundice (57%)

Among these patients, sple-

TABLE 3. Recommended Monitoring for Patients with PK Deficiency

Condition to Monitor	Recommended Monitoring and Frequency
Hemolytic anemia	Blood tests: CBCs, reticulocyte count, bilirubin • At least once per year • Frequency of these tests can vary based on a number of factors, including a patient's RBC transfusion needs
Gallstones	Ultrasound imaging • Once per year
Iron overload	Ferritin levels • Once per year in nontransfused patients • At least 2 times per year in transfused patients • Every 1-3 months in patients receiving iron chelation therapy MRI imaging in patients receiving regular transfusions • Once per year
Aplastic crisis (rapid onset of severe anemia)	• In patients with aplastic crisis and reticulocytopenia (ie, low numbers of immature RBCs), physicians should check parvovirus IgM or PCR once to confirm the diagnosis • Patients with PK deficiency should be aware of the risk for an aplastic crisis and the need for close monitoring after exposure to parvovirus B19
Blood-borne viral infections	HIV and hepatitis screening • Once per year in patients who received a transfusion in the prior year
Osteopenia	Vitamin D levels and possibly DEXA scans • Baseline assessment in adolescence or early adulthood • Frequency of DEXA scans and vitamin D monitoring depends on findings from the initial assessment
Endocrine disorders	Thyroid dysfunction, sex hormone abnormalities, and diabetes (evaluating fructosamine levels, not HbA_{1c}, is recommended when screening for diabetes) • Once per year in patients with iron overload
Pulmonary hypertension	Echocardiogram (heart imaging) • As needed in patients ≥30 years of age, prior to pregnancy, and at any age in patients with symptoms suggestive of poor heart function and/or pulmonary hypertension, such as shortness of breath
Extramedullary hematopoiesis (blood cell production occurring outside of the bone marrow)	Radiographic imaging • As needed to review the liver and spleen or other organs, based on the development of new symptoms

Adapted from Grace RF, Layton DM, Barcellini W. How we manage patients with pyruvate kinase deficiency. *Brit J Haematol*. 2019;184:721-734.
CBCs indicates complete blood cell counts; DEXA, dual-energy x-ray absorptiometry; Hb, hemoglobin; HIV, human immunodeficiency virus; IgM, immunoglobulin M; MRI, magnetic resonance imaging; PCR, polymerase chain reaction; PK, pyruvate kinase; RBC, red blood cell.

nectomy was associated with a median increase in hemoglobin levels of 1.6 g/dL.[18] Almost all (90%) patients required fewer RBC transfusions following splenectomy.[18] A common complication after splenectomy was clotting (thrombosis), which was reported in 11% of patients.[18]

Although data supporting its use are sparse, one type of treatment—hematopoietic stem-cell transplantation (HSCT)—has been shown to cure some individuals with PK deficiency.[9,19] HSCT involves taking healthy stem cells from the bone marrow or blood and putting them into the bloodstream of someone with a disease that causes stem-cell damage. These new stem cells then make healthy new blood cells.[20]

In 2018, a global effort was undertaken by van Straaten and colleagues to identify patients with PK deficiency who had undergone HSCT and to learn more about their outcomes. This resulted in a publication that describes 16 patients with PK deficiency who underwent HSCT in Europe or Asia between 1996 and 2015.[21] Although HSCT was successful in some of these patients, 5 patients died of transplant-related causes.[21] A lack of robust information about the risks associated with HSCT and the fact that most patients with PK deficiency live for years make some experts skeptical about use of this procedure.[9]

Currently, clinicians specializing in the management of patients with PK deficiency recommend splenectomy and/or regular RBC transfusions, rather than HSCT, for the treatment of PK deficiency–associated anemia. If HSCT approaches are refined to reduce the risk for long-term complications, the role of the procedure in patients with PK deficiency may change.[9]

Patients with PK deficiency should see their physicians routinely for monitoring. **Table 3** summarizes important monitoring recommendations from experts in PK deficiency.[9]

Part 2: Patient Perspectives on Living with PK Deficiency

Because PK deficiency is a rare disease, it is difficult for healthcare providers to understand and appreciate the burdens faced by patients with the disorder.[22] To learn more about the overall experience of living with PK deficiency, in 2018 Grace and colleagues conducted a qualitative study of adults to explore how signs and symptoms of the condition impact their health-related quality of life (HRQOL).[22] Interviews were conducted among a total of 21 individuals with PK deficiency from the United States, The Netherlands, and Germany.[22]

