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Stem Cell Transplantation for Primary Immunodeficiency

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1. Introduction

In 1968, hematopoietic stem cell transplantation (HSCT) was first performed for patients with inherited cellular immunodeficiencies: a child with severe combined immune deficiency (SCID) and another with Wiskott–Aldrich syndrome (WAS) transplanted from matched siblings [1, 2]. Since then, HSCT techniques have advanced enormously due to refined HLA-tissue typing, the increased use of alternative donors, the availability of new stem cell sources including umbilical cord blood as well as less toxic chemotherapeutic conditioning [3], and graft-versus host disease (GVHD) prophylaxis. Supportive care has also improved, with molecular detection of viral infection enabling pre-emptive antiviral treatment before organ damage supervenes [4]. Greater awareness of primary immunodeficiency (PID) amongst general paediatricians, highlighted by campaigns promoting warning signs has lead to earlier diagnosis and referral to specialist centres[5]. Indications for HSCT increase as advances in molecular immunology better define PIDs while parallel studies of the natural history of PIDs reveal which will benefit most from early HSCT before organ damage is present[6]. There are now nearly 200 molecularly defined PIDs. HSCT aims to give stable donor stem cell engraftment after partial or full ablation of the recipient's marrow and immune system using a combination of chemotherapy, antibody therapy, and a graft-versus-marrow effect. [7] Nearly 1,500 children in Europe who have received allogeneic HSCT for PIDs[8] were reported recently, as well as over 1,000 children from Northern America in 2008.[9] Overall survival has increased to 90% for SCID babies with a genoidentical donor and nearly 70% for those given matched unrelated donor (URD) HSCT. For non-SCID PIDs, the survival for both genoidentical and URD HSCT is between 70% and 80%.[8] Together with improved survival advances have led, and continue to focus on, improved quality of life longterm.

2. Who, when, what to transplant

SCID is one of the most severe forms of PID. Early diagnosis and management are essential and HSCT is curative. For other forms of PID, management and in particular the indications for HSCT are evolving. New data is emerging on the natural history of patients with diseases such as CGD and CD40L deficiency and the outcome from HSCT has improved dramatically and so the risks of performing HSCT at a young age before organ damage from recurrent infection and inflammation need to be carefully assessed and discussed with families balanced against a potentially shortened life span with poor quality of life due to multiple hospital admissions.

Table 1 gives a current list of primary immunodeficiency diseases in which HSCT is indicated.

2.1. Importance of molecular diagnosis

Precise molecular diagnosis is very helpful as prognosis can then be accurately assessed. The outcome following HSCT for patients with B negative forms of SCID such as RAG deficiency is less good than for those with B positive forms. Furthermore those with artemis deficiency have a worse prognosis than those with RAG deficiency because of the associated cellular radiosensitivity. In the long term post HSCT human papilloma virus warts predominantly occur in patients with common gamma chain or JAK3 deficient SCID[10], but not other SCID genotypes.

Ill-defined combined immunodeficiencies (CID) are a challenge to treat, as the outcome with HSCT is variable, and often poor[8]. This is because, at least in part, decisions about when to transplant are not made until a disease-defining illness has occurred (Table 2), which may leave significant organ damage or a persisting viral burden, which alters the prognosis following transplantation. Identification of a specific gene defect in a patient cohort can alter management decisions. For example, clear molecular definition has changed the management of an autosomal recessive form of hyper IgE syndrome caused by mutations in the dedicator of the cytokinesis 8 (DOCK8) gene. Previously this disease was managed conservatively, as a form of hyper IgE syndrome. Accurate molecular definition enabled data to be gathered on a patient cohort leading to the realisation that there is a high risk of infection, skin malignancy and death. Reports of cases being successfully transplanted have lead to HSCT becoming the treatment of choice for this diagnosis.[11-15]

Reticular dysgenesis is an autosomal recessive form of SCID characterised by an early differentiation arrest in myeloid lineage and impaired lymphoid maturation. Affected individuals also have bilateral sensorineural deafness. Mutations in AK2 (adenylate kinase 2) published in 2008 demonstrated that AK2 is expressed in the stria vascularis region of the inner ear providing an explanation for the deafness in addition to it's role in specific haematopoietic lineages. Again this can lead to an earlier specific diagnosis and appropriate intervention.[16]

