Diagnosis and Management of Osteopetrosis: Consensus Guidelines From the Osteopetrosis Working Group

Article *in* The Journal of Clinical Endocrinology and Metabolism · June 2017

DOI: 10.1210/jc.2017-01127

12 authors, including:

Michael Alan Levine
The Children's Hospital of Philadelphia
535 PUBLICATIONS **21,903** CITATIONS
SEE PROFILE

Paul J Orchard
University of Minnesota Twin Cities
397 PUBLICATIONS **14,976** CITATIONS
SEE PROFILE

Anna Petryk
Alexion
176 PUBLICATIONS **6,842** CITATIONS
SEE PROFILE

Eric Rush
Children's Mercy Kansas City
65 PUBLICATIONS **859** CITATIONS
SEE PROFILE

Diagnosis and Management of Osteopetrosis: Consensus Guidelines From the Osteopetrosis Working Group

Calvin C. Wu,[1] Michael J. Econs,[2] Linda A. DiMeglio,[3] Karl L. Insogna,[4] Michael A. Levine,[5] Paul J. Orchard,[6] Weston P. Miller,[6] Anna Petryk,[6,7] Eric T. Rush,[8] Dolores M. Shoback,[9] Leanne M. Ward,[10] and Lynda E. Polgreen[11]

[1]Department of Medicine, Los Angeles Biomedical Research Institute at Harbor-UCLA Medical Center, Torrance, California 90502; [2]Departments of Medicine and Medical and Molecular Genetics, Indiana University School of Medicine, Indianapolis, Indiana 46202; [3]Department of Pediatrics, Indiana University School of Medicine, Indianapolis, Indiana 46202; [4]Department of Medicine, Yale University, New Haven, Connecticut 06519; [5]Division of Endocrinology and Diabetes, Children's Hospital of Philadelphia, and Department of Pediatrics, University of Pennsylvania Perelman School of Medicine, Philadelphia, Pennsylvania 19104; [6]Department of Pediatrics, University of Minnesota, Minneapolis, Minnesota 55455; [7]Alexion Pharmaceuticals, Inc., New Haven, Connecticut 06510; [8]Departments of Pediatrics and Internal Medicine, University of Nebraska Medical Center, Omaha, Nebraska 68198; [9]Endocrine Research Unit, Department of Medicine, San Francisco Department of Veterans Affairs Medical Center, University of California, San Francisco, California 94143; [10]Department of Pediatrics, University of Ottawa, Ottawa, Ontario K1N 6N5, Canada; and [11]Department of Pediatrics, Los Angeles Biomedical Research Institute at Harbor-UCLA Medical Center, Torrance, California 90502

Background: Osteopetrosis encompasses a group of rare metabolic bone diseases characterized by impaired osteoclast activity or development, resulting in high bone mineral density. Existing guidelines focus on treatment of the severe infantile forms with hematopoietic cell transplantation (HCT) but do not address the management of patients with less severe forms for whom HCT is not the standard of care. Therefore, our objective was to develop expert consensus guidelines for the management of these patients.

Methods: A modified Delphi method was used to build consensus among participants of the Osteopetrosis Working Group, with responses to an anonymous online survey used to identify areas of agreement and conflict and develop a follow-up survey. The strength of recommendations and quality of evidence was graded using the Grading of Recommendations Assessment, Development and Evaluation system.

Results: Consensus was found in the areas of diagnosis, monitoring, and treatment. We recommend relying on characteristic radiographic findings to make the diagnosis and found that genetic testing adds important information by identifying mutations associated with unique disease complications. We recommend ongoing monitoring for changes in mineral metabolism and other complications, including cranial nerve impingement, anemia, leukopenia, and dental disease. We suggest that calcitriol should not be used in high doses and instead recommend symptom-based supportive therapy for disease complications because noninfantile osteopetrosis has no effective treatment.

Conclusions: Scarcity of published studies on osteopetrosis reduce the ability to develop evidence-based guidelines for the management of these patients. Expert opinion-based guidelines for this rare condition are nevertheless important to enable improved care. (***J Clin Endocrinol Metab*** **102: 3111–3123, 2017)**

ISSN Print 0021-972X ISSN Online 1945-7197
Printed in USA

Received 16 May 2017. Accepted 20 June 2017.
First Published Online 26 June 2017

Abbreviations: 25(OH)D, 25-hydroxyvitamin D; CAII, carbonic anhydrase II; DXA, dual-energy x-ray absorptiometry; HCT, hematopoietic cell transplantation; MIM, Mendelian Inheritance in Man; RBC, red blood cell.

Osteopetrosis is a group of rare bone disorders, within the family of sclerosing bone dysplasias, characterized by reduced osteoclastic bone resorption that results in a high bone mass. Rather than conferring strength, the overly dense bone architecture belies a structural brittleness that predisposes to fracture. The disruption of normal bone modeling and remodeling can give rise to skeletal deformity and dental abnormalities and can interfere with mineral homeostasis. In addition, expansion of bone into marrow cavities and cranial nerve foramina can compromise hematologic and neurologic function, respectively; the former may manifest as profound anemia, bleeding, frequent infections, and hepatosplenomegaly from extramedullary hematopoiesis. The latter can lead to blindness, deafness, and nerve palsies (1).

Not every patient is affected in the same way. Osteopetrosis comprises a clinical spectrum ranging from very mild to severe disease phenotypes that are fatal in the first year of life. Osteopetrosis has for decades been categorized descriptively by its clinical severity and inheritance pattern into a "malignant" autosomal recessive infantile form, a "benign" adult autosomal-dominant form (which can be severe), and an intermediate form. The incidence is variably estimated at 1:200,000 for autosomal-recessive osteopetrosis (2) and 1:20,000 for autosomal-dominant osteopetrosis (3). Recent molecular and genetic advances have begun to clarify the molecular basis of the disease and now allow the classification of osteopetrosis by its underlying molecular pathogenesis. The identification of specific genetic mutations can yield important prognostic and therapeutic implications (see following section on Genetic Testing). Unfortunately, incomplete penetrance and variable expressivity contribute to the wide phenotypic spectrum that can be seen even within kinships (4), limiting the ability to correlate the genotype and clinical phenotype.

The rarity and heterogeneity of osteopetrosis can present substantial challenges to clinicians. Existing consensus guidelines by the European Society for Immunodeficiencies and the European Society for Blood and Marrow Transplantation Inborn Errors Working Party provide a framework for the diagnosis, treatment, and follow-up of patients with infantile osteopetrosis, but they focus largely on the indications for, approaches to, and complications of hematopoietic cell transplantation (HCT) (5). They notably do not address the very different clinical course and treatment of patients with relatively milder forms of osteopetrosis, which can still be associated with substantial morbidity. Therefore, the objectives of these consensus guidelines are to summarize the collective experience of our panel of experts and provide a basis for standardizing the diagnostic approach and medical management of patients with noninfantile forms of osteopetrosis.

Methods

Methods for developing consensus recommendations

We began by formulating open-ended questions to elicit the minimum criteria for diagnosis, necessary baseline studies, practice patterns, monitoring strategies, and referral indications for specific medical subspecialties and for HCT. We distributed this survey electronically, inviting members of the Osteopetrosis Working Group, which consists of endocrinologists, geneticists, orthopedic surgeons, hematologists, and blood and marrow transplant physicians, to participate. Responses were collected anonymously and analyzed for areas of agreement and conflict; questions were then revised and recirculated for a second round of input from our experts. A summary of collective responses from the first round was also included, and participants were given an opportunity to rate their agreement with the suggestions of their peers. In this way, a modified Delphi method was used to build consensus.

Methods for developing evidence-based recommendations

We conducted a systematic review of the literature on osteopetrosis to ensure that our consensus guidelines were not only founded on expert opinion but also were evidence-based when possible. English-language publications with abstracts available and pertaining to humans from January 1970 to January 2017 were collected by searching PubMed using the keyword "osteopetrosis." Additional publications were then added to this collection on the basis of relevant citations in those publications.

We applied the Grading of Recommendations Assessment, Development and Evaluation system (6) to weigh the quality of evidence and strength of our recommendations. Because osteopetrosis is a rare disease, the body of literature is sparse and composed primarily of case reports and case series. High-quality evidence in the form of randomized controlled trials and meta-analyses is lacking. The strength of each recommendation was indicated as 1 (strong recommendation) or 2 (weak recommendation). The quality of the evidence was indicated by cross-filled circles, such that ⊕◯◯◯ denotes very-low-quality evidence; ⊕⊕◯◯, low quality; ⊕⊕⊕◯, moderate quality; and ⊕⊕⊕⊕, high quality.