In the Grace 2018 study, patients with PK deficiency most often reported yellow eyes, tiredness, yellow skin, fatigue, low energy, and shortness of breath.[22] **Table 4** shows how these PK deficiency–related signs and symptoms affect various aspects of patients' lives, including activities of daily living, leisure time, work/school functioning, and social life.[22] This research by Grace and colleagues is a key step in the development of surveys that can be used to learn more about the signs and symptoms of PK deficiency and their impact on patients' HRQOL. Data derived from these types of surveys are important because they can

> "I didn't have any problems with my gallbladder until my 30s. It was misdiagnosed as an ulcer, so it was treated for an ulcer for a while. Then GERD [gastroesophageal reflux disease] for a while. I just kept having these attacks. Finally, I had such an attack and turned so bright yellow that the doctor sent me directly to the emergency room. Then things were taken care of; I had my gallbladder removed. There were a lot of painful episodes."—JS (patient)

TABLE 4. PK Deficiency Burden

Symptoms of Anemia	Physical Deficits	Appearance	Activities of Daily Living	Social/Emotional Impact
Tired/fatigued	Constant need for more rest	Negative impact on appearance	Lack of motivation	Negative impact on relationships/social activities
Low energy/stamina	Physical exertion more difficult (climbing stairs, exercise, sports)		Low productivity	Unwanted attention
General weakness	Weakened immune system/susceptibility to illness		Difficulty with chores and other daily tasks	Fear/concern over future effects of condition
Dizziness/short of breath				
Appearance-related symptoms				
Jaundice, pale skin				
Other symptoms				
Cognitive impairment				
Bone/joint pain				

Signs and symptoms that were reported by ≥3 of the 21 participants with PK deficiency are listed on the left. Impacts (ie, physical limitations, changes in appearance) are arranged from left to right to indicate how closely related each is to the signs and symptoms of PK deficiency.[22]
PK indicates pyruvate kinase.

help healthcare professionals better appreciate the real challenges faced by individuals living with PK deficiency. These surveys also help drug researchers learn more about the effects of new pharmacologic interventions on HRQOL of patients with PK deficiency.[22]

Another important initiative for the PK deficiency community was the externally led Patient-Focused Drug Development meeting, which was held on September 20, 2019, in Hyattsville, Maryland.[23] This meeting was important because patients and caregivers were able to share their experiences with physicians and researchers, as well as with representatives from the US Food and Drug Administration (FDA). This was also the largest gathering of individuals with PK deficiency in the world at any time in history, with 17 patients being in the room.[23] The meeting was held in response to the FDA's interest in more systematically obtaining patients' perspectives on the burden of PK deficiency and the impact of current treatments for the disease. A detailed report of these findings reviews patients' and caregivers' unique insights on the effect of PK deficiency, currently available management approaches, and their expectations about future treatments. Some of the critical insights garnered from this meeting are summarized in **Table 5**.[23]

To learn more about the natural history and day-to-day impact of PK deficiency, 2 additional research studies are currently underway. One of these—the Pyruvate Kinase Defi-

> "The ongoing research could be really important for future people who would be diagnosed with PK deficiency. In fact, we have earlier diagnosis now; people are being diagnosed as babies. This is valuable, because parents don't have to keep wondering what's going on with their kid. It's scary for parents, but [with the PK deficiency diagnosis] I think they feel very supported. And at least they know what's going on. Parents can now anticipate what's going to be needed, like splenectomy or partial splenectomy. They can anticipate iron overload and begin to treat that earlier on, so the kids don't have organ problems."—JS (patient)

TABLE 5. Summary of Findings for Patient-Focused Drug Development Meeting – September 2019

Patients with pyruvate kinase (PK) deficiency continuously deal with very difficult issues in their daily lives, the most important of which include the following:
- Tiredness, exhaustion, fatigue
- Shortness of breath
- Difficulty concentrating (ie, "brain fog")
- Memory loss
- Anemia

Patients with PK deficiency experience a high level of emotional and social stresses, including the following:
- Anxiety
- Low self-esteem
- Social isolation
- Depression
- Bullying

Patients living with PK deficiency find that their disease limits them in various ways:
- Disconnected from others and stigmatized
- Unable to participate in sports/physical activities
- Miss work or school too often
- General limitations in daily function

Current treatments for PK deficiency do not sufficiently manage patients' most significant symptoms:
- 11% of polled participants stated that their current treatments work "very well"
- 58% responded with "moderately well"
- 14% noted that their treatments work "poorly"
- 8% indicated that their treatments "do not work at all"