Severe combined immunodeficiency	Cytokine signalling	CγC JAK 3 IL7 Rα
	Nucleotide biosynthesis salvage pathway defects	ADA deficiency
	Defects affecting signalling through the T cell antigen receptor	CD45 CD3δ CD3ε CD3ζ
	VDJ recombination defects	RAG 1 & 2 Artemis Cernunnos DNA Ligase 4 DNA PK
	Other	AK2 deficiency (RD) ORAI1 STIM1
T cell immunodeficiency	CD4 lymphopenia Zap 70 kinase deficiency MHC class II deficiency PNP deficiency Omenn's syndrome CD40 ligand deficiency Wiskott Aldrich syndrome X-Linked lymphoproliferative disease MHC Class I deficiency Combined Immune Deficiency with skeletal dysplasia Cartilage hair hypoplasia Severe Di George syndrome (22q 11 del)* CHARGE association* Undefined	
Phagocytic cell disorders	Familial Hemophagocytic lymphohistiocytosis Griscelli disease Immunodeficiency with partial albinism Interferon-γ receptor deficiency Kostmann disease ** Shwachman-Diamond syndrome ** Leucocyte Adhesion Defect XL and AR Chronic Granulomatous Disease Chediak-Higashi syndrome Undefined	
Severe Immune dysregulation	Autoimmune lymphoproliferative syndrome (homozygotes) ** IPEX syndrome	

Abbreviations: CγC Common gamma chain, XL X linked, Jak 3 Janus associated kinase 3, AR Autosomal recessive, ADA Adenosine deaminase, RAG Recombinant activating gene, MHC Major histocompatibility, PNP Purine nucleoside phosphorylase, IPEX Immunodeficiency polyendocrinopathy enteropathy X linked, CHARGE coloboma, heart anomalies, choanal atresia, retardation of growth and development, and genital and ear anomalies due to CHD7 gene mutations.

*Thymic transplant recommended in preference to HSCT
**Not all require HSCT

Table 1. Indications for HSCT in immunodeficiencies

2.2. Severe combined immunodeficiency

Severe combined immunodeficiency is usually fatal by 1 year of age unless an infant receives a new immune system and should be considered a paediatric emergency which is immediately life threatening. In the most recent analysis of 699 SCID patients transplanted across Europe there was a survival of 90% for those transplanted with a genoidentical sibling donor and nearly 70% for those receiving a matched unrelated donor[8]. The outcome for those without pre-existing infection such as those diagnosed at birth is even better: in the UK series the survival for those transplanted having being diagnosed at or before birth was 91.5% compared to 61% for those transplanted having being diagnosed at a median age of 143 days and a significant number of these children died from infection before reaching transplant[5]. These data support neonatal screening programmes, which are being introduced in North America, and pick up patients with SCID in the newborn period, before infection supervenes[17]. Similar screening programmes are being considered in Europe. Recognition of the specific molecular defect may alter the approach to HSCT. Patients with adenosine deaminase-deficient SCID will develop adequate immunological reconstitution following an unconditioned infusion from HLA-identical sibling stem cells[18], whereas those with RAG-deficient SCID will require chemotherapy conditioning to achieve stem cell engraftment and immunological reconstitution.

2.3. Other primary immunodeficiencies

For other non-SCID PID, debate continues about the optimum age for transplantation. As registry data for specific diseases becomes available, the role of HSCT is increasingly clear, but the optimum age for transplantation remains to be determined. Earlier transplantation is favoured for T cell immunodeficiencies.

Filipovich et al published results of 170 transplants for Wiskott Aldrich syndrome and demonstrated that boys receiving an unrelated donor transplant before the age of 5 had as high a survival rate to matched sibling donor recipients of any age[19]. A recent international report of 194 patients with Wiskott Aldrich syndrome, transplanted in 12 centers, reported an overall survival of 84%, rising to 89% for those transplanted since the year 2000. Younger age and milder clinical phenotype was associated with better outcome[20]. Whilst survival has improved over the last decade, young age at transplant before complications of the underlying disease supervene, also improves outcome.[21]

The outcome of patients transplanted for CD40 ligand deficiency is also dependent on age at transplantation, with pre-existing lung disease being a significant factor in predicting survival[22, 23]. Thirty eight children were reported who were transplanted in Europe for CD40L between 1993 and 2002. Of the 34 engrafted, 26 survived (68%), and 20 no longer required immunoglobulin replacement therapy. This survival and cure rate was little better than the survival of non-transplanted patients at the age of 20 years. However, many patients in this series were over 10 years of age when transplanted and already had significant lung and/or liver damage. A total of 14 patients with CD40L deficiency have been transplanted in our centre: 2 received MSD and 10 URD; 4 died (overall survival 71%), but

none since the year 2000, and 3 out of the 4 deaths were in children over 12 years of age who already had sclerosing cholangitis. All the survivors express CD40L and only one needs immunoglobulin. These data further emphasize the importance of early age at transplant before organ damage supervenes.