Diagnosis

Imaging

We recommend that demonstration of the classic radiographic features (Fig. 1) of osteopetrosis is the minimum needed to make the diagnosis (1|⊕⊕⊕◯).

Osteopetrosis, derived from the Greek words for "bone" ("osteo") and "stone" ("petros"), is a fitting name for a disease in which generalized osteosclerosis identifiable on standard radiographs is pathognomonic. Parallel bands of dense bone can give the appearance of "bone-within-bone" or "endobones." This finding is

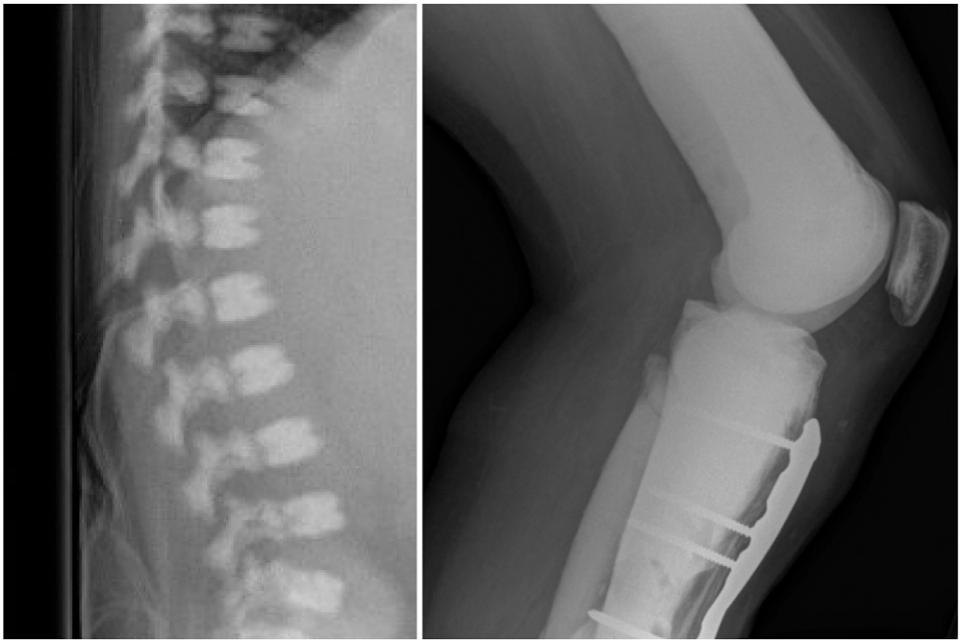

Figure 1. Classic radiographic features. Left: Vertebral midbodies sandwiched between dense bands along superior and inferior endplates ("sandwich vertebrae"). Right: Bone-within-bone appearance.

often prominent in the pelvis, long bones, phalanges, and vertebrae. Vertebrae can also be uniformly dense or take on a "sandwich vertebrae" or "rugger-jersey" appearance (when a normal-appearing vertebral midbody is sandwiched between dense bands along the superior and inferior endplates). Club-shaped flaring of the metaphyses of long bones with cortical thinning (Erlenmeyer flask bone deformity) and transverse metaphyseal lucent bands reflect a failure of metaphyseal remodeling but are not exclusive to osteopetrosis (7). Evidence of new or healing fractures may be found, and skull changes can include calvarial and basilar thickening as well as poor sinus development.

Because of the wide gamut of pathognomonic radiographic features of osteopetrosis, a skeletal survey is sufficient to make the diagnosis.

Genetic testing

We recommend genetic testing once the radiographic diagnosis of osteopetrosis has been established because it can distinguish among forms of osteopetrosis with unique complications (1|⊕⊕⊕○).

Before osteoclasts can achieve bone resorption, they must first embark on a path (Fig. 2) that involves differentiation from granulocyte-macrophage precursors, attachment to bone, creation of a ruffled border (resorptive surface), and acidification of the bone to dissolve and digest the mineral matrix. The last step requires the generation of protons by carbonic anhydrase II (CAII), transport of protons through the ruffled border by a vacuolar proton pump, and maintenance of electroneutrality in the ruffled border by chloride exchange (1). Most defects in osteopetrosis represent loss-of-function mutations that impair the acidification process through trafficking and/or fusion of lysosome-related organelles to the ruffled border. Mutations in the osteoclast vacuolar proton pump [*TCIRG1*, Mendelian Inheritance in Man (MIM) 604592] and the H^+-Cl^- exchange transporter 7 (*CLCN7*, MIM 602727) account for nearly 70% of autosomal-recessive osteopetrosis, whereas rarer variants encode the *CAII* enzyme (MIM 611492), a stabilizing β-subunit of the H^+-Cl^- exchange transporter 7 (*OSTM1*, MIM 607649), and a lysosome-associated protein involved in vesicular trafficking *(PLEKHM1,* MIM 611466) (8). On the other hand, mutations in tumor necrosis factor ligand superfamily member 11 (*TNFSF11,* MIM 602642) and tumor necrosis factor receptor superfamily member 11A (*TNFRSF11A,*MIM 603499) are associated with osteoclast-poor forms of autosomal recessive osteopetrosis by interfering with the receptor activator of nuclear factor κ-B receptor and ligand signaling pathway through which osteoblasts activate osteoclastogenesis.

Genetic testing may provide critical information on prognosis and clinical associations that have a meaningful effect on management decisions. CAII deficiency presents with a constellation of renal tubular acidosis and cerebral calcifications, and special attention needs to be paid to the potential for nephrocalcinosis and nephrolithiasis. HCT provides great potential as a means of remodeling the bony changes associated with osteopetrosis, because repopulating the marrow with the necessary hematopoietic precursors allows recovery of functional osteoclasts. However,

