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Authors

Dorsey, Morna J

Dvorak, Christopher C

Cowan, Morton J

et al.

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Treatment of Infants Identified by Newborn Screening for Severe Combined Immunodeficiency

Morna J. Dorsey, MD, MMSc, **Christopher C. Dvorak, MD**, **Morton J. Cowan, MD**, and **Jennifer M. Puck, MD**

Department of Pediatrics, Division of Allergy, Immunology, and Bone Marrow Transplantation, University of California, San Francisco

Abstract

Background—Severe combined immunodeficiency (SCID) is characterized by severely impaired T cell development and is fatal without treatment. Newborn screening (NBS) for SCID permits identification of affected infants before development of opportunistic infections and other complications. Substantial variation exists between treatment centers with regard to pre-transplant care and transplant protocols for NBS identified SCID infants, as well as for infants with other T lymphopenic disorders detected by NBS.

Methods—We developed approaches to management based on the study of infants identified by SCID NBS who received care at UCSF.

Results—From August 2010 through October 2016, 32 NBS SCID and leaky SCID cases from California and other states were treated and 42 NBS identified non-SCID T cell lymphopenia (TCL) cases were followed.

Conclusions—Our center's approach supports successful outcomes; systematic review of our practice provides a framework for diagnosis and management, recognizing that more data will continue to shape best practices.

Introduction

Severe combined immunodeficiency (SCID) is a genetically heterogeneous group of inherited defects characterized by severely impaired T cell development combined with inability to make specific antibodies [1–4]. While fatal without treatment, SCID is treatable by allogeneic hematopoietic cell transplantation (HCT), or in certain genotypes by enzyme

Corresponding Author: Morna J. Dorsey, Department of Pediatrics, Division of Allergy, Immunology, and Bone Marrow Transplantation, University of California, San Francisco, 550 16th Street, 4th Floor, Box 0434, San Francisco CA 94143; morna.dorsey@ucsf.edu.

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replacement (ERT) or gene therapy (GT). Unless diagnosed in the neonatal period, affected infants develop severe, opportunistic infections early in life. Avoidance of infection was the impetus for population-based SCID newborn screening (NBS) using newborn dried blood spots (DBS). All infants with SCID fail to generate a diverse repertoire of mature T cells, and consequently have absent or very low numbers of T cell receptor excision circles (TRECs), DNA byproducts of T cell receptor gene rearrangement [5,6]. An exception is late-onset adenosine deaminase (ADA) SCID where progressive loss of T cells occurs with time. Newborn screening for insufficient TRECs makes possible identification of SCID before infections occur, permitting optimal care of affected infants.

As SCID NBS has become widespread, and in conjunction with establishment of the Primary Immune Deficiency Consortium (PIDTC) funded by the National Institute of Allergy and Infectious Diseases and Office of Rare Diseases, National Center for Advancing Translational Sciences, NIH, new definitions for SCID have evolved for healthy-appearing affected infants [3, 4, 13]. In contrast to classical descriptions of SCID with infections [1], the new criteria are based upon laboratory parameters. Typical SCID cases have <300 autologous CD3 T cells/uL, <10% of the lower range of normal proliferation to the mitogen phytohemmaglutinin (PHA), and/or detectable maternal T cell engraftment as well as deleterious mutations in recognized SCID genes (Figure 1). In addition to typical SCID, TREC screening identifies “leaky” SCID due to hypomorphic mutations in known SCID genes; 26% of the SCID cases found by screening were leaky, as reported in an 11-program study of NBS for SCID in the US [1]. Leaky SCID cases have 300–1500 T cells/uL or more, but lack naïve CD4 T cells expressing CD45RA. Their T cells are functionally impaired and have limited diversity, and maternal cells are not detected. A subset of infants with leaky SCID have expansion of oligoclonal, dysregulated T cells leading to adenopathy, erythroderma with cutaneous and intestinal T cell infiltration, hepatosplenomegaly, eosinophilia, and highly elevated IgE, features collectively known as Omenn syndrome (OS) [3].

NBS also identifies infants with low TRECs who do not have SCID, but who nonetheless have few T lymphocytes in the peripheral blood, termed T cell lymphopenia (TCL) [3,15]. While most of these infants have recognized conditions, such as DiGeorge syndrome, others have TCL with no apparent underlying cause [10,11]. Establishing a definitive diagnosis and managing these infants are new challenges for physicians, who must recognize the level of TCL that is medically significant, select diagnostic tests, and apply appropriate interventions.

We provide here our center’s approach to infants with SCID, leaky SCID, and non-SCID TCL identified by NBS, recognizing that individual state screening programs and providers currently employ a spectrum of approaches [3] and that more data are needed to identify best practices.

Approach to infants with TREC results that are not normal

In California, all infants with non-normal TREC tests immediately have a CBC, differential and flow cytometry analysis to distinguish the 43% of infants with confirmed TCL from the

57% with >1500 T cells/uL, for whom no further intervention is undertaken as long as naïve T cells are observed (Figure 1, right) [11,12,15]. Infants with <300 T cells/uL, or with more T cells/uL but <200 naïve CD4 T cells, representing ~16% of all those with non-normal TREC results (Figure 1, left), are immediately hospitalized due to the high likelihood of having typical or leaky SCID, respectively, or complete DiGeorge syndrome. The management of SCID cases at our center is discussed below. The remaining infants with 300–1500 T cells/uL, but with naïve CD4 T cells present (Figure 1, middle right), have multisystem syndromes, secondary, and undiagnosed or idiopathic TCL. Diagnosis and management of infants with non-SCID TCL is undertaken on a case-by-case basis, and generally includes (in addition to flow cytometry), proliferative responses to mitogens, serum immunoglobulin levels, and DNA arrays to reveal genomic copy number variation.