For non-T cell immunodeficiencies, timing of transplantation has been more controversial, although with registry data becoming available, the natural history of disease on conventional treatment is becoming more clear. Lifelong antibacterial and antifungal prophylaxis with cotrimoxazole and itraconazole has improved short- and medium-term survival for patients with chronic granulomatous disease. However, although steroids and aminosalicylates ameliorate colitis and other inflammatory complications they do not cure the underlying genetic defect and longterm immunosuppression is required to maintain symptom control. Quality of life is poor with frequent hospital admissions and poor growth. Even with the best prophylactic treatment, only 50% of patients are alive at 30 years[6] (Figure 1). HSCT can cure CGD with resolution of infection and colitis but was previously considered to be a high risk procedure. A European multicenter experience of replete marrow HSCT with mainly matched sibling donor stem cells following myeloablative conditioning gave good results; 23/27 patients survived, 22 were cured (81%), with deaths confined to high-risk patients with active fungal infection[24]. A more recent single centre study demonstrated similar outcomes with either matched sibling or matched unrelated donors, with a survival and cure of 90% and low incidence of significant GvHD. Mean weight and height for age Z scores on recovery from HSCT rose significantly. Transplant-associated complications were restricted to those with pre-existing infection or inflammation, supporting the argument for early HSCT for all CGD patients with a well matched donor[25]. As transplantation techniques improve, and survival increases, earlier transplantation becomes a more attractive option for other primary immunodeficiencies requiring long term antimicrobials and immunosuppression to control symptoms.

2.4. Donor choice and degree of HLA matching

Unlike patients with haematological malignancy in whom a graft versus leukaemia effect is desirable, GVHD confers no benefit to patients with PID. The best HLA matched donor is a sibling and so any siblings of the patient should be tissue typed. Many PID patients come from consanguineous families and so it may be possible to find a donor from the extended family. When tissue typing reveals more than one possible donor, other factors such as age, sex, parity, blood group and cytomegalovirus (CMV) status are taken into consideration. If no family donor is found a search of the National and International unrelated donor registries should be undertaken. There are currently 19 million adult and over 500,000 cord blood donors that can be accessed through the Bone Marrow Donors Worldwide registry (www.bmdw.org).

3. Stem cell source

Bone marrow has been the traditional source of stem cells and is harvested under general anaesthetic from the posterior iliac crests. Adult donors are increasingly being offered the

option of donating peripheral blood stem cells (PBSC) rather than bone marrow. This may be especially useful for donors who may have a medical condition of their own that would increase the risks of general anaesthesia. PBSC collection is generally carried out as a day case procedure and some donors find this prospect less difficult than the short period of hospitalisation and general anaesthetic needed for bone marrow donation. However, PBSC will require the donor to receive a short course of injections of granulocyte-colony stimulating factor (G-CSF) prior to commencing the first collection. Typically G-CSF at 10 micrograms per kilogram is given daily for 5 days before 1-2 leukapheresis procedures are performed. This procedure is not licensed in children in most countries and so sibling donations from children continue to be bone marrow.
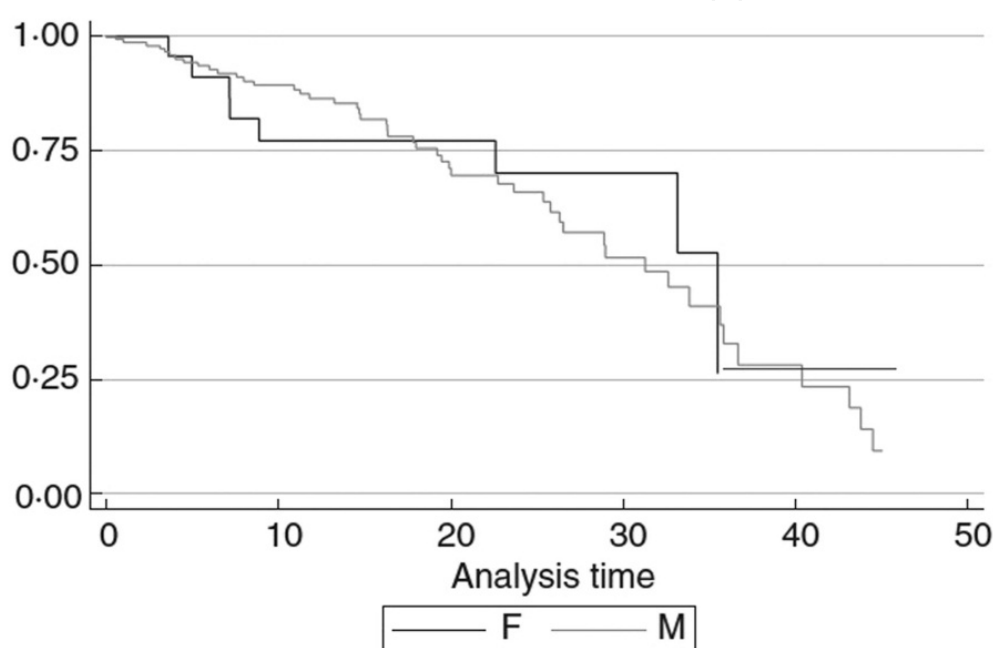
(Permission granted from Jones, *et al*[6]. Copyright by John Wiley and Sons, Ltd.)