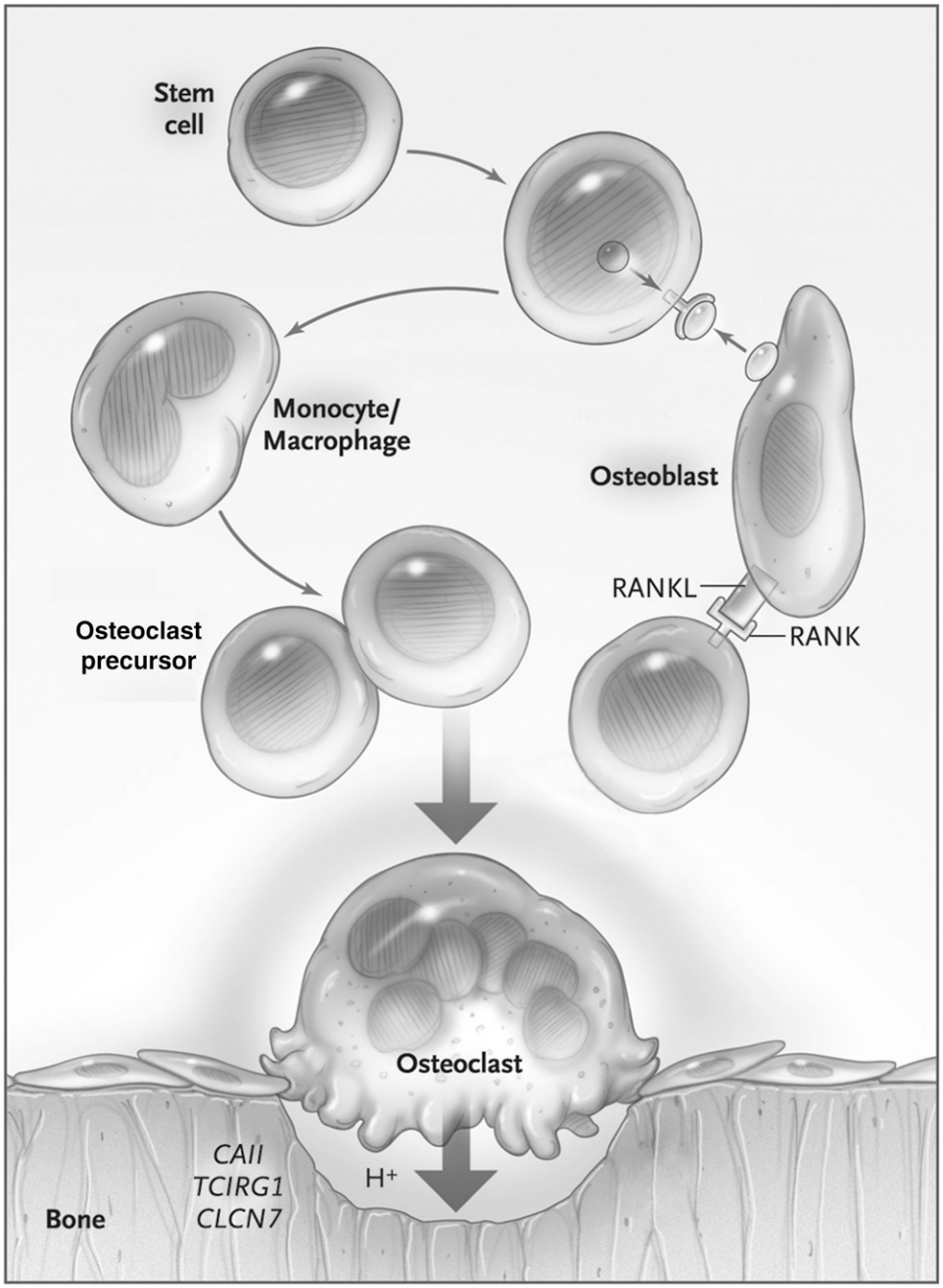

Figure 2. Molecules affecting osteoclast differentiation and function. The binding of receptor activator of nuclear factor κ-B (RANK) ligand (RANKL) produced by osteoblasts to RANK on osteoclast precursors induces osteoclast differentiation; in animals with defects in these genes, no osteoclasts are observed. In disorders of acidification—including defects in carbonic anhydrase II (*CAII*), the osteoclast-specific proton pump (encoded by *TCIRG1*, also termed *ATP6i*), and the gene encoding a chloride channel (*CLCN7*)—there are no primary defects in differentiation. (Modified from *N Engl J Med* Tolar J, Teitelbaum SL, and Orchard PJ, Osteopetrosis, 351:2839-49, copyright © 2004 Massachusetts Medical Society. Reprinted with permission.)

this holds true only for mutations intrinsic to the osteoclast. Therefore, although the *TNFRSR11A* and *TNFSF11* mutations both lead to an osteoclast-poor phenotype, HCT can treat the former but not the latter because nuclear factor κ-B receptor is expressed on osteoclasts, whereas nuclear factor κ-B receptor ligand is produced by cells of mesenchymal origin. *OSTM1* defines a rare subset of patients with very severe central nervous system involvement marked by cerebral atrophy and loss of myelination, such that even though their osteoclast defect might be rescued by HCT, their extremely short life expectancy limits the utility of this approach. On the other hand, *CLCN7* mutations of the chloride transporter are responsible for a wide range of clinical presentations within the infantile autosomal-recessive, "benign" autosomal-dominant, and intermediate forms of osteopetrosis. Thus, identification of a *CLCN7* mutation provides little information on how the disease will progress.

Management Guidelines

Baseline and monitoring studies

Laboratory evaluations

Once osteopetrosis has been diagnosed, we recommend obtaining minimum initial studies of serum calcium, intact parathyroid hormone, phosphorus, creatinine, 25-hydroxyvitamin D [25(OH)D], complete blood count with differential, creatine kinase isoenzymes (specifically the BB isozyme of creatine kinase), and lactate dehydrogenase (1|⊕⊕◯◯) (Table 1).

Serum calcium, phosphorus, 25(OH)D, and intact parathyroid hormone provide a measure of mineral homeostasis and can guide supplementation. A complete blood count with differential allows monitoring for evidence of bone marrow failure that may require transfusion, referral to a bone marrow transplant specialist, or other supportive measures.

In the absence of genetic testing or typical radiographic findings, elevated levels of lactate dehydrogenase, aspartate aminotransferase, tartrate-resistant acid phosphatase (not currently clinically available), as well as the BB isozyme of creatine kinase when normalized for age, have all been found to correlate with autosomal dominant osteopetrosis due to *CLCN7* defects but not other sclerosing bone diseases (9–13). Nevertheless, levels of these biomarkers have not been shown to correlate with disease severity or elucidate carrier status (11, 14), and normalcy does not rule out the presence of a mutation in *CLCN7*.

Imaging

In an effort to minimize radiation exposure, we recommend baseline magnetic resonance imaging of the brain to evaluate for cranial nerve involvement, hydrocephalus, and vascular abnormalities, followed by computed tomography if clinical findings of cranial nerve compression are identified (1|⊕⊕◯◯).

There is a high prevalence of optic nerve involvement reported in all forms of osteopetrosis (15, 16); the resultant visual impairment often has its onset early in life but can be variable. Although a patient who reportedly lost vision in the sixth decade of life has been briefly described (17), we (16) and others (18) have not observed visual impairment after growth cessation. Given the potential for progressive and severe disability, baseline neuroimaging with special attention to the optic foramina is warranted to enable early detection and treatment of these neurologic complications, which when present should be evaluated by an ophthalmologist every 6 months. The need for and frequency of subsequent imaging can then be guided by ophthalmologic findings.

We suggest that dual-energy x-ray absorptiometry (DXA) is not required as part of routine diagnosis or monitoring of disease progression (2|⊕⊕◯◯).

In patients with osteopetrosis, DXA reveals a markedly increased bone mineral density throughout the entire skeleton, especially at the lumbar spine, with increasing density with age pointing to the progressive nature of the disease (19). Unfortunately, despite its utility in the general population, in which low bone mineral density/osteoporosis is a risk factor for fracture, DXA cannot accurately predict the risk for fractures in patients with osteopetrosis (20). This limits its utility in this population.

Bone biopsy

We suggest that a bone biopsy is not useful except in exceptional situations, such as when there is a need to distinguish between osteoclast-poor and osteoclast-rich osteopetrosis (2|⊕⊕◯◯).

Osteoclasts are often present in normal or even increased numbers in bone specimens from patients with some disease-causing mutations but are absent in others. Bone biopsy can distinguish between osteoclast-poor and osteoclast-rich subtypes of osteopetrosis. Although this invasive and technically challenging study may have

Table 1. Evaluations for Screening and Monitoring Therapeutic Interventions in Patients With Osteopetrosis

Intervention	Timing
Serum calcium	At diagnosis and every 6–12 mo (every 3 mo during calcitriol therapy)
Serum phosphorus	At diagnosis and every 6–12 mo (every 3 mo during calcitriol therapy)
Serum creatinine	At diagnosis (every 3 mo during calcitriol therapy)
25(OH)D	At diagnosis and every 6–12 mo
Parathyroid hormone	At diagnosis and every 6–12 mo
Complete blood count with differential	At diagnosis and every 6–12 mo
BB isozyme of creatine kinase	At diagnosis
Lactate dehydrogenase	At diagnosis
MRI to evaluate optic nerves	At diagnosis and as clinically indicated
Urinary calcium/creatinine ratio (random or 24 h)	At initiation of calcitriol and every 3 mo during calcitriol therapy
Renal ultrasonography	At diagnosis and every 12 mo in patients with *CAII* mutations (every 12 mo during calcitriol therapy)

Abbreviation: MRI, magnetic resonance imaging.

guided treatment options in the previous era, it now offers little that genetic testing cannot provide and is therefore rarely performed.

Multidisciplinary approach

Optimal care of patients with osteopetrosis requires the involvement of a multidisciplinary team that, depending on mutation and clinical comorbidities, may include specialists in endocrinology, ophthalmology, genetics, dentistry, orthopedic surgery, neurology, neurosurgery, otolaryngology, hematology, infectious disease, nephrology, pain management, and developmental pediatrics (Table 2). We recommend evaluations for all patients by specialists in endocrinology, ophthalmology, genetics, and dentistry at the time of initial diagnosis (1|⊕⊕○○).

Endocrinology

The human skeleton as a reservoir comprises >99% of the total body calcium, which it can store or release under the tightly regulated influence of hormones, principally parathyroid hormone and vitamin D, to maintain serum levels within the physiologic range. In osteopetrosis, defective osteoclast activity renders the bone unable to mobilize its calcium pool and participate in normal calcium homeostasis, giving rise to hypocalcemia and resultant tetany and seizures. Paradoxically, despite the markedly positive total body calcium and phosphate balances in osteopetrosis, having mineral stores locked away in this manner also deprives the body of the substrate it needs to mineralize newly formed bone, which can result in rickets in autosomal-recessive osteopetrosis, otherwise aptly known in this setting as osteopetrorickets (21). Many of these complications pertaining to mineral balance can be effectively managed by the endocrinologist, who can direct calcium, phosphate, and vitamin D therapy, including the use of calcitriol when indicated, and monitor bone development.