How we treat the infant with suspected SCID

Infants suspected to have SCID or leaky SCID based upon laboratory testing (and those identified by a family history or clinical features), are admitted to protective isolation in our hospital; any sign of infection or other abnormality is immediately pursued with cultures and treated (Table 1). Our immune evaluation is listed in Table 2. Early discussions between the family and the immunology and HCT teams present information about SCID, the diagnostic approach, and expectations for hospitalization. The HCT team works with the family to identify the optimal donor. A social worker versed in SCID is involved to identify needs for support. Infectious disease workup includes assessing for cytomegalovirus (CMV) by PCR, which may be transmitted via breast milk of CMV seropositive mothers prior to confirmation of TCL. Two infants in our 32-patient cohort (6%) were infected by CMV at baseline, with breast milk from a CMV seropositive mother the only identified potential source. Baseline lymphocyte subset confirmation and functional testing are obtained, along with HLA typing of the infant and family members to identify potential HCT donors. Evidence of maternal T cell engraftment is sought by analysis of maternal and infant DNA; maternal chimerism, detected in >50% of SCID cases [14], suggests that a maternal graft can be accepted without conditioning. An infant with CD45RO, but not CD45RA, T cells may have expansion of a limited repertoire of T cells of either autologous or maternal origin. A T-B-NK- lymphocyte profile suggests ADA deficiency, often accompanied by neutropenia; screening for ADA enzyme activity and metabolite levels is quickly accomplished before pursuing gene sequencing.

Special considerations

Iatrogenic anemia

Iatrogenic anemia is a concern in small infants with SCID. Blood draws are planned to minimize discomfort and use the smallest possible volume for each test. Transfusions are avoided to minimize risks of infection and allosensitization. If transfusions are needed, only CMV negative, irradiated, leukoreduced packed red cells are given. A standard operating procedure developed with our clinical laboratory uses specific minimum volumes for every test (Appendix 1).

Adenosine deaminase (ADA) deficiency

ADA-SCID, diagnosed in 6 of our initial 32 SCID infants (19%), is a systemic metabolic disorder in which elevated concentrations of purine intermediates are particularly toxic to lymphocytes, but affect all tissues. Early complications of ADA deficiency included neutropenia, seen in 4 of our 6 ADA infants, and pulmonary alveolar proteinosis (PAP) seen in 2, one of whom required intubation for respiratory distress. Two others with tachypnea at diagnosis were suspected to have PAP. Bronchoscopy to obtain cultures as well as histology is required to distinguishable PAP from infection [16]. Elevations in liver function tests can occur with ADA deficiency, but also can be a sign of a viral infection. Prompt administration of ERT with PEG-ADA has led to resolution of abnormalities over several days in our patients. Therefore, unless there is an HLA matched sibling, we start ERT immediately at full doses of 30 units/kg or more to achieve rapid detoxification. After T cells appear, the infants are cared for at home with intramuscular PEG-ADA injections (125 units) twice weekly. Prior to HCT without other conditioning, we discontinue ERT, monitoring rising metabolites until approaching pre PEG-ADA levels. One of our ADA deficient patients received a matched sibling HCT, but 5 others who lacked a matched related or unrelated donor received ERT for several months. After 6 months of age, 4 infants enrolled in the experimental gene therapy (GT) protocol of Dr. Donald Kohn at UCLA, all with successful development of functional gene corrected lymphocytes. GT is planned for a fifth patient.

Leaky SCID and Omenn Syndrome (OS)

Infants with defects in any SCID gene that permit residual protein function can have more T cells than those with typical SCID; without screening the diagnosis may be delayed many months or years. Although nearly all are identified by TREC NBS, the mildest cases may not have low TREC numbers at birth. OS, due to hypomorphic mutations in RAG1, RAG2, requires immunosuppression while awaiting HCT, which should be implemented as swiftly as possible [3]. Aggressive autoreactive oligoclonal T cell expansion occurring within a few days to weeks of life can result in a severe scaly rash, diarrhea, and failure to thrive. Systemic corticosteroids; calcineurin inhibitors, and/or anti-thymocyte globulin or anti-CD52 monoclonal antibody treat this condition effectively until an HCT can be done.

Radiation Sensitive SCID (RS-SCID)

RS-SCID includes deficiencies of Artemis, DNA Ligase IV, DNA-dependent protein kinase catalytic subunit (DNA-PKcs), Cernunnos-XLF, and nibrin (the protein associated with Nijmegen breakage syndrome). Infants presenting with abnormal NBS generally have a T- (or T low) B-NK+ phenotype as the process of recombination of T and B cell antigen receptor genes require non-homologous end-joining by the DNA repair complex [17]. While RS-SCID is treatable with HCT, donor selection and conditioning must be adjusted because of increased sensitivity to typically used alkylating agents, such as busulfan, melphalan, thiotepa or cyclophosphamide [17,18]. T-B-NK+ lymphocyte profiles and developmental abnormalities, such as microcephaly and bird-like facial appearance, suggest heightened radiation sensitivity, prompting radiation sensitivity testing of patient fibroblasts, Western blots and/or gene sequencing (e.g. *LIG4*, *NHEJ1*) to establish the diagnosis. Although these take several weeks to perform, they must be obtained prior to HCT if conditioning is

planned. Despite this delay, it is our practice to hospitalize these infants while the workup is performed. While no absolute contraindication exists for clinically indicated X-rays, exposure should be minimized. Ultrasound and magnetic resonance imaging are safe alternatives.