Figure 1. Kaplan–Meier survival estimates, by sex for UK patients with chronic granulomatous disease.

Umbilical cord blood stem cell transplantation (UCSCT) offers a stem cell source when a matched sibling donor is unavailable.

Advantages of UCSCT include

i.   quick access to the cord blood unit and ease of arranging date of transplant
ii.   absence of risk to donor
iii.   lower risk of latent viral transmission and graft versus host disease (GvHD)
iv.   higher chance of matching rare HLA haplotypes.

Additionally, the HSC telomere length is longer in patients who have received UCSC than in those who have received PBSC from older donors. UCSCs may have greater self renewing capacity and longevity than those derived from an adult donor. As many transplanted PID patients are infants or young children, giving HSC with a greater proliferative life span is theoretically more attractive.

Disadvantages of UCSCT include

i.   low stem cell dose, particularly for an older child,
ii. Lack of availability of the donor should any boost procedure be required
iii. Cord blood units are virologically naïve and have been reported to have slower engraftment which increases the risks from pre-existing infections.

However, the conditioning regimen employed may influence this, as omission of serotherapy enables viral clearance, albeit with an increased risk of GvHD[26].

For PID patients who often present before 1 year of age, UCSCT can offer a suitable stem cell source. In a recent European study mismatched-related donor HSCT was compared to unrelated donor cord HSCT in children with severe T cell deficiencies. There was no significant difference in survival, but cord blood recipients had a higher frequency of complete donor chimerism at day 100 and faster total lymphocyte recovery.[27]

4. Assessment of donor

A Physician or Paediatrician who is independent to that of the recipient should perform a pre-transplant assessment of any family donor. A medical history should be taken including:

•   Vaccination history
•   blood transfusion history
•   allergy history
•   history of travel to tropical countries
•   number of pregnancies in women

A routine physical examination is performed and a chest radiograph and ECG if indicated.

For unrelated donors the examination is performed by the donor assessment centre. However the transplant centre will need to request blood for confirmatory tissue typing, DNA analysis for post transplant chimerism studies and the opportunity is usually taken to check the donor virology and serology status. The donor's fitness to donate must be ascertained before conditioning of the patient begins. The donor centre is responsible for the consent of the donor. The donor needs to be fully informed about the procedure for collecting the stem cells, the blood tests that will be performed including HIV status, the possibility of a second donation for the same patient and the emphasis on anonymity for the donor and patient. Anonymity may be relaxed in time and regulations may vary in different countries.

For related donors in the UK in addition to a physical examination, under the Human Tissue Act 2004, any potential donation of bone marrow or PBSC from adults who lack capacity to consent and children who lack competence to consent, must be assessed by an Accredited Assessor and a report submitted to the Human Tissue Authority for consideration.

Psychological aspects for the donor are important particularly when children are donating for siblings, for example if the recipient does not survive the donor may feel it is their fault. Therefore preparation and counselling for the donor is important

For cord blood donations at the time of cord collection tests for the following are performed on the mother:

The current minimum serology testing requirements for these products are summarised below:

Anti-HIV-1,2
HBsAg
Anti HBc (hepatitis B core antibody)
Anti-HCV-Ab (anti-hepatitis C antibody)
A validated testing algorithm to exclude the presence of syphilis/active infection
with Treponema pallidum (Anti-T. pallidum)
HTLV-I antibody testing for maternal donors living in, or originating from, high-incidence areas or with sexual partners originating from those areas, or where the donor's parents originate from those areas.