"I've learned to balance...to accept. I see that I have strengths in some areas and limitations in others. You have to accept your weaknesses, as well as your strengths. Balance is important. Sleep is important. Doing some form of exercise—to the extent that you can tolerate it—and continuing to try to build strength are still important. Do it to help you as an individual, not to keep up with the neighbor or somebody else who is in their 50s at your gym. Find a gym that caters to older people. I do great in senior yoga, by the way!"—JS (patient)

ciency Natural History Study (PKD NHS)—was initiated in December 2013.[24] The goal of PKD NHS is to describe the range and incidence of symptoms, treatments, and complications associated with PK deficiency.[24] The study is collecting retrospective data on the medical history and routine clinical care of patients with PK deficiency at baseline and annually for up to 2 years.[24] This effort is expected to be useful in the design of future clinical trials of new therapies for patients with PK deficiency.[24,25]

The first report of findings from PKD NHS, which was authored by Grace and colleagues in 2018, included 254 patients with molecularly confirmed PK deficiency.[25] After collecting and analyzing their medical data, researchers learned many important things about PK deficiency[25]:

- Complications during pregnancy were common, including high levels of bilirubin, anemia that required transfusions, hydrops fetalis (a life-threatening condition that can occur in fetuses and newborns), and premature delivery
- Almost all newborns with PK deficiency (93%) were treated with phototherapy (a special type of light therapy) and many were treated with exchange transfusions (46%), with both of these treatments used to counteract the effects of jaundice
- Children under 5 years of age were often transfused until they underwent splenectomy
- More than half (59%) of the 254 patients with PK deficiency who were evaluated in the NHS had undergone splenectomy[25]
  - Splenectomy resulted in a median increase in hemoglobin of 1.6 g/dL and lowered transfusion burden for 90% of these patients[25]
- Approximately one-third (34%) of 254 patients underwent both a splenectomy and a cholecystectomy
- The most frequent complications of PK deficiency included
  - Iron overload (48%)
  - Gallstones (45%)
- Other complications included
  - Bone fracture (17%)
  - Aplastic crisis (14%)
  - Extramedullary hematopoi-

esis (ie, RBC production in parts of the body other than the bone marrow; 9%)
- Delayed puberty (8%)
- Thyroid disease (5%)
- Pulmonary hypertension (3%)
- Liver cirrhosis (3%)
- Leg ulcers (2%)

Although the risk for complications increased if patients had more severe anemia, complications of PK deficiency were reported among all patients in this registry. Despite these complications, life expectancy for patients with PK deficiency appears to be long.[25] Grace and colleagues concluded that close monitoring by a healthcare professional is important for all patients with PK deficiency, including those with mild anemia.[25]

> "We are now recognizing that older patients with PK deficiency go from being more or less functional to being tired all the time as they age. We are just learning about this now as we review registry data. If this decline is related to anemia and we have a drug available, these are the people who could benefit."—BG (PK deficiency specialist)

The other ongoing initiative is the Pyruvate Kinase Deficiency Global Longitudinal Registry, or Peak Registry, which is a global registry for patients with PK deficiency.[26] The registry will be open for enrollment for 7 years, with all enrolled participants followed prospectively for a minimum of 2 years and a maximum of 9 years.[26] Data regarding symptoms, signs, and treatment of PK deficiency will be collected from participating registry physicians, patients, and, if appropriate, parents/guardians of children with PK deficiency.[26] As of July 2020, no information from the Peak Registry has been published.

Part 3: Summaries of PK Deficiency Literature (2010-2020)

Below are summaries of important PK deficiency–focused publications that have appeared in the medical literature since 2010. By reviewing this information, patients with PK deficiency, as well as their caregivers and family members, can become aware of the medical research updates about their disease that their physicians may be reading. These summaries may also help patients and caregivers to inform physicians about these publications and ensure that all individuals involved are working together to manage the disorder.