Ophthalmology

Ocular complications are especially prevalent and severe in autosomal-recessive infantile osteopetrosis [affecting half of patients with a median age of 2 months at onset in one case series (15)] but do also frequently develop in patients with less severe forms of osteopetrosis as well (17, 22). The most common complication is optic nerve atrophy, resulting in visual impairment (22, 23). Although vision loss has in some cases been attributed to processes unrelated to bony changes, including retinal degeneration and impaired optic nerve myelination (24), the primary mechanism by which patients develop optic atrophy appears to be extrinsic nerve compression by the surrounding optic foramen (17, 24–27). A comprehensive ophthalmologic evaluation together with monitoring of visual evoked potentials can allow early detection of visual dysfunction, prompt necessary investigations, and facilitate timely intervention. Dedicated CT can evaluate patency of

Table 2. Complications by Subspecialty

Subspecialty	Complication
Endocrinology	Osteopetrorickets
	Hypocalcemia
Ophthalmology	Papilledema
	Ptosis
	Strabismus
	Paralysis of extraocular muscles
	Optic nerve atrophy
	Exophthalmos
	Nystagmus
	Retinal degeneration
	Tearing (from nasolacrimal duct obstruction)
Dentistry	Delay/failure of tooth eruption
	Malformed crowns/roots
	Periodontal ligament defects
	Odontoma
	Tooth agenesis
	Enamel hypoplasia
	Tooth decay/caries
	Thickened lamina dura
	Osteomyelitis (most frequently of the mandible)
Orthopedics	Skeletal deformities
	Scoliosis
	Spondylolisthesis
	Fractures (particularly of the long bones)
	Delayed union/nonunion
	Degenerative arthritis
	Spondylolysis
Neurology/ neurosurgery	Compressive cranial neuropathies (often optic and facial nerves, but can involve any of cranial nerves I–VIII)
	Increased intracranial pressure
	Craniosynostosis
	Arnold–Chiari I malformation
	Neuromuscular scoliosis
	Developmental delay/regression, seizures (*OSTM1* mutation)
	Calcifications of the basal ganglia, thalami (*CAII* deficiency)
	Hydrocephalus
	Cerebrovascular stenosis/occlusion
	Acquired encephalocele
Otolaryngology	Conductive hearing loss
	Recurrent otitis media
	Chronic congestion (poorly pneumatized sinuses)
	Rhinorrhea
	Choanal atresia
	Rhinosinusitis
	Obstructive sleep apnea
Hematology	Thrombocytopenia with bleeding
	Anemia
	Leukopenia with frequent infections
	Hepatosplenomegaly
	Transfusion dependence
Nephrology	Renal tubular acidosis, nephrocalcinosis, and nephrolithiasis (*CAII* deficiency)

the optic canals and confirm optic nerve entrapment, and electroretinography can determine whether pathology originates at the retina rather than the optic nerve (28, 29). It is important to note that many patients can be asymptomatic at diagnosis but have vision loss that is slowly progressive (2).

Dentistry

It is not surprising that many cases of osteopetrosis are first identified by dentists because the range of dental abnormality includes tooth agenesis, a delay or failure of tooth eruption, enamel hypoplasia, malformed crowns and roots, and tooth loss due to early decay and caries (Table 2) (30, 31). Osteomyelitis in osteopetrosis most frequently occurs in the mandible, where it typically follows odontogenic infections and oral procedures, may leave necrotic bone exposed, and is notoriously difficult to eradicate (32) because of a reduced blood supply and often accompanying anemia and neutropenia. Surgical debridement and antibiotics are the mainstays of therapy, whereas refractory cases may require hyperbaric oxygen to promote local healing and reperfusion (33). Tooth extraction should be approached judiciously because carious teeth can be precursors to osteomyelitis, but the operation itself can also induce infection, fractures, and other sequelae of poor healing, including irregular alveolar ridges (34). Routine dental surveillance and maintenance of proper oral hygiene by the dentist in this high-risk population is of utmost importance and can prevent, identify, and manage the myriad complications.

Orthopedics

When bone resorption is hampered and bone formation proceeds unopposed, as occurs in osteopetrosis, both the quantity and quality of bone suffer—total bone mass increases (35) but is distributed unevenly (20), and haphazard woven bone and calcified cartilage remnants of primary spongiosa can be seen histologically (36). The resulting fragility makes fracture a frequent problem for patients with osteopetrosis. Osteopetrotic bone fails under tensile stress (fractures occur at right angles to the cortex) rather than compressive stress (37), reflected in the fact that vertebral compression fractures have not been reported (38). Fractures are particularly common in autosomal-dominant CLCN7-deficient osteopetrosis and were the most common complication overall in two large case series (16, 18), routinely involving long bones, such as the proximal femur. The material properties of osteopetrotic bone make operative repair particularly challenging; the combined hardness and brittleness is resistant to drilling and prone to iatrogenic fracture, reduces the bone's ability to hold screws, and can necessitate creation of a medullary cavity for intramedullary fixation (39, 40). The postoperative course can be further complicated by defective remodeling, implant failure, and refracture risk; osteomyelitis can follow and often requires aggressive surgical debridement and en bloc resection. In some cases, fractures can be appropriately treated nonoperatively, although they should be monitored carefully for delayed union and nonunion as well as disabling deformities, including coxa vara and lateral bowing of the femur and long bones (38). For these reasons, the orthopedic surgeon should be consulted for any fractures and skeletal deformities.

Neurology and neurosurgery

The most prevalent neurologic complications of osteopetrosis are the compressive neuropathies that stem from progressive constriction of cranial foramina and/or the failure of these foramina to enlarge proportionately with growth. The optic nerve is disproportionately affected in patients with autosomal-recessive osteopetrosis, whereas the most frequent cranial nerve deficit in autosomal-dominant CLCN7-deficient osteopetrosis is the facial nerve (22, 41, 42), followed by the acoustic nerve, although any of cranial nerves I through VIII can be involved (43). Many of the cranial palsies can be subclinical (41) and are detected only on careful evaluation or after they have progressed. Optic nerve decompression (17, 25) and optic nerve sheath fenestration (44) in the appropriate clinical settings have been demonstrated to improve visual function and may better preserve visual acuity if performed early (45), although it is important to note that such positive outcomes are not universal (28, 46–48) and can be difficult to achieve in very young patients. Successful results have also been reported to follow decompression of the facial and acoustic nerves (49), although these operations are not without their risks and limitations (42).

Less commonly reported are the consequences of calvarial thickening reducing cranial capacity, which can include increased intracranial pressure, posterior fossa crowding, cerebellar tonsillar herniation, and hydrocephalus (45, 49). Case reports have also described cerebrovascular stenosis and occlusion (51–55), craniosynostosis (56, 57), and Arnold–Chiari I malformation (58, 59). Finally, neurologic disease is associated with some specific genetic mutations. Developmental delay or regression, unexplained seizures, and neuroradiologic findings should alert to the possibility of an *OSTM1* mutation. The cognitive defects of CAII deficiency are more variable, but the calcifications involving the basal ganglia, thalami, and/or gray-matter junction are almost always present (60). Altogether, the neurologist and neurosurgeon have a crucial part to play in recognizing and mitigating sources of disability in osteopetrotic

patients, and the latter can provide the necessary surgical intervention when indicated.

Otolaryngology

Extrinsic bony compression can lead to sensorineural deafness in a manner similar to the other cranial nerve palsies already discussed, but the hearing deficits seen in osteopetrosis can also arise from one of several conductive mechanisms, including recurrent otitis media, narrowing of the bony portion of the eustachian tube, obliteration of the round and oval windows, and immobilization or bony replacement of the ossicles (23, 49). The failure of bone modeling to enlarge bony cavities also applies to the midface, where narrowed and distorted nasal architecture along with poorly pneumatized sinuses contribute to the chronic nasal congestion with rhinorrhea and obligate mouth breathing (21, 61, 62) that can afflict patients with autosomal-recessive osteopetrosis; fortunately, some of these problems may improve with age (23). Although some areas of focus may overlap with those of other specialists, the otolaryngologist can provide essential expertise in addressing the many head and neck manifestations of osteopetrosis.

Hematology

Because of encroachment of osteopetrotic bone on the marrow cavity, the presence of hepatosplenomegaly and early myeloid cells in the periphery often indicate the development of extramedullary hematopoiesis. This encroachment eventually results in bone marrow failure in severe phenotypes and may also be seen to some degree in dominant and intermediate disease; profound anemia may be apparent, and thrombocytopenia with subsequent hemorrhage and infectious complications may prove life-threatening. Although a small minority of patients may recover sufficient hematopoiesis spontaneously (63), patients may become transfusion dependent. Early evidence has suggested that hypersplenic hemolysis may contribute to the anemia and thrombocytopenia, but splenectomy is not typically performed or recommended and corticosteroids have shown inconsistent results (see following section on Treatment). A reticulocyte count and blood smear may provide insight into whether anemia is a production problem or whether there is sequestration. Given the complexities that can be involved, it would be prudent to enlist the services of a hematologist when attempting to tackle refractory blood dyscrasias in patients with osteopetrosis.