Furthermore, if the cord blood unit has been stored for more than 180 days the additional testing is required:

Nucleic acid testing for HIV (HIV-NAT)
Nucleic acid testing for hepatitis B (HBV-NAT)
Nucleic acid testing for hepatitis C (HCV-NAT)
Nucleic acid testing for HTLV-I (HTLV-I-NAT) of maternal donors living in, or originating from, high-incidence areas or with sexual partners originating from those areas, or where the donor's parents originate from those areas.

Confirmatory tests 3 to 6 months after delivery are also performed. The health of the baby is also assessed. Cord blood banks will supply the transplant centre with the required viral status of the mother and sometimes the cord blood itself. The cell doses contained within the cord donation and cell viability are also recorded. They will also perform extra tests for confirmatory tissue typing and virology and serology on small aliquots of the cord. Some centres may wish to perform their own confirmatory typing. A small sample is usually taken at the time of thawing for DNA analysis for post transplant chimerism studies.

5. Preparation of patient

The clinical condition of patients undergoing stem cell transplantation varies enormously depending on diagnosis, age, previous treatment, and organ damage. Once the decision to transplant has been made and a donor selected each organ system should be assessed so that any organ damage is known about prior to transplant and therefore potential harmful effects of chemotherapy, risks for GVHD and recurrence of infections can be anticipated. A full medical history should be taken and a physical examination performed. It is useful to perform the assessment at least 6 weeks before the transplant date in order that any necessary investigations can be arranged. A checklist is essential (Figure 2). It is helpful to perform microbiology screening for resistant bacteria e.g. methicillin resistant staphylococcus aureus (MRSA) and glycopeptide resistant enterococci (GRE/VRE). This will depend on unit policy.

Name of patient		MRN		D.O.B.	
Description			**Date**		**Initials**
Full blood count					
Coagulation Screen					
Blood group					
Tissue typing					
DCT, Isohaemagglutinins					
U & E's, LFT's, Bone profile, Glucose					
Thyroid function tests					
Immunoglobulins (+Ig Subclasses)					
Specific antibodies (as appropriate)					
Lymphocyte subsets					
Proliferations					
Viral serology (CMV, EBV, Hep B & C, VZV, HSV, Measles, Toxoplasma, Treponema, HIV 1&2)					
Viral PCRs (CMV, HHV6, EBV, Adenovirus)					
DNA genetic markers					
Amino glycoside Study					
Other blood tests					
Referral for sperm donation (if applicable) **date:**					
Stool samples (C and S, Virology)					
Urine (C and S, +/- Virology)					
Nose /Throat Swab/NPA					
Chest X-ray					
Lung Function					
Ultrasound abdomen					
Echocardiogram					
Dental check					
Audiology					
Medical photography					
Height	**Weight**				

With permission from Kath Grey. Liaison team nursing sister. Great North Children's Hospital, Newcastle upon Tyne

Figure 2. Checklist for patients requiring an immunology assessment for HSCT

A number of infectious agents, particularly viruses, can now be detected by sensitive molecular techniques such as polymerase chain reaction (PCR) at an early stage of the infectious process. This in turn means that pre-emptive therapy can be given before organ damage such as life threatening pneumonitis or hepatitis, occurs. Such early detection together with treatments such as cidofovir for Adenovirus has led to a dramatic improvement in outcome following viral infection.

Many children with PID fail to thrive and are malnourished and it is important to maximise nutritional status prior to transplant, which may mean high calorie enteral feeding via nasogastric tube or parenteral nutrition through a central venous catheter.

Some diseases such as IPEX syndrome require immunosuppression to control autoimmune enteropathy and it is important to suppress any active inflammation prior to transplant, but also to minimise immunosuppression in order to reduce risk of infection.

Familial haemophagocytic lymphohistiocytosis is a genetically determined disorder characterized by the early onset of fever, hepatosplenomegaly, central nervous system disease, thrombocytopenia, coagulation disorders, and haemophagocytosis. It is caused by genetic defects that impair T cell–mediated and natural cytotoxicity. Chemotherapy and/or immunotherapy-based treatments can achieve remission, but HSCT is the only curative option. Outcome of HSCT is much more favourable if active disease is controlled prior to transplantation[28].