Unique psychosocial vulnerability of parents with SCID-affected infants diagnosed by NBS

The combination within the first month of life of the postpartum period and the infant's hospitalization for SCID treatment present a situation of compounded vulnerability with the potential to destabilize family functioning and parental ability to provide care during and after HCT. At our institution, care providers create an early, open dialogue about psychosocial health impacts in this unique situation. Social workers interact extensively with the families during the hospitalization and after discharge.

Post-Partum Depression

Given the high stress experienced by these new parents, special efforts are made to recognize post-partum depression (PPD) and post-traumatic stress disorder (PTSD). PPD is a significant health concern affecting 10–15% of US women after normal births [19]. PPD has also been reported in new fathers [19]. The onset of PPD is typically 2 weeks after childbirth, coinciding with the diagnosis of SCID by NBS. Mothers who initiate or maintain breastfeeding are at lower risk for PPD, and interruption of breastfeeding may contribute to PPD [20]. Thus, interrupting breastfeeding while serologic status for CMV is tested adds to the stress during the postpartum period.

Post-Traumatic Stress Disorder

Parents learning of an abnormal metabolic newborn screen result have reported shock as if a death in the family occurred [21]. Likewise, the emotional distress experienced by parents of newborns with SCID may stem from shock, grief, guilt, and apprehension regarding losing the child. During the first weeks of hospitalization, confirmation of the SCID diagnosis and characterization of the genotype are sought through genetic testing. Additional testing for HLA typing is conducted on siblings and parents. Genetic testing is highly personal and is often associated with anxiety, fear and guilt for passing on an inherited condition [22, 23]. Our pilot study in which parents were interviewed revealed the majority of mothers and fathers caring for infants with SCID suffer from PPD and PTSD (M. Dorsey, unpublished data). Thus involvement of experienced social workers and nurses is a critical component of overall care for SCID families, with early introduction to available services and support serving as a key to decreasing stress. In addition, routine screening for PPD and PTSD can identify families in need of referral to mental health providers.

Treatment

Transplant: Allogeneic and Autologous Gene-Modified Hematopoietic Stem Cells (HSC)

Historically, single centers reported transplant outcomes for SCID in which the contributions of specific gene defects, infections, and variations in treatment protocols could not be

isolated. The PIDTC is addressing these issues by enrolling large numbers of patients from centers across North America [4,14]. In a retrospective report, Pai et al. (2014) showed that infants who received transplants before 3.5 months of age had a 5-year survival rate of 94%, as did infants older than 3.5 months but with no history of infection (90%) or whose infections had resolved by the time of HCT (82%) [4]. Children over 3.5 months of age with active infection at the time of HCT and no HLA matched sibling had the lowest survival (50%). HLA-matched sibling donors resulted in the best outcomes regardless of age or infection. While reduced intensity (RIC) or myeloablative (MAC) conditioning versus no conditioning or serotherapy alone had a higher likelihood of full T cell reconstitution and no need for immunoglobulin replacement therapy, long term effects of chemotherapy remain to be addressed [4]. For patients infected at the time of transplant at any age, the use of haploidentical donors without conditioning had the best 5-year survival. In a prospective PIDTC study, Dvorak et al. (2013) reported that the time needed for an unrelated adult donor (URD) or umbilical cord blood (UCB) search delayed HCT by approximately a month compared to using a mismatched related donor (MMRD) [14]. Additional studies will determine whether earlier HCT produces superior outcomes, or if closer HLA-matching in unrelated donors justifies the prolonged time to transplant in uninfected infants.

Gene correction of autologous HSC has been successful for children with ADA and X-linked (*IL2RG)* SCID [24–31]. The initial success in *IL2RG* SCID was tempered by the development of T cell leukemia due to retroviral vector insertional mutagenesis (IM) in 5 of the initial 20 recipients [26,27]. Current trials use self-inactivating retroviral or lentiviral vectors, which appear to have a significantly lower risk of IM [28]. Initial gene therapy attempts without conditioning for ADA SCID failed; when low dose busulfan was added as conditioning T and sometimes B cell immunity was reconstituted [29]. In early *IL2RG* SCID gene therapy trials without conditioning, T cell reconstitution occurred, but B and NK cell reconstitution were limited [28, 30]. Results from an initial trial using low dose busulfan for *IL2RG* SCID suggest that B and NK cells will be reconstituted [31].

Treatment outcomes at UCSF

Twenty-six California NBS identified SCID and leaky SCID cases and 6 from other states were treated at UCSF between September 2010 and October 2016. Twenty-nine (94%) are alive and well, 19 of them >2 years post-HCT. All of 26 who are >6 months post-HCT have reconstituted T cells and 13/26 (50%) have B cell reconstitution. Only 4 (14%) had a recognized family history of SCID. Gene diagnoses were: 7 *IL2RG*, 6 *ADA*, 5 *DCLRE1C*, 4 *IL7R*, 4 *RAG1*, 2 *RAG2*, 1 *JAK3*, 1 *RMRP*, 1 *BCL11B* [31]. One infant with unknown genotype (despite family exome and genome sequencing) died of refractory CMV infection present upon admission. One of 5 Artemis-deficient Navajo infants died of refractory HHV6 infection. Four ADA patients underwent busulfan-conditioned autologous HCT with gene corrected CD34+ cells; all are reconstituted with T cells and 2/4 who are >6 months post-GT have B cell reconstitution.