Infectious disease

Patients with osteopetrosis, depending on the severity of their disease, are at variably increased risk for infections. Impaired vascularity, concomitant anemia and leukopenia, and impaired healing are among the factors at play. Osteomyelitis, particularly of the mandible but also of the long bones, is well reported in the literature and often is tenacious and difficult to treat. The activation of neutrophils and their ability to mount a respiratory burst response may also be impaired in autosomal-recessive osteopetrosis (64). The infectious disease specialist can therefore be a vital resource in coordinating appropriate antimicrobial therapy and overseeing chronic and recalcitrant infections.

Nephrology

Renal manifestations of osteopetrosis are not well documented, with the exception of those associated with CAII deficiency: renal tubular acidosis, nephrocalcinosis, and nephrolithiasis (65). The nephrologist can be consulted to monitor and address specific concerns as they arise in patients with known CAII mutations.

Treatments

Calcium and ergocalciferol/cholecalciferol

We recommend calcium and vitamin D as first-line therapy for the treatment of hypocalcemia and secondary hyperparathyroidism in patients with osteopetrosis (1|⊕⊕◯◯). We suggest that calcium supplementation be considered when dietary intake is below daily recommended dietary allowances (2|⊕⊕◯◯). We suggest that vitamin D (*i.e.*, ergocalciferol or cholecalciferol) be given to maintain 25(OH)D levels at >30 ng/mL (2|⊕⊕◯◯).

When the understanding of osteopetrosis was more limited, a strategy of limiting the amount of calcium available for mineralization was attempted in one case report that described use of a low-calcium diet (66); the addition of corticosteroids (for hematologic reasons) likely further interfered with gut absorption of calcium but together led to varying degrees of growth retardation. Calcium and vitamin D are frequently taken by patients with osteopetrosis, but their use outside osteopetrorickets is often not specifically discussed in the literature. No studies have addressed optimal calcium or vitamin D intake in patients with osteopetrosis. Given the many physiologic processes that depend on calcium and vitamin D sufficiency, their requirements in osteopetrosis should likely mirror those of the general population. A reasonable approach is to start standard age-appropriate doses of calcium and vitamin D replacement to meet recommended dietary allowances as outlined by the Institute of Medicine (67).

The optimal concentration of 25(OH)D for skeletal and nonskeletal benefits remains a point of contention among professional societies and scientific organizations. The American Academy of Pediatrics (68) and the

Institute of Medicine (67) have focused on the prevention of rickets and osteomalacia and propose a 25(OH)D concentration >20 ng/mL (50 nmol/L), whereas other groups, such as the Endocrine Society (69) and International Osteoporosis Foundation (70), suggest a minimum level of 30 ng/mL (75 nmol/L), citing evidence of a plateau in parathyroid hormone levels, fracture prevention, and improved intestinal calcium absorption corresponding to this target in the general population. In the absence of harm and specific correlations with fracture in the unique setting of osteopetrosis, we favor the latter approach of targeting a 25(OH)D level of ≥30 ng/mL (75 nmol/L).

Osteopetrorickets most typically develops in patients with *TCIRG1* mutations as a result of reduced gastric acidity, which in turn decreases calcium absorption; calcium supplementation may be sufficient to correct the bony and metabolic abnormalities in these patients. A case series of five osteopetrotic patients with biochemical (low calcium and phosphorus levels, high alkaline phosphatase for age) and radiographic findings of rickets (rachitic rosary, appendicular metaphyseal enlargement) together with baseline calcium–phosphorus products <30 mg^2/dL^2 (2.4 $mmol^2/L^2$) demonstrated resolution of rickets once the calcium–phosphorus product had risen above 40 mg^2/dL^2 (3.2 $mmol^2/L^2$) with calcium and vitamin D treatment (21).

Calcitriol

We suggest that calcitriol should not be used in high doses for treatment of osteopetrosis (2|⊕⊕○○).

Calcitriol is known to play a physiologic role in promoting the differentiation of osteoclasts from monocyte-macrophage stem cell precursors and in high doses can foster osteoclastic bone resorption by stimulating nuclear factor κ-B ligand production by osteoblasts. In 1984, Key *et al.* (71) first described the treatment of an infant diagnosed with severe recessive osteopetrosis by using high-dose calcitriol (titrated up to 32 μg/d) along with a low-calcium diet (to prevent hypercalcemia), resulting in improvements in measures of osteoclast and monocyte function. Despite these seemingly encouraging findings, the patient continued to deteriorate and calcitriol was ultimately withdrawn after 3 months because of poor neurologic prognosis. Subsequent case reports using high-dose calcitriol to stimulate bone resorption have similarly failed to demonstrate any meaningful clinical benefit in bone resorption or hematologic recovery (72–74). Animal studies have shown that not only does calcitriol not increase osteoclastic bone resorption (75, 76) but it may also have the opposite intended effect of increasing bone mass (77). Calcitriol at high doses has thus largely fallen out of favor given that any advantages are anecdotal at best.

Red blood cell transfusion

The decision for transfusion of red blood cells (RBCs) is recommended to be guided by the presence of symptomatic anemia as well as the hemoglobin level in consultation with a hematologist (1|⊕⊕○○).

RBC transfusions are not benign and can lead to several adverse outcomes. Any decision to transfuse should first trigger a clinical evaluation that accounts for such factors as baseline hemoglobin level, the rate of decline in hemoglobin level, intravascular volume status, existing cardiovascular conditions, and alternative therapies and should proceed only if the benefits outweigh the risks.

In hemodynamically stable adult patients in the inpatient and critical care settings, we agree with a restrictive threshold by which RBC transfusion is not indicated until the hemoglobin level is ≤7 g/dL. These recommendations are in line with those from multiple societal guidelines, including those from the AABB (formerly known as the American Association of Blood Banks) published in 2016 (78), which are, in turn, derived from now 31 randomized clinical trials examining hemoglobin thresholds for RBC transfusion demonstrating that a restrictive transfusion strategy is as safe as a liberal one that treats at hemoglobin levels of ≤10 g/dL. A randomized controlled trial in stable critically ill children drew similar conclusions and suggests that this threshold might apply to pediatric populations as well (79).

Interferon γ-1b

We suggest that the use of interferon γ-1b be considered experimental in noninfantile osteopetrosis (2|⊕⊕○○).

Early studies in autosomal-recessive osteopetrosis demonstrated a defect in nitroblue tetrazolium reduction and decreased intracellular bacterial killing among leukocytes (64, 80), suggesting that dysfunctional superoxide generation might contribute to the susceptibility to infections. Leukocytes and osteoclasts were recognized to arise from a common hematopoietic precursor, and subsequent studies found that osteoclasts also relied on superoxide generation to resorb bone (81) and that osteoclasts from an osteopetrotic patient exhibited similar deficits in oxygen radical formation (82). Breakthroughs from chronic granulomatous disease in which interferon γ-1b increased superoxide production and improved clinical outcomes led to trials of this agent in patients with autosomal-recessive osteopetrosis that revealed not only increases in leukocyte superoxide activity but also hematologic recovery and decreases in trabecular bone volume after up to 18 months of therapy (83, 84), although the desired benefits are not always seen (74). These findings culminated in the U.S. Food and Drug

Administration approval for the use of interferon γ-1b (Actimmune; Horizon Pharma, Lake Forest, IL) in severe malignant osteopetrosis in 2000. Of note, that indication pertains only to severe infantile osteopetrosis at this time. Clinical experience with this therapy remains limited; however, clinical trials are under way to better clarify the role of interferon γ-1b in the noninfantile osteopetrosis patient population.

Corticosteroids

Corticosteroid therapy may be considered as a second-line therapy to HCT in cases of severe infantile osteopetrosis in whom HCT is determined to not be appropriate (2|⊕⊕◯◯).

A reduced RBC survival time (85), excess of nucleated RBCs, reticulocytosis, and absence of hemolyzing antibodies (86) in early studies seemed to implicate a hemolytic anemia (87) from hypersplenism in autosomal-recessive osteopetrosis, for which trials of corticosteroids (26, 66) and splenectomy (66, 86) benefited the anemia and thrombocytopenia in some but not all cases. Perhaps the most promising evidence comes from two case reports of infantile osteopetrosis, followed for 3 months and 3 years, treated in the newborn period with prednisone; spontaneous remission occurred, obviating the need for HCT (although mutation analysis does not appear to have been done) (88, 89). Nevertheless, the mechanisms by which corticosteroids rectify the hematologic abnormalities have never been fully understood, and other studies have shown a variable response at best and no alteration in the natural course of the disease (29, 90). In addressing the bone pathology, corticosteroids have a known osteoporotic effect by directly interfering with mineralization of bone and cartilage (91) and intestinal calcium absorption (90, 92). However, given the present knowledge of the primarily osteoclastic acidification defects that generate the disease phenotype, the mineralization deficiencies that already exist with impaired turnover of bone, the generally transient effects, and the potential for adverse events, including growth delay, there is insufficient evidence to support the routine use of corticosteroids in osteopetrosis at this time.