Sometimes a combination of antimicrobials and immunosuppression may be the optimal treatment. For example, Leiding et al have described 9 cases of severe life threatening liver abscesses in patients with CGD that progressed despite appropriate antibacterial therapy and drainage but responded to moderately high doses of corticosteroids tapered over several months.[29]

It is essential to address family and psychosocial issues and so each patient should be referred to a social worker and psychologist. Each unit will have their own system for information giving including written information about the transplant procedure, a visit to the unit and a home visit. It may be appropriate for patients to meet the dietician and physiotherapist prior to transplant. Fertility issues need to be discussed. In particular sperm banking should be arranged if appropriate. It is vital that written consent is taken once the patient/family are fully aware of all the aspects of the transplant.

6. Conditioning

Conditioning aims to create space in the recipient marrow niche to enable donor stem cells to engraft more easily. A range of conditioning regimens are available from non conditioning, through immunosuppressive regimens to myeloablative combinations (Figure 3).

For many years, myeloablative chemotherapy with busulphan and cyclophosphamide was given prior to HSCT for PIDs. However, busulphan is associated with significant toxicity including veno-occlusive disease (VOD). A strong correlation between blood levels of cyclophosphamide metabolites and VOD has previously been shown due to depletion of glutathione from the liver[30]. Combinations of cyclophosphamide with reduced-dose busulphan may also lead to severe hepatic toxicity and VOD [31, 32]. The combination of fludarabine and full dose busulfan has gradually replaced the combination of busulfan and cyclophosphamide in most centres. Malar et al. have recently reported on the importance of therapeutic drug monitoring for intravenous busulphan therapy in 34 paediatric patients. Seven children all weighing less than 12 kg had VOD despite not exceeding the targeted

area under the curve: six of them had AUCs below the target range, highlighting the difficulty in giving busulphan to this young group of children despite careful therapeutic monitoring.[33]
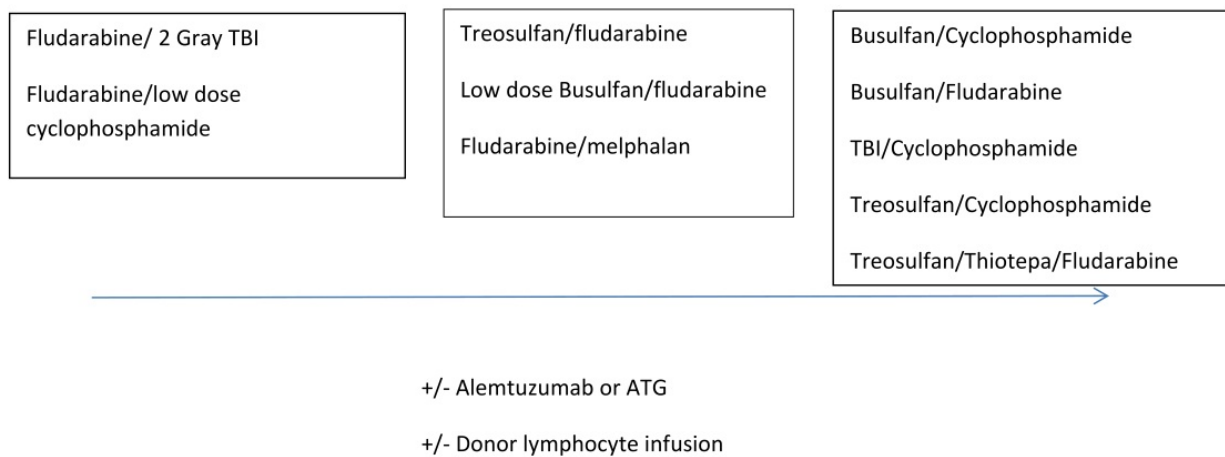