Our approach to selection of the best donor and conditioning regimen for patients with typical SCID is summarized in Figure 2. The guiding principles are to (1) use the donor associated with the lowest risk of graft vs. host disease (GvHD); (2) minimize the duration

of pre-HCT lymphopenia and risk for infections; and (3) use the least amount of chemotherapy possible to ensure donor T-cell engraftment but minimize the risk of short-and long-term toxicity. Unless there is an urgent need to proceed with HCT, such as a CMV infection, busulfan is not given to infants <3 months old. This approach accepts that booster HCT may be needed and B cell immunity will typically not be restored when a donor other than a matched sibling is used, except in patients with intrinsically normal B cells, e.g., *IL7R* SCID, leaving many patients dependent upon gammaglobulin replacement. In many patients with typical SCID, irrespective of donor type, no chemotherapy is required, though use of serotherapy may be beneficial when utilizing T-cell replete stem cell sources or when maternal chimerism is present and the mother is the donor [33]. For patients with leaky SCID or Omenn syndrome, a conditioning regimen is required. When using busulfan, we carefully monitor pharmacokinetics, which is highly variable in small infants, and modify divided doses to achieve the target cumulative exposure predicted to allow stem cell engraftment [34]. GvHD prophylaxis is aggressive, since there is no benefit to alloreactivity. Stem cells from haploidentical donors currently undergo CD34-selection [35], though in the near future, alternate *ex vivo* T-cell depletion strategies are planned. With the identification of maternal engraftment as a risk factor for the development of GvHD despite CD34-selection [33], low-dose rabbit anti-thymocyte globulin (rATG) serotherapy has also been successfully utilized in the most recent patients. For unrelated donors, we also employ pre-HCT serotherapy [36]. Serotherapy may abrogate an early (<100 days) wave of homeostatic donor T-cell expansion, but this typically can be tolerated in an uninfected patient identified by NBS, and naïve T-cell generation occurs at around 3 months post-HCT.

Post-transplant Management

Before hospital discharge, close attention must be given to availability of immediate medical care as well as T cell immunity and infection risk at home. If there are no siblings or if siblings are attending grade school, SCID patients are considered for discharge when they have $>50\times10^6$/L CD4+ cells and >10% of the lower limit of normal CD45+ lymphocyte proliferation to PHA. If preschool-age siblings are in the home, they should not attend daycare or group activities, and patient T-cell counts and proliferation need to be higher before discharge is considered.

Immune reconstitution is monitored with lymphocyte evaluation including naïve and memory T cell, NK cell and B cell subsets every 4 weeks until normal then every 3 months for the first year. Vaccines are initiated in patients who have B cell reconstitution when IgG replacement has been discontinued. At our institution, this is defined as total CD19 B cells >150 cells/uL; normal IgA and IgM for age; and IgM isohemagglutinin titers ≥1:8. If one of these criteria is not met, normal percentages of CD27+IgM-IgD- B cells is considered. Baseline titers against vaccine antigens are obtained 3 months after the last IgG infusion and 3–4 weeks following administration of the primary series of inactivated vaccines. If adequate responses are demonstrated, including a normal lymphocyte proliferation to tetanus toxoid, live attenuated vaccines are given.

Non-SCID TCL workup and management

For infants with 300 – 1500 T cells/uL, reduced but present naïve CD4 T cells (can be variable and <200 cells/uL), and no maternal cells (Figure 1, right panel; Figure 3), initial immune evaluation is similar to that for SCID, but hospitalization may not be required if immunodeficiency is not profound.

Non-SCID TCL conditions evaluated by UCSF immunologists (aside from 10 cases of TCL due to extreme preterm birth alone, which resolved in survivors) included multisystem syndromes in which T cells are variably reduced. Of 28 syndromic infants, diagnoses were: 17 DiGeorge syndrome/22q11.2 deletion or *TBX1* intragenic mutation (61% of syndromic TCL); 2 each of ataxia telangiectasia (AT), CHARGE syndrome and trisomy-21; and 1 each of Noonan syndrome, Kabuki makeup syndrome, CLOVES syndrome, Fryns syndrome, and immuno-osseous dysplasia. Of these, 1 each with CHARGE, CLOVES, Fryns and Noonan syndromes died of severe congenital anomalies. Nine infants have had secondary TCL, in which T cell generating capacity is normal but circulating T cells were diminished. Causes included 2 cases of hydrops, 3 of severe congenital heart disease; and 1 each of chylothorax, neonatal leukemia and maternal immunosuppressive medication (fingolomod) taken during pregnancy for multiple sclerosis. An additional 5 infants have had idiopathic lymphopenia with no underlying cause identified [11, 12, 14], although the syndromic infants with AT and immuno-osseous dysplasia and maternal medication were also in this category at initial presentation. Resolution of idiopathic TCL occurred in 1 infant, a fraternal twin, while 2 continue to be followed with low, but functional T cells, and 2 have been lost to follow-up. Long-term follow up is important in identifying underlying diagnoses when TCL persists (Figure 3).

It is important to remember that not all serious disorders of T cells are identified by TREC screening; combined immunodeficiencies (CIDs) that are associated with intact T cell development beyond the point of TCR gene recombination in the thymus, including Zap-70 deficiency and MHC class I and II non-expression, may have normal numbers of TRECs even though T cell function is severely impaired [37].