PK DEFICIENCY REVIEW

Grace, *Am J Hematol* 2015: This article reviews the clinical diversity of PK deficiency, complications with the disease, current treatments, and future treatment directions. Researchers' understanding of the genetic variations, causes, and complications of the hemolytic anemia associated with PK deficiency has expanded. Significant gaps remain, however, regarding knowledge about clinical care and monitoring of patients with PK deficiency. Treatments such as phototherapy and exchange transfusion in the newborn period, regular or intermittent RBC transfusions in children and adults, and splenectomy to decrease transfusion requirements and symptoms of anemia all can be valuable. **Grace and colleagues conclude that the need for more evidence exists, in order to develop PK deficiency–specific guidelines for supportive care and monitoring of complications. They refer to the Pyruvate Kinase Deficiency Natural History Study as an important step in understanding more about PK deficiency.[27]**

Grace RF, Zanella A, Neufield EJ, et al. Erythrocyte pyruvate kinase deficiency: 2015 status report. *Am J Hematol.* 2015;90:825-830. Link to article

Grace, *Brit J Haematol* 2019: This review article summarizes the approach that experts in the management of PK deficiency currently use for the diagnosis, monitoring, and treatment of children and adults with the disorder. Tables 3 and 4 are particularly helpful, as they review monitoring and supportive care guidelines for patients with PK deficiency. Table 5 of this article summarizes the risks and benefits of transfusions versus splenectomy in patients with PK deficiency. The article also discusses novel treatment options for PK deficiency, including PK activators and gene therapy, suggesting that they may transform the way in which patients with the disease are managed in the future. **Key takeaways from this publication include the following: (1) Decisions to transfuse and/or perform a splenectomy should be individualized to each patient with PK deficiency; (2) Because all patients with PK deficiency are at risk for long-term complications, they should be monitored regularly; and (3) The development of disease-specific therapies in the future makes formal diagnosis of PK deficiency even more important today.[9]**

Grace RF, Layton DM, Barcellini W. How we manage patients with pyruvate kinase deficiency. *Brit J Haematol.* 2019;184:721-734. Link to article

Bianchi, *Am J Hematol* 2019: In 2016, a global PK deficiency International Working Group was created, which included 24 experts from 20 centers located around the world. This group studied the current gaps in the diagnosis of PK deficiency to establish diagnostic guidelines. Broad consensus was reached among these experts on many clinical and technical aspects involved in the diagnosis of PK deficiency. These guidelines increase awareness of this rare condition among the medical community, helping physicians make more timely and accurate diagnoses of PK deficiency. **Key takeaways from this publication include the following: (1) Both a PK enzyme test and *PKLR* genetic testing are important for diagnosing PK deficiency; (2) A timely and accurate diagnosis is important for patients to understand more about their PK deficiency, thus allowing hematologists to proactively manage disease symptoms and monitor complications; and (3) A diagnosis of PK deficiency should be considered in patients with chronic hemolysis who do not have an acquired autoimmune disorder or other inherited hemolytic anemias.[4]**

Bianchi P, Fermo E, Glader B, et al. Addressing the diagnostic gaps in pyruvate kinase deficiency: consensus recommendations on the diagnosis of PK deficiency. *Am J Hematol*. 2019;94:149-161. Link to article

Al-Samkari, *Haematologica* 2020: Recognizing the complicated nature of PK deficiency and its management, and the limitations of the literature, an international working group of 10 PK deficiency experts convened to better define the disease burden and manifestations. This article summarizes the conclusions of this working group, and serves as a guide for clinicians and investigators who care for patients with PK deficiency. Case studies of patients with PK deficiency are provided in this article to highlight the different ways in which the disease can affect people. These cases can help physicians be aware of the various types of symptoms that they might encounter. The researchers also underscore the importance of close patient monitoring and individualized care. **Key conclusions drawn by the authors include the following: (1) Patient symptoms do not always match the degree of anemia in a person with PK deficiency; RBC transfusions should be based on symptoms and lifestyle choices, not on the patient's hemoglobin level; (2) The spectrum of signs and symptoms of PK deficiency is broad; symptoms can change over a patient's lifetime; and (3) Complications such as iron overload can arise, even if patients did not receive RBC transfusions.[28]**

Al-Samkari H, van Beers EJ, Kuo KHM, et al. The variable manifestations of disease in pyruvate kinase deficiency and their management. *Haematologica*. 2020 Mar 12. doi: 10.3324/haematol.2019.240846. Online ahead of print. Link to article