Referral for HCT

Bone marrow failure and age <1 year at diagnosis are primary indications to refer to a specialized center with experience in HCT treatment of patients with osteopetrosis. Genetic testing may reveal specific mutations that favor or preclude (*e.g., OSTM1* or *RANKL* mutations) bone marrow transplantation. Neurologic involvement and unremitting pain may be reasonable indications to warrant an evaluation as well.

A detailed discussion of HCT is outside the scope of these guidelines; however, a few key points merit emphasis. Historically the best outcomes have been achieved by using a genotypically HLA-identical bone marrow donor (93), which is not commonly available. Serious limitations include high mortality rates within the first year (93, 94). Importantly, HCT is an option only for mutations intrinsic to the osteoclast and remains experimental for intermediate osteopetrosis at this time. Each individual case warrants careful consideration of the risks vs benefits through consultation with an experienced HCT physician before proceeding with transplantation.

Conclusions/Future Directions

Scarcity of published studies on noninfantile osteopetrosis reduces the ability to develop evidence-based guidelines for the clinical management of these patients. Despite this limitation, the current effort is important to highlight the evidence that does exist and how it can be used within the medical community in a fashion that, although not completely evidence based, is at least consistent with known physiologic principles.

Studies on disease progression and effects of current supportive therapies are desperately needed. Despite an extensive literature review, most of our recommendations are based solely on expert opinion and, in some areas, small case series. A multicenter, longitudinal study of a large cohort of patients with noninfantile osteopetrosis is needed to better define disease progression, quantify response to current supportive therapies, and provide natural history data to be used in future therapeutic clinical trials. In addition, because noninfantile osteopetrosis has no cure, future research should also focus on curative therapies for this devastating disease.

Acknowledgments

Address all correspondence and requests for reprints to: Lynda E. Polgreen, MD, MS, David Geffen School of Medicine–UCLA, Pediatric Endocrinology, Los Angeles Biomedical Research Institute at Harbor-UCLA, 1124 W. Carson Street, Liu Research Building, Torrance, California 90502. E-mail: lpolgreen@labiomed.org.

The Osteopetrosis Working Group meetings were supported by an educational grant from Horizon Pharma. L.E.P. and M.J.E. are supported by research grants from Horizon Pharma. L.M.W. is supported by a University of Ottawa Research Chair Award in Pediatric Bone Health.

Disclosure Summary: The authors have nothing to disclose.

References

1. Tolar J, Teitelbaum SL, Orchard PJ. Osteopetrosis. *N Engl J Med*. 2004;**351**(27):2839–2849.
2. Johnston CC Jr, Lavy N, Lord T, Vellios F, Merritt AD, Deiss WP Jr. Osteopetrosis. A clinical, genetic, metabolic, and morphologic

study of the dominantly inherited, benign form. *Medicine (Baltimore)*. 1968;**47**(2):149–167.
3. Bollerslev J, Andersen PE Jr. Radiological, biochemical and hereditary evidence of two types of autosomal dominant osteopetrosis. *Bone*. 1988;**9**(1):7–13.
4. Campos-Xavier AB, Casanova J-L, Doumaz Y, Feingold J, Munnich A, Cormier-Daire V. Intrafamilial phenotypic variability of osteopetrosis due to chloride channel 7 (CLCN7) mutations. *Am J Med Genet A*. 2005;**133A**(2):216–218.
5. Schulz A, Moushous D, Steward CG, Villa A, Sobacchi C. Osteopetrosis: consensus guidelines for diagnosis, therapy and follow-up. 2015. Available at: https://www.ebmt.org/Contents/Research/TheWorkingParties/IEWP/Documents/00_OP_Guidelines_V3.pdf. Accessed 16 May 2017.
6. Swiglo BA, Murad MH, Schünemann HJ, Kunz R, Vigersky RA, Guyatt GH, Montori VM. A case for clarity, consistency, and helpfulness: state-of-the-art clinical practice guidelines in endocrinology using the grading of recommendations, assessment, development, and evaluation system. *J Clin Endocrinol Metab*. 2008;**93**(3):666–673.
7. Faden MA, Krakow D, Ezgu F, Rimoin DL, Lachman RS. The Erlenmeyer flask bone deformity in the skeletal dysplasias. *Am J Med Genet A*. 2009;**149A**(6):1334–1345.
8. Sobacchi C, Schulz A, Coxon FP, Villa A, Helfrich MH. Osteopetrosis: genetics, treatment and new insights into osteoclast function. *Nat Rev Endocrinol*. 2013;**9**(9):522–536.
9. Yoneyama T, Fowler HL, Pendleton JW, Sforza PP, Lui CY, Iranmanesh A, Gerard RD. Elevated levels of creatine kinase BB isoenzyme in three patients with adult osteopetrosis. *N Engl J Med*. 1989;**320**(19):1284–1285.
10. Yoneyama T, Fowler HL, Pendleton JW, Sforza PP, Gerard RD, Lui CY, Eldridge TH, Iranmanesh A. Elevated serum levels of creatine kinase BB in autosomal dominant osteopetrosis type II—a family study. *Clin Genet*. 1992;**42**(1):39–42.
11. Waguespack SG, Hui SL, White KE, Buckwalter KA, Econs MJ. Measurement of tartrate-resistant acid phosphatase and the brain isoenzyme of creatine kinase accurately diagnoses type II autosomal dominant osteopetrosis but does not identify gene carriers. *J Clin Endocrinol Metab*. 2002;**87**(5):2212–2217.
12. Whyte MP, Chines A, Silva DP Jr, Landt Y, Ladenson JH. Creatine kinase brain isoenzyme (BB-CK) presence in serum distinguishes osteopetroses among the sclerosing bone disorders. *J Bone Miner Res*. 1996;**11**(10):1438–1443.
13. Whyte MP, Kempa LG, McAlister WH, Zhang F, Mumm S, Wenkert D. Elevated serum lactate dehydrogenase isoenzymes and aspartate transaminase distinguish Albers-Schönberg disease (chloride channel 7 deficiency osteopetrosis) among the sclerosing bone disorders. *J Bone Miner Res*. 2010;**25**(11):2515–2526.
14. Takacs I, Cooper H, Weaver DD, Econs MJ. Bone mineral density and laboratory evaluation of a type II autosomal dominant osteopetrosis carrier. *Am J Med Genet*. 1999;**85**(1):9–12.
15. Gerritsen EJ, Vossen JM, van Loo IH, Hermans J, Helfrich MH, Griscelli C, Fischer A. Autosomal recessive osteopetrosis: variability of findings at diagnosis and during the natural course. *Pediatrics*. 1994;**93**(2):247–253.
16. Waguespack SG, Hui SL, Dimeglio LA, Econs MJ. Autosomal dominant osteopetrosis: clinical severity and natural history of 94 subjects with a chloride channel 7 gene mutation. *J Clin Endocrinol Metab*. 2007;**92**(3):771–778.
17. Haines SJ, Erickson DL, Wirtschafter JD. Optic nerve decompression for osteopetrosis in early childhood. *Neurosurgery*. 1988;**23**(4):470–475.
18. Bénichou OD, Laredo JD, de Vernejoul MC. Type II autosomal dominant osteopetrosis (Albers-Schönberg disease): clinical and radiological manifestations in 42 patients. *Bone*. 2000;**26**(1):87–93.
19. Grodum E, Gram J, Brixen K, Bollerslev J. Autosomal dominant osteopetrosis: bone mineral measurements of the entire skeleton of adults in two different subtypes. *Bone*. 1995;**16**(4):431–434.
20. Arruda M, Coelho MCA, Moraes AB, de Paula Paranhos-Neto F, Madeira M, Farias MLF, Vieira Neto L. Bone mineral density and microarchitecture in patients with autosomal dominant osteopetrosis: a report of two cases. *J Bone Miner Res*. 2016;**31**(3):657–662.
21. Kaplan FS, August CS, Fallon MD, Gannon F, Haddad JG. Osteopetrorickets. The paradox of plenty. Pathophysiology and treatment. *Clin Orthop Relat Res*. 1993; (294):64–78.
22. Steward CG. Neurological aspects of osteopetrosis. *Neuropathol Appl Neurobiol*. 2003;**29**(2):87–97.
23. Wong ML, Balkany TJ, Reeves J, Jafek BW. Head and neck manifestations of malignant osteopetrosis. *Otolaryngology*. 1978;**86**(4 Pt 1):ORL-585–ORL-594.
24. Vrabec F, Sedlackova J. Neuro-histological Findings in Osteopetrosis (Albers-Schoenberg Disease). *Br J Ophthalmol*. 1964;**48**:218–222.
25. Al-Mefty O, Fox JL, Al-Rodhan N, Dew JH. Optic nerve decompression in osteopetrosis. *J Neurosurg*. 1988;**68**(1):80–84.
26. Moe PJ, Skjaeveland A. Therapeutic studies in osteopetrosis. Report of 4 cases. *Acta Paediatr Scand*. 1969;**58**(6):593–600.
27. Cummings TJ, Proia AD. Optic nerve compression in infantile malignant autosomal recessive osteopetrosis. *J Pediatr Ophthalmol Strabismus*. 2004;**41**(4):241–244.
28. Hoyt CS, Billson FA. Visual loss in osteopetrosis. *Am J Dis Child*. 1979;**133**(9):955–958.
29. Keith CG. Retinal atrophy in osteopetrosis. *Arch Ophthalmol*. 1968;**79**(3):234–241.
30. Dick HM, Simpson WJ. Dental changes in osteopetrosis. *Oral Surg Oral Med Oral Pathol*. 1972;**34**(3):408–416.
31. Droz-Desprez D, Azou C, Bordigoni P, Bonnaure-Mallet M. Infantile osteopetrosis: a case report on dental findings. *J Oral Pathol Med*. 1992;**21**(9):422–425.
32. Steiner M, Gould AR, Means WR. Osteomyelitis of the mandible associated with osteopetrosis. *J Oral Maxillofac Surg*. 1983;**41**(6):395–405.
33. Mainous EG, Hart GB, Soffa DJ, Graham GA. Hyperbaric oxygen treatment of mandibular osteomyelitis in osteopetrosis. *J Oral Surg Am Dent Assoc*. 1975;**33**(4):288–291.
34. Bansal V, Kumar S, Arunkumar KV, Mowar A, Khare G. Dental management in autosomal recessive (intermediate) osteopetrosis: a case report. *Pediatr Dent*. 2010;**32**(7):542–545.
35. Kovanlikaya A, Loro ML, Gilsanz V. Pathogenesis of osteosclerosis in autosomal dominant osteopetrosis. *AJR Am J Roentgenol*. 1997;**168**(4):929–932.
36. Helfrich MH, Aronson DC, Everts V, Mieremet RH, Gerritsen EJ, Eckhardt PG, Groot CG, Scherft JP. Morphologic features of bone in human osteopetrosis. *Bone*. 1991;**12**(6):411–419.
37. de Palma L, Tulli A, Maccauro G, Sabetta SP, del Torto M. Fracture callus in osteopetrosis. *Clin Orthop Relat Res*. 1994; (308):85–89.
38. Shapiro F. Osteopetrosis. Current clinical considerations. *Clin Orthop Relat Res*. 1993; (294):34–44.
39. Chhabra A, Westerlund LE, Kline AJ, McLaughlin R. Management of proximal femoral shaft fractures in osteopetrosis: a case series using internal fixation. *Orthopedics*. 2005;**28**(6):587–592.
40. Landa J, Margolis N, Di Cesare P. Orthopaedic management of the patient with osteopetrosis. *J Am Acad Orthop Surg*. 2007;**15**(11):654–662.
41. Bollerslev J, Grøntved A, Andersen PE Jr. Autosomal dominant osteopetrosis: an otoneurological investigation of the two radiological types. *Laryngoscope*. 1988;**98**(4):411–413.
42. Benecke JE Jr. Facial nerve dysfunction in osteopetrosis. *Laryngoscope*. 1993;**103**(5):494–497.
43. Klintworth GK. The neurologic manifestations of osteopetrosis (Albers-Schonberg's disease). *Neurology*. 1963;**13**:512–519.
44. Allen RC, Nerad JA, Kattah JC, Lee AG. Resolution of optic nerve edema and improved visual function after optic nerve sheath fenestration in a patient with osteopetrosis. *Am J Ophthalmol*. 2006;**141**(5):945–947.
45. Al-Tamimi YZ, Tyagi AK, Chumas PD, Crimmins DW. Patients with autosomal-recessive osteopetrosis presenting with hydrocephalus and