Alpha-fetoprotein (AFP) levels obtained before the age of 6 months are generally not helpful in the diagnosis of AT and should be interpreted with caution. It is known that AFP may be elevated in healthy infants but trends down with age unlike AT where AFP levels will remain elevated [38]. Similarly, functional antibody responses cannot reliably be assessed before completion of the primary series of killed vaccines. Those infants with <1500 T cells/mL are considered immune deficient and advised to avoid live rotavirus vaccination, due to the risk of acquiring diarrheal disease from the vaccine strain rotavirus [39]. Need for home isolation, prophylactic antibiotics, and gammaglobulin infusions are made on a case-by-case basis.

Conclusion

Early detection of SCID through TREC NBS creates the opportunity to provide immune reconstitution while infants are healthy and uninfected. While NBS optimizes the chances of

successful definitive treatment, newborns with SCID present new issues in management, including the parental emotional difficulties as well as defining best treatment regimens. Early detection of non-SCID TCL also poses new challenges. Further experience and multi-center studies will establish best practices.

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Appendix 1. Workup Outline for Infants with SCID to Prepare for Hematopoietic Cell Transplant

Blood tests for infant	Amount of blood*	Tube type, notes
CBC w/diff	0.25 mL	EDTA
Lymphocyte subsets and lymphocyte T cells subsets, naïve and memory (CD45 RA/RO)	0.5 ml	Purple microtainer, ordered as 2 separate tests
IgM, IgA, IgG, IgE	2 ml (1 ml without IgE)	SST
Blood chemistries, albumin, total bilirubin, LFTs	0.5 ml	Light green microtainer
HLA typing (prefer buccal swab if maternal engraftment suspected)	(2.6 mL if blood)	2 Buccal swabs or small ACD yellow top tube
Maternal engraftment by STR markers on CD3 selected cells	2.6 mL	2.6 ml (small) ACD
Serum CMV PCR	1.5 mL	EDTA
Lymphocyte proliferation to PHA (and other mitogens)	5 mL	Na heparin
ABO, Rh, Antibody screen	Microtainer	EDTA
SCID Gene molecular diagnosis (sequencing)	1 ml	Blood or Buccal swabs
ADA and PNP enzyme studies (if appropriate SCID)	1 mL	EDTA
Infectious Disease Markers (Should be done within 30 days of admission to BMT)		
HIV DNA PCR- immunology lab	1 ml	EDTA
HBsAg	0.6 mL	SST
EBV PCR	1.5 mL	EDTA
HCV DNA PCR	2 ml	EDTA
Other Pre HCT procedures		
Cardiac Echo		
Audiology Exam		
Other tests and procedures to consider		
Central line placement for infusions and blood draws		

Blood tests for infant	Amount of blood*	Tube type, notes
BAL only if indicated, to be sent for PCP, respiratory virus panel, cultures for bacteria, fungi, AFB, CMV; pertussis PCR		
Stool for cultures, rotavirus antigen, Norovirus, C. difficile if diarrhea present, O&P		
Respiratory virus PCR panel		

* Minimum amounts by special arrangement with our laboratories. Suggest working with local lab to achieve acceptable minimum amounts that are institution specific.

Appendix 2. Prophylactic medications for infants with SCID

Medication	Dose	Notes
IVIG	Initial loading dose 1 gm/kg. Get trough IgG at 1–2 weeks. After loading give at least 0.5 gm/kg every 3–4 wks (use full vial, e.g. 2.5 or 5g)	Maintain IgG trough >800 mg/dL, usually 0.5 gm/kg every 3 wk
TMP/SMX for PJP prophylaxis*	2.5 mg/kg TMP component BID PO 2 days/week	Start when >30 days old; monitor bilirubin and CBC
Leukovorin (folinic acid)	1.25 mg TMP/SMX 2 days/week with TMP/SMX	Minimizes TMP/SMX neutropenia
Atovaquone for PJP prophylaxis	30 mg/kg PO once daily, 1–3 mo 45 mg/kg PO once daily, 4 mo – 2 yr	Alternative to TMP/SMX
Dapsone	4 mg/kg PO weekly	Alternative to TMP/SMX
Acyclovir for herpesvirus prophylaxis	12.5 mg/kg PO BID or 8 mg/kg IV q12hr (≤12kg), 400mg/m2 PO BID or 250mg/m2 IV q12hr (>12kg)	
Fluconazole for fungal prophylaxis	6 mg/kg/q48 hours (full term <1 month) 6mg/kg/day ≥ 1 month	Ensure normal LFTs prior to starting; follow LFTs
Palivizumab	Per institutional protocol, q 4 weeks	During RSV season
PEG-ADA (Adagen)	At least 30 units/kg (up to half of a vial) twice weekly; start with full dose	Start for ADA deficient patients, while awaiting HCT or gener therapy

* Inhaled pentamidine is not recommended in children ≤ 5 years of age. There are no recommendations for IV pentamidine for prophylaxis in infants and would avoid this in the newly diagnosed babies given the risk of nephrotoxicity and possible hepatoxicity.