PK DEFICIENCY SYMPTOMS

Grace, *Blood* 2018: An international, multicenter registry was established to collect clinical data on patients with PK deficiency. Medical history and laboratory data were obtained at enrollment from 254 patients with molecularly confirmed PK deficiency. The most frequent complications of PK deficiency included iron overload (48%) and gallstones (45%). Additional complications included aplastic crises, osteopenia/bone fragility, RBC production outside of the bone marrow (extramedullary hematopoiesis), postsplenectomy infection (sepsis), pulmonary hypertension, and leg ul-

cers. Although the risk associated with these PK deficiency–related complications increases with the severity of a patient's anemia, complications were common among all patients with the disorder. **Because there are few early predictors of the severity of PK deficiency and because even patients with mild anemia can experience long-term complications, all individuals with PK deficiency should be closely monitored for complications, particularly iron overload and the development of gallstones.**[25]
Grace RF, Bianchi P, van Beers EJ, et al. Clinical spectrum of pyruvate kinase deficiency: data from the Pyruvate Kinase Deficiency Natural History Study. *Blood*. 2018;131:2183-2192. Link to article

Grace, *Eur J Haematol* 2018: This is the first study that shares patients' perspectives on the burden of living with PK deficiency. Based on interviews conducted among a total of 21 patients with PK deficiency, the most common signs and symptoms reported included scleral icterus (n = 19), tiredness (n = 18), jaundice (n = 17), fatigue (n = 15), low energy (n = 13), and shortness of breath (n = 13). These symptoms of PK deficiency negatively impact patients' ability to perform physical activities, as well as their appearance, social activities, leisure activities, work and/or school, sleep, emotional states, and cognitive states. Bone pain and joint pain, reported in 8 patients and 3 patients, respectively, are not recognized as symptoms of PK deficiency and represent a new finding. Similarly, difficulty concentrating and memory loss, reported in 4 patients and 2 patients, respectively, are of particular interest, considering the relatively young age of the cohort of participants in this study (median age, 38.9 years; range, 19-58 years).[22]
When summarizing their learnings, the researchers noted that patients living with a chronic disease such as PK deficiency often describe a "new normal" way of living, which is borne from acceptance of and adjustments to living with their condition. The investigators noted that it is possible this "normalization" trend may result in underestimating the true impact of the patients' symptom experience. **This research lays the foundation for future studies designed to explore the effects of new treatments on the overall HRQOL of patients living with PK deficiency.**[22]
Grace RF, Cohen J, Egan S, et al. The burden of disease in pyruvate kinase deficiency: patients' perception of the impact on health-related quality of life. *Eur J Haematol*. 2018;101:758-765. Link to article

PK DEFICIENCY GENETICS

Canu, *Blood Cells Mol Disease* 2015: Clinical PK deficiency is transmitted to children as an autosomal recessive trait. Two PK genes are present: (1) the pyruvate kinase liver and red blood cells (*PKLR*) gene, and (2) the pyruvate kinase muscle (*PKM*) gene. Reports have been published showing a variety of genetic defects in the *PKLR* gene. The researchers of this analysis reviewed about 250 published mutations and 6 forms of the *PKLR* gene, with the corresponding clinical and molecular data. **This review provides useful information for clinicians and laboratory professionals about reported *PKLR* gene mutations and helps experts harmonize the data on patients with PK deficiency.**[29]
Canu G, De Bonis M, Minucci A, Capoluongo E. Red blood cell PK deficiency: an update of *PK-LR* gene mutation database. *Blood Cells Mol Dis*. 2016;57:100-109. Link to article

Bianchi, *J Pediatr* 2020: This study reports on the molecular features of 257 patients with PK deficiency who enrolled in the Pyruvate Kinase Deficiency Natural History Study. Of the 127 different variants in *PKLR* that were detected, 66% were missense and 34% were non-missense. Among 177 unrelated patients, 20% were homozygous and 80% were compound heterozygous. Overall, 55 patients (21%) were found to have at least 1 previously unreported variant, and 45 new mutations in *PKLR* were described. **This technical report confirms the wide variety of genetic mutations in patients with PK deficiency. Researchers also indicated that (1) Signs and symptoms of PK deficiency at birth were not predictable, even among siblings; (2) Patients with 2 non-missense mutations had a more severe clinical picture—that is, higher rates of splenectomy, lower hemoglobin levels after splenectomy, higher numbers of lifetime transfusions, and higher rates of iron overload; and (3) Pregnancy was similarly tolerated in women with PK deficiency regardless of their mutation, but women with 2 non-missense *PKLR* variants required transfusion support during pregnancy.**[30]
Bianchi P, Fermo E, Lezon-Geyda K, et al. Genotype-phenotype correlation and molecular heterogeneity in pyruvate kinase deficiency. *Am J Hematol*. 2020;95:472-482. Link to article