hindbrain posterior fossa crowding. *J Neurosurg Pediatr*. 2008;**1**(1):103–106.
46. Gerritsen EJ, Vossen JM, Fasth A, Friedrich W, Morgan G, Padmos A, Vellodi A, Porras O, O'Meara A, Porta F, Cant A, Hermans J. Bone marrow transplantation for autosomal recessive osteopetrosis. A report from the Working Party on Inborn Errors of the European Bone Marrow Transplantation Group. *J Pediatr*. 1994;**125**(6 Pt 1):896–902.
47. Ellis PP, Jackson WE. Osteopetrosis. A clinical study of optic-nerve involvement. *Am J Ophthalmol*. 1962;**53**:943–953.
48. Ainsworth JR, Bryce IG, Dudgeon J. Visual loss in infantile osteopetrosis. *J Pediatr Ophthalmol Strabismus*. 1993;**30**(3):201–203.
49. Miyamoto RT, House WF, Brackmann DE. Neurotologic manifestations of the osteopetroses. *Arch Otolaryngol*. 1980;**106**(4):210–214.
50. Scott WW, Weprin BE, Swift DM. A unifying theory for the multifactorial origin of cerebellar tonsillar herniation and hydrocephalus in osteopetrosis. *J Neurosurg Pediatr*. 2014;**14**(6):665–673.
51. Wilms G, Casaer P, Alliet P, Demaerel P, Smet M, Baert AL. Cerebrovascular occlusive complications in osteopetrosis major. *Neuroradiology*. 1990;**32**(6):511–513.
52. Curé JK, Key LL, Shankar L, Gross AJ. Petrous carotid canal stenosis in malignant osteopetrosis: CT documentation with MR angiographic correlation. *Radiology*. 1996;**199**(2):415–421.
53. Tasdemir HA, Dagdemir A, Celenk C, Albayrak D. Middle cerebral arterial occlusion in a child with osteopetrosis major. *Eur Radiol*. 2001;**11**(1):145–147.
54. Makin GJ, Coates RK, Pelz D, Drake CG, Barnett HJ. Major cerebral arterial and venous disease in osteopetrosis. *Stroke*. 1986;**17**(1):106–110.
55. Curé JK, Key LL, Goltra DD, VanTassel P. Cranial MR imaging of osteopetrosis. *AJNR Am J Neuroradiol*. 2000;**21**(6):1110–1115.
56. Krimmel M, Niemann G, Will B, Reinert S. Surgical correction of craniosynostosis in malignant osteopetrosis. *J Craniofac Surg*. 2004;**15**(2):218–220, discussion 221.
57. Dowlati D, Winston KR, Ketch LL, Quinones R, Giller R, Frattini A, van Hove J. Expansion cranioplasty with jackscrew distracters for craniosynostosis and intracranial hypertension in transplanted osteopetrosis. *Pediatr Neurosurg*. 2007;**43**(2):102–106.
58. Dlouhy BJ, Menezes AH. Osteopetrosis with Chiari I malformation: presentation and surgical management. *J Neurosurg Pediatr*. 2011;**7**(4):369–374.
59. Kulkarni ML, Marakkanavar SN, Sushanth S, Pradeep N, Ashok C, Balaji MD, Sajith KP, Srinivas G. Osteopetrosis with Arnold Chiari malformation type I and brain stem compression. *Indian J Pediatr*. 2007;**74**(4):412–415.
60. Bosley TM, Salih MA, Alorainy IA, Islam MZ, Oystreck DT, Suliman OSM, al Malki S, Suhaibani AH, Khiari H, Beckers S, van Wesenbeeck L, Perdu B, AlDrees A, Elmalik SA, Van Hul W, Abu-Amero KK. The neurology of carbonic anhydrase type II deficiency syndrome. *Brain*. 2011;**134**(Pt 12):3502–3515.
61. Bartynski WS, Barnes PD, Wallman JK. Cranial CT of autosomal recessive osteopetrosis. *AJNR Am J Neuroradiol*. 1989;**10**(3):543–550.
62. Elster AD, Theros EG, Key LL, Chen MY. Cranial imaging in autosomal recessive osteopetrosis. Part I. Facial bones and calvarium. *Radiology*. 1992;**183**(1):129–135.
63. Totan M, Albayrak D. Osteopetrosis—improvement of hematologic findings with age. *Indian J Pediatr*. 1999;**66**(5):809–812.
64. Beard CJ, Key L, Newburger PE, Ezekowitz RA, Arceci R, Miller B, Proto P, Ryan T, Anast C, Simons ER. Neutrophil defect associated with malignant infantile osteopetrosis. *J Lab Clin Med*. 1986;**108**(5):498–505.
65. Ismail EA, Abul Saad S, Sabry MA. Nephrocalcinosis and urolithiasis in carbonic anhydrase II deficiency syndrome. *Eur J Pediatr*. 1997;**156**(12):957–962.
66. Dent CE, Smellie JM, Watson L. Studies in osteopetrosis. *Arch Dis Child*. 1965;**40**:7–15.
67. Ross AC, Taylor CL, Yaktine AL, Del Valle HB, eds. 2011 DRI dietary reference intakes for calcium and vitamin D. Institute of Medicine. 2011. Available at: https://www.nap.edu/read/13050. Accessed 16 May 2017.
68. Wagner CL, Greer FR; American Academy of Pediatrics Section on Breastfeeding; American Academy of Pediatrics Committee on Nutrition. Prevention of rickets and vitamin D deficiency in infants, children, and adolescents. *Pediatrics*. 2008;**122**(5):1142–1152.
69. Holick MF, Binkley NC, Bischoff-Ferrari HA, Gordon CM, Hanley DA, Heaney RP, Murad MH, Weaver CM; Endocrine Society. Evaluation, treatment, and prevention of vitamin D deficiency: an Endocrine Society clinical practice guideline. *J Clin Endocrinol Metab*. 2011;**96**(7):1911–1930.
70. Dawson-Hughes B, Mithal A, Bonjour J-P, Boonen S, Burckhardt P, Fuleihan GE-H, Josse RG, Lips P, Morales-Torres J, Yoshimura N. IOF position statement: vitamin D recommendations for older adults. *Osteoporos Int*. 2010;**21**(7):1151–1154.
71. Key L, Carnes D, Cole S, Holtrop M, Bar-Shavit Z, Shapiro F, Arceci R, Steinberg J, Gundberg C, Kahn A, et al. Treatment of congenital osteopetrosis with high-dose calcitriol. *N Engl J Med*. 1984;**310**(7):409–415.
72. van Lie Peters EM, Aronson DC, Everts V, Dooren LJ. Failure of calcitriol treatment in a patient with malignant osteopetrosis. *Eur J Pediatr*. 1993;**152**(10):818–821.
73. Blazar BR, Fallon MD, Teitelbaum SL, Ramsay NK, Brown DM. Calcitriol for congenital osteopetrosis. *N Engl J Med*. 1984;**311**(1):55.
74. Kubo T, Tanaka H, Ono H, Moriwake T, Kanzaki S, Seino Y. Malignant osteopetrosis treated with high doses of 1 alpha-hydroxyvitamin D3 and interferon gamma. *J Pediatr*. 1993;**123**(2):264–268.
75. Safadi FF, Hermey DC, Popoff SN, Seifert MF. Skeletal resistance to 1,25-dihydroxyvitamin D3 in osteopetrotic rats. *Endocrine*. 1999;**11**(3):309–319.
76. Popoff SN, McGuire JL, Zerwekh JE, Marks SC Jr. Treatment of congenital osteopetrosis in the rabbit with high-dose 1,25-dihydroxyvitamin D. *J Bone Miner Res*. 1989;**4**(1):57–67.
77. Alam I, Gray AK, Acton D, Gerard-O'Riley RL, Reilly AM, Econs MJ. Interferon Gamma, but not Calcitriol Improves the Osteopetrotic Phenotypes in ADO2 Mice. *J Bone Miner Res*. 2015;**30**(11):2005–2013.
78. Carson JL, Guyatt G, Heddle NM, Grossman BJ, Cohn CS, Fung MK, Gernsheimer T, Holcomb JB, Kaplan LJ, Katz LM, Peterson N, Ramsey G, Rao SV, Roback JD, Shander A, Tobian AAR. Clinical practice guidelines from the AABB: red blood cell transfusion thresholds and storage. *JAMA*. 2016;**316**(19):2025–2035.
79. Lacroix J, Hébert PC, Hutchison JS, Hume HA, Tucci M, Ducruet T, Gauvin F, Collet J-P, Toledano BJ, Robillard P, Joffe A, Biarent D, Meert K, Peters MJ; TRIPICU Investigators; Canadian Critical Care Trials Group; Pediatric Acute Lung Injury and Sepsis Investigators Network. Transfusion strategies for patients in pediatric intensive care units. *N Engl J Med*. 2007;**356**(16):1609–1619.
80. Reeves JD, August CS, Humbert JR, Weston WL. Host defense in infantile osteopetrosis. *Pediatrics*. 1979;**64**(2):202–206.
81. Key LL Jr, Ries WL, Taylor RG, Hays BD, Pitzer BL. Oxygen derived free radicals in osteoclasts: the specificity and location of the nitroblue tetrazolium reaction. *Bone*. 1990;**11**(2):115–119.
82. Key LL Jr, Ries WL. Osteopetrosis. The pharmaco-physiologic basis of therapy. *Clin Orthop Relat Res*. 1993; (294):85–89.
83. Key LL Jr, Ries WL, Rodriguiz RM, Hatcher HC. Recombinant human interferon gamma therapy for osteopetrosis. *J Pediatr*. 1992;**121**(1):119–124.
84. Key LL Jr, Rodriguiz RM, Willi SM, Wright NM, Hatcher HC, Eyre DR, Cure JK, Griffin PP, Ries WL. Long-term treatment of osteopetrosis with recombinant human interferon gamma. *N Engl J Med*. 1995;**332**(24):1594–1599.