Abbreviations

SCID	Severe Combined Immunodeficiency
NBS	Newborn Screening
TCL	T cell lymphopenia
TRECs	T cell receptor excision circles
PIDTC	Primary Immune Deficiency Consortium
HCT	Hematopoietic cell transplant
ADA	Adenosine deaminase
HLA	Human leukocyte antigen

MMRD	Mismatched related donor
MUD	Matched unrelated donor
RIC	Reduced intensity conditioning
MAC	Myeloablative conditioning
CMV	Cytomegalovirus
PPD	Post partum depression
PTSD	Post traumatic stress disorder

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What is Known on This Subject

In the new era of severe combined immunodeficiency newborn screening establishing a diagnosis and managing these infants are new challenges for pediatricians.

What This Study Adds

This paper offers the first systematic approach to infants with severe combined immunodeficiency, leaky severe combined immunodeficiency and T cell lymphopenia identified by newborn screening.

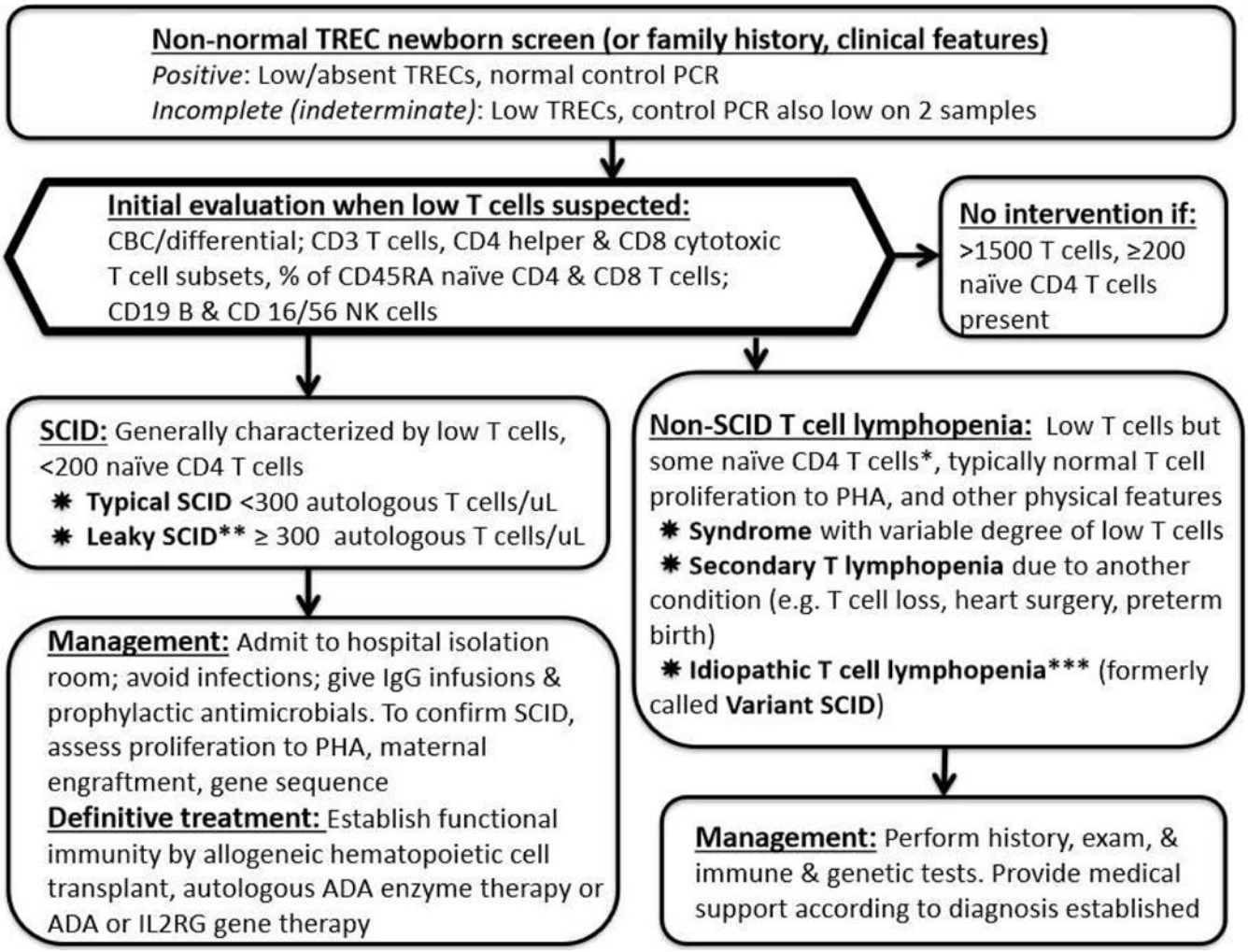

Figure 1.

Identification of T cell immune defects by newborn TREC screening; primary immune defects may also be diagnosed due to a history affected family members or clinical features. *Variable can be < 200 naïve CD4 T cells. ****Omenn syndrome** is a form of leaky SCID with rash; eosinophilia; autoreactive, oligoclonal T cells; and variable CD3 T cell count which can be >1500. ***Some infants never leave this group but some move out of this category when other diagnoses are made. These infants need to be followed over time.