Qin, *Int J Lab Hematol* 2020: PK deficiency is an autosomal recessive disorder in which the affected individuals are either homozygous or compound heterozygous for *PKLR* mutations. Although measurement of PK enzymatic activity is essential for the diagnosis of PK deficiency, testing for PK activity could yield false-negative results if the patient recently received an RBC transfusion. **Even when laboratory tests reveal low PK activity, testing for the *PKLR* gene is strongly recommended to confirm a diagnosis of PK deficiency. Next-generation sequencing technology is particularly helpful in the diagnosis of PK deficiency and in genetic counseling.[31]**
Qin L, Nie Y, Chen L, et al. Novel *PKLR* mutations in 4 families with pyruvate kinase deficiency. *Int J Lab Hematol*. 2020;42:e84-e87. Link to article

PK DEFICIENCY MANAGEMENT

Iolascon, *Haematologica* 2017: It is well understood that splenectomy is one possible therapeutic approach to the management of patients with severe anemias. Except for hereditary spherocytosis (a condition for which the effectiveness of splenectomy has been well documented), however, the efficacy of splenectomy in other anemias, including PK deficiency, is unclear. Concerns also exist about short- and long-term infections, as well as other complications associated with splenectomy, including blood clots. In this article, experts provide specific recommendations for splenectomy in anemia disorders, including PK deficiency. Since no randomized clinical trials of splenectomy were found in the literature that was reviewed by the experts, recommendations for each disease were based on expert opinion. **This expert panel recommends consideration of splenectomy in patients who are transfusion-dependent or severely anemic and/or who do not tolerate anemia. They also recommend that cholecystectomy (ie, removal of the gallbladder) be performed concurrently with splenectomy.[32]**
Iolascon A, Andolfo I, Barcellini W, et al. Recommendations regarding splenectomy in hereditary hemolytic anemias. *Haematologica*. 2017;102:1304-1313. Link to article

van Beers, *Haematologica* 2018: This analysis describes the prevalence and clinical characteristics of iron overload in patients enrolled in the Pyruvate Kinase Deficiency Natural History Study, with a focus on those individuals who are not regularly transfused with RBCs. Researchers found a high rate of iron overload in patients who are regularly transfused as well as in those who are not regularly transfused. Iron overload was found to occur at all ages and regardless of hemoglobin level. **These findings suggest that iron screening is important in all patients with PK deficiency. Iron levels should be monitored regularly, beginning in childhood. Prompt treatment of iron overload is critical. The authors recommend that MRI scans be performed in patients with PK deficiency whose ferritin levels are above 500 ng/mL.[10]**
van Beers EJ, van Straaten S, Morton DH, et al. Prevalence and management of iron overload in pyruvate kinase deficiency: report from the Pyruvate Kinase Deficiency Natural History Study. *Haematologica*. 2019;104:e51-e53. Link to article

Al-Samkari, *Am J Hematol* 2020: Using data collected from the Pyruvate Kinase Deficiency Natural History Study, the researchers compared 2 groups of patients with PK deficiency based on severity of disease. They found that patients with PK deficiency who underwent splenectomy and did not require regular blood transfusions have similar rates of disease-associated complications compared with those who underwent splenectomy and did require regular blood transfusions (ie, ≥6 every year). **Iron overload was the only complication of PK deficiency that was more common in patients who require regular transfusions. Because transfusion requirements fluctuate considerably over time, they are not a good marker of disease severity in patients with PK deficiency. Stated differently, transfusion dependence following splenectomy does not necessarily imply a worse clinical outcome for patients with PK deficiency.[33]**
Al-Samkari H, van Beers EJ, Morton DH, et al. Characterization of the severe phenotype of pyruvate kinase deficiency. *Am J Hematol.* 2020 Jul 3. doi: 10.1002/ajh.25926. Online ahead of print. Link to article

Glossary of Key Terms in PK Deficiency[2]

Aplastic crisis: the production of new red blood cells temporarily stops completely

ATP: adenosine triphosphate; a high-energy molecule made by glycolysis, which red blood cells use for energy

Bilirubin: a substance released from red blood cells when they break down; bilirubin causes jaundice (yellowing of the skin)

Capillaries: small narrow blood vessels found throughout the body

Chelation therapy: drugs that remove excess iron from the blood by binding with the iron to form substances that can be excreted from the body easily

Cholecystectomy: surgical removal of the gallbladder

Gallstones: small stones that form in the gallbladder

Gene mutation: a permanent change in the DNA sequence of a gene, altering the gene's instructions to make a protein such as pyruvate kinase so that the protein stops working properly