Figure 3. Chemotherapy regimens, increasing in intensity from left to right

Reduced-intensity conditioning regimens using drugs such as fludarabine and melphalan have diminished treatment-related toxicity in some PID patients,[3] but toxicity remains a problem for children under one year of age,[34] and cardiac toxicity is associated with melphalan[35]. Minimal-intensity conditioning with fludarabine and low dose cyclophosphamide can reduce toxicity even further, but may be associated with poor donor myeloid chimerism or an increased incidence of GVHD.[36] Consequently, new conditioning regimens for PIDs have been developed that give adequate myeloablation but less toxicity, particularly in patients under one year of age. Treosulfan (l-treitol-1, 4-bis-methanesulfonate) is the pro-drug of l-epoxybutane, a water-soluble bifunctional alkylating agent with myeloablative and immunosuppressive properties,[37] and is effective in HSCT conditioning with less toxicity, particularly in VOD, compared to busulphan. A recent study of 70 children in the UK, transplanted for various PIDs and given treosulfan with either fludarabine or cyclophosphamide, showed an overall survival of 81%. Of 46 children less than 12 months of age at HSCT, only eight died (overall survival 83%). Two children had VOD when treosulfan was given in combination with cyclophosphamide, but none when it was given with fludarabine. In the treosulfan cohort, there was no cardiactoxicity, such as seen with melphalan or pulmonary fibrosis as seen with busulphan[38-40]. T cell chimerism was significantly better when treosulfan was given in combination with fludarabine, and although numbers were small, there was a suggestion that chimerism was better when PBSCs were used as the stem source. Conversely, patients receiving cord blood stem cells in conjunction with serotherapy had a tendency to more mixed donor chimerism. While donor myeloid chimerism was sufficiently high to cure most patients with CGD and WAS with all donor types/stem cell sources, where 100% donor chimerism might be preferred in all cell lineages—for example, WAS[20] — then the use of PBSCs or full myeloablation with busulphan might be required. Early results using PBSCs with treosulfan and fludarabine look promising in terms of good chimerism without an increased incidence of greater than

grade II GVHD. Prospective studies are needed comparing treosulfan/ fludarabine with busulphan and fludarabine,[41-43] where early reports also indicate reduced transplant-related toxicities and excellent survival. Long-term follow-up is needed to see if this "modified intensity" regimen results in less long-term toxicity and, in particular, infertility, as the gonadal toxicity of busulphan is already well documented[44]. Radioimmunotherapy, in which targeted irradiation of the bone marrow is achieved by using radiolabeled monoclonal antibodies, may be used as a potent myeloablative agent with low intrinsic organ toxicity. Schulz et al. recently reported on the use of radioimmunotherapy with 90Y-anti-CD66 for conditioning in 30 pediatric patients undergoing HSCT, including 16 with non-malignant disorders with high co-morbidities. Patients received radioimmunotherapy with fludarabine or melphalan alone or in combination, usually with ATG; one patient was given radioimmunotherapy alone. A highly favorable ratio of marrow dose to other organ dose was demonstrated, and stable engraftment was achieved with complete donor chimerism in 13 out of 16 (81%) patients. One patient with Griscelli syndrome and who had secondary graft failure after the first HLA haploidentical transplantation achieved stable complete donor chimerism with radioimmunotherapy alone[45].

Antibodies to eliminate host stem cells prior to transplantation could mean that toxic conditioning regimens would not be needed. Administration of ACK2 and an antibody that blocks c-Kit function led to transient removal of >98% of endogenous HSCs in immune-deficient mice[46]. Subsequent transplantation with donor HSCs led to chimerism levels up to 90% compared to only 3% in those without this preconditioning. The pharmacological agent AMD3100, which is a CXCR4 inhibitor, has been shown to induce egress of HSCs out of the marrow and improve levels of donor HSC engraftment relative to untreated recipients[47]. Plerixafor, a CXCR4 inhibitor, may also be a more effective mobilizer of stem cells for donors of PBSCs than granulocyte colony stimulating factor (G-CSF), allowing larger doses of stem cells to be administered, which may lead to more rapid engraftment.

7. Methods of T cell depletion

In 1981 the introduction of T lymphocyte depletion to remove of alloreactive lymphocytes from the bone marrow source enabled transplantation across HLA barriers. This meant that patients without an HLA-identical donor could receive T lymphocyte depleted marrow from an HLA-haploidentical parent.

Profound T-cell depletion is a fundamental prerequisite for haploidentical donor transplantation to avoid severe GVHD and B cell depletion lessens the risk of Epstein Barr Virus (EBV)-related lymphoproliferative disease. Various methods have been used to remove viable T lymphocytes from the graft, including E-rosette lectin depletion and in vitro anti CD52 (CAMPATH-1M anti-lymphocyte antibody which is no longer available). Since the late 1990s European centers performing T lymphocyte–depleted HSCT for patients with PID have used CD34+ stem cell selection rather than T-lymphocyte depletion. The most commonly used method, the Miltenyi Clini-MACS system, uses an organic iron bead attached to an anti-CD34 antibody to isolate purified CD34+ HSCs from the other cells by

passing the HSC source through a magnetic column[48]. The purified CD34+ HSC fraction is infused into the patient. By using this method, 4 log depletions of T-lymphocyte numbers can be achieved. There are important differences between CD34+ stem cell–selected and T lymphocyte–depleted bone marrow. Although the residual T-lymphocyte count in anti-CD52–treated marrow can be relatively high, very few of the T lymphocytes remain viable because they are still coated with anti-CD52 when infused into the recipient and are then destroyed by complement-mediated lysis. In the anti-CD34 selected HSC product, although the number of T lymphocytes infused into the patient might be very low, those that are infused are viable and could cause GvHD. The anti-CD52– treated product contains component blood cell precursors and cells already in early differentiation from the stem cell, as well as other stromal factors that might aid engraftment of HSCs into the bone marrow space thus achieving better engraftment.