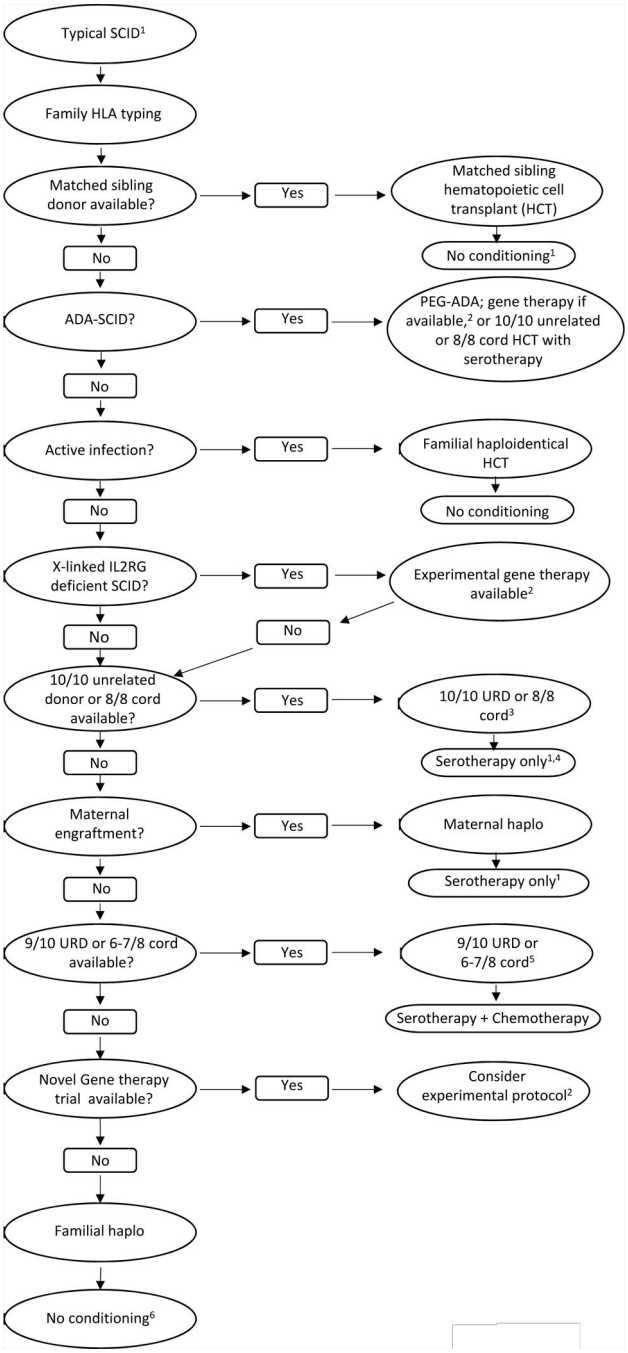

Figure 2.

Donor selection and conditioning for typical SCID. [1]Excludes Omenn syndrome and leaky SCID, both classified as atypical SCID for transplant purposes and requiring at least some conditioning; also excludes radiation sensitive SCID (DCLREC1, LIG4, etc.), for which donor selection and conditioning are individualized to balance risks of rejection vs. chemotherapy toxicity. [2]Gene therapy clinical trials should be considered for X-linked or ADA SCID when there is no HLA matched sibling. [3]Based on availability, CMV status, donor age, and other variables. [4]Non-radiation-sensitive T-B-NK+ SCID generally requires

chemotherapy plus serotherapy for unrelated donor transplant. [5]For X-linked and JAK3 SCID a maternal donor with serotherapy alone is preferred over a 9/10 unrelated or 6–7/8 cord donor. [6]Consider chemotherapy conditioning for enhanced B and/or T cell reconstitution and to prevent rejection.