Glycolysis: a multistep process in which glucose (a sugar) is converted into pyruvate and energy

Hematocrit: the volume of red blood cells in the blood, typically reported as a percentage

Hematopoiesis: red blood cell production

Hemoglobin: a protein in red blood cells that transports oxygen around the body, typically reported as grams per deciliter

Hemolysis: the destruction of red blood cells

Hemolytic anemia: low numbers of red blood cells or a low hemoglobin level due to destruction of red blood cells

Iron overload: an excess of iron in the body

Jaundice: yellowing of the skin

Phlebotomy: withdrawal of blood from the body

Pyruvate: an important metabolic molecule that is the end product of glycolysis

Pyruvate kinase: an enzyme that makes the last step in glycolysis happen, converting phosphoenrolpyruvate into pyruvate and energy

Reticulocyte: young (not fully mature) red blood cell

Splenectomy: surgical removal of the spleen

Splenomegaly: enlargement of the spleen

References

1. National Organization for Rare Disorders (NORD) website. Rare disease database: Pyruvate kinase deficiency. 2019. https://rarediseases.org/rare-diseases/pyruvate-kinase-deficiency. Accessed August 30, 2020.
2. Grace R. *Fast Facts for Patients and Supporters*. Pyruvate Kinase Deficiency: A rare genetic disease that affects red blood cells. Basel, Switzerland: S. Karger Publishers Limited; 2018. www.karger.com/Book/Home/278171. Accessed August 30, 2020.
3. Storm M, Secrest MH, Carrington C, et al. Prevalence of red cell pyruvate kinase deficiency: a systematic literature review. *Blood*. 2019;134(suppl 1):3513.
4. Bianchi P, Fermo E, Glader B, et al; EuroBloodNet, the European Reference Network in Rare Hematological Diseases. Addressing the diagnostic gaps in pyruvate kinase deficiency: consensus recommendations on the diagnosis of pyruvate kinase deficiency. *Am J Hematol.* 2019;94:149-161.
5. Kanno H, Balas SK, Miwa S, et al. Molecular abnormality of erythrocyte pyruvate kinase deficiency in the Amish. *Blood*. 1994;83:2311-2316.
6. Rider NL, Strauss KA, Brown K. Erythrocyte pyruvate kinase deficiency in an old-order Amish cohort: longitudinal risk and disease management. *Am J Hematol*. 2011;86:827-834.
7. Machado P, Manco L, Gomes C, et al. Pyruvate kinase deficiency in sub-Saharan Africa: identification of a highly frequent missense mutation (G829A;Glu277Lys) and association with malaria. *PLoS One*. 2012;7:e47071.
8. van Bruggen R, Gualtieri C, Iliescu A, et al. Modulation of malaria phenotypes by pyruvate kinase (*PKLR*) variants in a Thai population. *PLoS One*. 2015;10:e0144555.
9. Grace RF, Layton DM, Barcellini W. How we manage patients with pyruvate kinase deficiency. *Brit J Haematol.* 2019;184:721-734.
10. van Beers EJ, van Straaten S, Morton DH, et al. Prevalence and management of iron overload in pyruvate kinase deficiency: report from the Pyruvate Kinase Deficiency Natural History Study. *Haematologica*. 2019;104:e51-e53.
11. Hoffbrand AV, Taher A, Cappellini MD. How I treat transfusional iron overload. *Blood*. 2012;120:3657-3669.
12. WebMD.com. What does hemoglobin (Hb or Hgb) mean from a complete blood count (CBC)? Reviewed by Louise Chang on December 22, 2018. www.webmd.com/a-to-z-guides/qa/what-does-hemoglobin-hb-or-hgb-mean-from-a-complete-blood-count-cbc. Accessed August 30, 2020.
13. Pyruvate Kinase Deficiency website. Diagnosis of PKD. https://pyruvatekinasedeficiency.com/diagnose-of-pkd. Accessed August 30, 2020.
14. National Human Genome Research Institute. www.genome.gov/genetics-glossary/Allele. Accessed August 25, 2020.
15. National Human Genome Research Institute. www.genome.gov/genetics-glossary/heterozygous. Accessed August 25, 2020.
16. MedicineNet.com. Medical definition of compound heterozygote. Last editorial review January 25, 2017. www.medicinenet.com/script/main/art.asp?articlekey=33675. Accessed August 30, 2020.