85. Zetterstrom R. Osteopetrosis (marble bone disease); clinical and pathological review. *Bibl Paediatr*. 1958;**14**(66):488–508.
86. Sjolin S. Studies on osteopetrosis. II. Investigations concerning the nature of the anaemia. *Acta Paediatr*. 1959;**48**:529–544.
87. Gamsu H, Lorber J, Rendle-Short J. Haemolytic anaemia in osteopetrosis. A report of two cases. *Arch Dis Child*. 1961;**36**: 494–499.
88. Iacobini M, Migliaccio S, Roggini M, Taranta A, Werner B, Panero A, Teti A. Apparent cure of a newborn with malignant osteopetrosis using prednisone therapy. *J Bone Miner Res*. 2001;**16**(12): 2356–2360.
89. Colarizi P, Fiorucci P, Roggini M, Papoff P. Circulating thrombopoietin levels in a neonate with osteopetrosis. *Pediatrics*. 1999; **103**(3):700–701.
90. Del Fattore A, Peruzzi B, Rucci N, Recchia I, Cappariello A, Longo M, Fortunati D, Ballanti P, Iacobini M, Luciani M, Devito R, Pinto R, Caniglia M, Lanino E, Messina C, Cesaro S, Letizia C, Bianchini G, Fryssira H, Grabowski P, Shaw N, Bishop N, Hughes D, Kapur RP, Datta HK, Taranta A, Fornari R, Migliaccio S, Teti A. Clinical, genetic, and cellular analysis of 49 osteopetrotic patients: implications for diagnosis and treatment. *J Med Genet*. 2006;**43**(4): 315–325.
91. Silbermann M, Maor G. Mechanisms of glucocorticoid-induced growth retardation: impairment of cartilage mineralization. *Acta Anat (Basel)*. 1978;**101**(2):140–149.
92. Hahn TJ, Halstead LR, Baran DT. Effects off short term glucocorticoid administration on intestinal calcium absorption and circulating vitamin D metabolite concentrations in man. *J Clin Endocrinol Metab*. 1981;**52**(1):111–115.
93. Orchard PJ, Fasth AL, Le Rademacher J, He W, Boelens JJ, Horwitz EM, Al-Seraihy A, Ayas M, Bonfim CM, Boulad F, Lund T, Buchbinder DK, Kapoor N, O'Brien TA, Perez MAD, Veys PA, Eapen M. Hematopoietic stem cell transplantation for infantile osteopetrosis. *Blood*. 2015;**126**(2):270–276.
94. Martinez C, Polgreen LE, DeFor TE, Kivisto T, Petryk A, Tolar J, Orchard PJ. Characterization and management of hypercalcemia following transplantation for osteopetrosis. *Bone Marrow Transplant*. 2010;**45**(5):939–944.