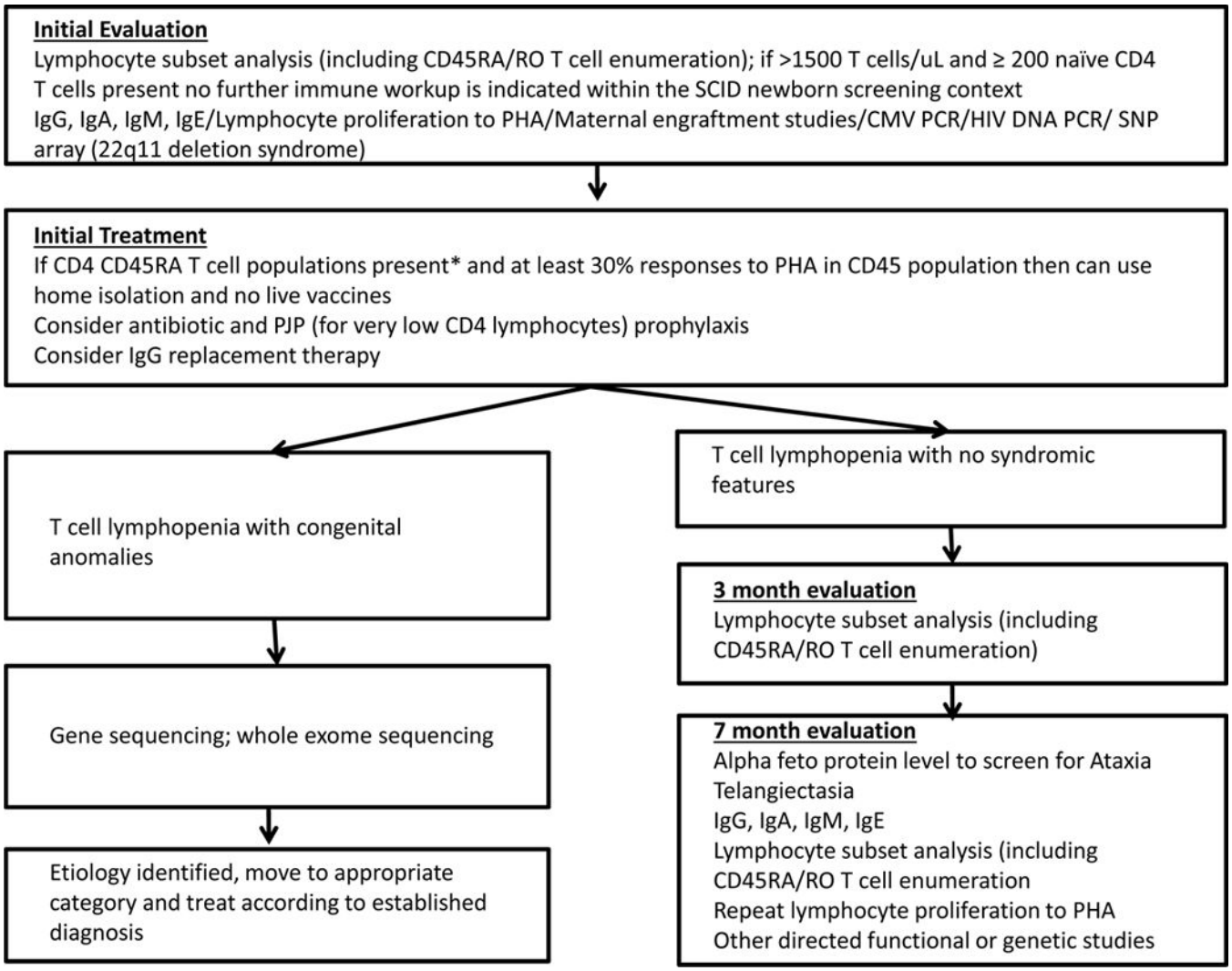

Figure 3.

Evaluation and management of non-SCID T cell lymphopenia (persistently <1500 CD3 T cells); continued follow-up is needed to determine degree and persistence of immune impairment, whether prophylactic therapy is needed, and whether an underlying etiology can be found.

*Variable can be <200 naïve CD4 T cells

Table 1

Approach to early management of new infant with lymphocyte subset profile consistent with SCID or leaky SCID.

- Isolate in a single room in SCID treatment center; prohibit ill contacts.
- Diagnose and treat any clinical abnormalities such as respiratory distress or signs of infectious or autoimmune conditions.
- Introduce social worker to help with support and services for family.
- Omit live vaccines for patient or household contacts including rotavirus vaccine
- Avoid infection:
  - Advise mother to suspend nursing while evaluating her prior exposure by CMV IgG serology.
  - If mother CMV seronegative, encourage to resume nursing.
  - If mother CMV seropositive and baby CMV negative by blood and urine PCR, advise mother to avoid nursing, given risk of breast milk transmission of CMV.
  - Obtain infant blood CMV PCR weekly for 4 weeks and periodically thereafter (more often if CMV clinically suspected, such as with elevation of liver transaminases).
- Intravenous access needed for gammaglobulin replacement with coordination of blood draws to decrease frequency of venipuncture. Establish more permanent intravenous access just prior to eventual hematopoietic cell transplant (HCT).
- Provide nutritional support, including monitoring for iron deficiency.
- Transfuse if symptomatic or hemoglobin <8 mg/dL, using only CMV-negative, leukoreduced, irradiated packed red cells.
- Administer immunoglobulin to maintain IgG >800 mg/dL [37]
- Administer palivizumab during respiratory syncytial virus season.
- Begin prophylactic fluconazole, acyclovir and trimethoprim-sulfamethoxazole (TMP/SMP) (the latter after 30 days of age).
- Perform HLA typing on parents and full siblings to evaluate as potential HCT donors; in absence of matched sibling, initiate search for an unrelated adult or cord blood donor.

* See appendix 2 for prophylactic medication dosing

Table 2

Early clinical and laboratory assessment for new infant with suspected SCID

• Directed history, including infections, family history, pedigree analysis, consanguinity.
• Physical exam, including documentation of any signs of infection, skeletal features and congenital anomalies associated with certain SCID variants, rash of GVHD or erythroderma of Omenn syndrome, desaturation associated with pulmonary alveolar proteinosis in ADA deficiency.
• Lymphocyte subsets, including T cell CD45RA/RO by flow cytometry, to confirm the initial flow cytometry done by the screening program.
• Blood chemistries, albumin, liver function tests and total bilirubin.
• PCR or antigen tests for HIV, HSV, EBV, HepB, parvovirus B19, adenovirus, respiratory viruses and rotaviruses. Do not rely on serologic tests. Serial maternal serology and/or PCR may suffice to rule out some of the above.
• Quantitative Immunoglobulins: IgG (reflects maternal transfer to baby, must be drawn prior to IVIG administration), IgA, IgM and IgE.
• Lymphocyte proliferation to PHA: send large enough sample to account for low lymphocyte count, and inform the testing laboratory of infant's low T cell count.
• ADA and PNP enzyme and purine metabolites (unless there is a family history or strong suspicion of another genotype).
• Maternal chimerism studies by DNA testing of polymorphic alleles.
• Molecular diagnosis directed by sex and lymphocyte profile (T-B+ or T-B−, and whether NK cells are present). Sequence single leading candidate gene or a panel of SCID genes.
• SNP array (or other copy number DNA array), preferred over Chromosome 22 FISH for infants with cardiac anomalies or any features suggesting DiGeorge syndrome.
• Unless concern for radiation sensitive SCID exists *, chest X-ray to document whether thymus is visible and view lung parenchyma and osseous structures. In asymptomatic infants, imaging can be deferred until documenting central venous line placement.

* microcephaly and bird-like facial appearance; infants with Navajo background