17. Cleveland Clinic. A patient's guide to blood transfusions. https://my.clevelandclinic.org/health/treatments/14755-a-patients-guide-to-blood-transfusions. Accessed August 30, 2020.
18. Grace RF, Bianchi P, van Beers EJ, et al. Clinical spectrum of pyruvate kinase deficiency: data from the Pyruvate Kinase Deficiency Natural History Study. *Blood*. 2018;131:2183-2192.
19. Kim M, Park J, Lee J, et al. Hemolytic anemia with null *PKLR* mutations identified using whole exome sequencing and cured by hematopoietic stem cell transplantation combined with splenectomy. *Bone Marrow Transplant*. 2016;51:1605-1608.
20. Cancer.net. What is a bone marrow transplant (stem cell transplant)? Approved by the Cancer.Net Editorial Board, July 2020. www.cancer.net/navigating-cancer-care/how-cancer-treated/bone-marrowstem-cell-transplantation/what-bone-marrow-transplant-stem-cell-transplant. Accessed August 30, 2020.
21. van Straaten S, Bierings M, Bianchi P, et al. Worldwide study of hematopoietic allogeneic stem cell transplantation in pyruvate kinase deficiency. *Haematologica*. 2018;103:e82-e86.
22. Grace RF, Cohen J, Egan S, et al. The burden of disease in pyruvate kinase deficiency: patients' perception of the impact on health-related quality of life. *Eur J Haematol*. 2018;101:758-765.
23. National Organization for Rare Disorders (NORD) and Foundation for Rare Blood Diseases (SZB). Voice of the Patient Report: Pyruvate Kinase Deficiency. Meeting Date: September 20, 2019. Report Date: January 21, 2020. https://rarediseases.org/wp-content/uploads/2020/01/NRD-2029-Voice-of-the-Patient-Report-PKD_FNL-1.pdf. Accessed August 30, 2020.
24. Clinicaltrials.gov. Pyruvate Kinase Deficiency Natural History Study (PKD NHS). https://clinicaltrials.gov/ct2/show/NCT02053480. Accessed August 31, 2020.
25. Grace RF, Bianchi P, van Beers EJ, et al. Clinical spectrum of pyruvate kinase deficiency: data from the Pyruvate Kinase Deficiency Natural History Study. *Blood*. 2018;131:2183-2192.
26. Clinicaltrials.gov. Pyruvate Kinase Deficiency Global Longitudinal Registry (PEAK Registry). https://clinicaltrials.gov/ct2/show/NCT03481738. Accessed August 31, 2020.
27. Grace RF, Zanella A, Neufield EJ, et al. Erythrocyte pyruvate kinase deficiency: 2015 status report. *Am J Hematol*. 2015;90:825-830.
28. Al-Samkari H, van Beers EJ, Kuo KHM, et al. The variable manifestations of disease in pyruvate kinase deficiency and their management. *Haematologica*. 2020 Mar 12. doi: 10.3324/haematol.2019.240846. Online ahead of print.
29. Canu G, de Bonis M, Minucci A, Capoluongo E. Red blood cell PK deficiency: an update of *PK-LR* gene mutation database. *Blood Cells Mol Dis*. 2016;57:100-109.
30. Bianchi P, Fermo E, Lezon-Geyda K, et al. Genotype-phenotype correlation and molecular heterogeneity in pyruvate kinase deficiency. *Am J Hematol*. 2020;95:472-482.
31. Qin L, Nie Y, Chen L, Zhang D, Lin Y, Ru K. Novel PKLR mutations in 4 families with pyruvate kinase deficiency. *Int J Lab Hematol*. 2020;42:e84-e87.
32. Iolascon A, Andolfo I, Barcellini W, et al. Recommendations regarding splenectomy in hereditary hemolytic anemias. *Haematologica*. 2017;102(8):1304-1313.
33. Al-Samkari H, van Beers EJ, Morton DH, et al. Characterization of the severe phenotype of pyruvate kinase deficiency. *Am J Hematol.* 2020 Jul 3. doi: 10.1002/ajh.25926. Online ahead of print.

NOTES

NOTES

NOTES

Interested in what's next for PK deficiency?
Agios is dedicated to the science and studies behind medicines to treat rare genetic disorders, like PK deficiency.

To stay informed about the latest news, resources, and research related to PK deficiency, register for updates at **KnowPKDeficiency.com**.

agios

©2020 Agios Pharmaceuticals, Inc.
All rights reserved.