An alternative means of stem cell enrichment has been used with the aim of achieving an optimal balance between the competing risks of GVHD, poor engraftment and delayed immune reconstitution. By targeting T- and B- cells specifically CD3/CD19 depleted grafts not only retain CD34+ stem cells but also CD34 negative progenitors, natural killer, dendritic and graft-facilitating cells which enhance engraftment.

Further developments are now focusing on the depletion of TcR$\alpha\beta$+ T cells which can prevent GvHD after allogeneic stem cell transplantation from HLA non identical donors and may lead to more rapid immune reconstitution than CD3+ depletion of grafts. In contrast to depletion of CD3+ T cells valuable TcR$\gamma\delta$+ T cells are spared.[49]

8. Results

8.1. Severe combined immunodeficiency

A successful HSCT procedure in patients with primary immunodeficiency should result in a cure of the underlying defect with normal immuno-reconstitution leading to normal immune function and a normal life. The likelihood of a successful outcome depends on the underlying diagnosis, degree of HLA matching from the donor, and pre existing end organ damage. Overall the outcome for patients with SCID is better than that for patients with other PID and has improved over time[8]. The likelihood of cure following HSCT is up to 90% in those SCID patients with a genoidentical donor. Even for those with no HLA identical donor, survival following a T cell depleted haploidentical transplant is approaching 60%. Whilst infusion of the stem cell product without the use of chemotherapy eliminates the potentially fatal effects of conditioning including infection from aplasia and increased risk of pneumonitis and mucositis, the quality of engraftment is compromised. Thus for patients with common gamma chain or JAK3 SCID thymopoiesis may be achieved as T cell precursors engraft in the thymus[50], but B cell engraftment is unlikely and patients will remain on lifelong immunoglobulin replacement as recipient B cells are unable to produce IgM through the IL 21 mediated signalling path and do not undergo immunoglobulin class switching.[51] Patients thus remain at risk of bronchiectasis in the

long term. However, for patients with VDJ recombination defects leading to T- B-NK+ SCID, stem cell infusion will lead to no B cell engraftment and only peripheral T cell engraftment with poor T cell numbers, no thymopoiesis and risk of immunosenescence in the medium term.[52]. So, although conditioning may increase the short term risks, immune function is better in the long term following chemotherapy. Full donor chimerism is not necessary as stable mixed donor chimerism may give adequate immune reconstitution. Best results are achieved in patients diagnosed early with no infection and no end organ damage. Newborn transplantation gives the best results of all[5, 53, 54], giving weight to the argument for newborn screening by detection of T cell receptor excision circles (TRECS) on the neonatal blood spot (Guthrie) card and thus detection of SCID before presentation with a SCID defining illness such as pneumocystis pneumonitis (Table 2).

Common Presentations	Rare Presentations
Persistent or recurrent viral gastroenteritis	Bacterial septicaemia
	Disseminated BCG infection
Persistent or recurrent viral lower respiratory tract infection	Haemophagocytosis
Pneumocystis jiroveci pneumonitis	Lymphoid malignancy
Recurrent or recalcitrant candidiasis	Autoimmune cytopenias
Fungal abscess	Maternofetal GvHD
Recurrent bacterial lymphadenitis	
Persistant cutaneous human papillomavirus warts	
Persistant molluscum contagiosum	

Table 2. Disease defining illness in Primary Immunodeficiency

For those patients presenting late with infection, graft manipulation such as new methods of T cell depletion and add back of virus specific cytotoxic T cells has enabled more rapid immunoreconstitution and viral clearance[55]. For those patients requiring additional support through the transplant period, improvement in paediatric intensive care, close liaison between BMT and PICU teams, and earlier interventions are leading to improved outcomes of patients receiving HSCT but needing to go to PICU. Survival rates are now approaching up to 60%, better survival for those who receive only invasive ventilation and worse survival for those with multi organ failure[56].

8.2. Other primary immunodeficiencies

The results of transplant for other primary immunodeficiencies are historically not so good as those for patients with SCID but are improving over time[8]. This is likely due to a number of factors. Foremost is the adoption of stem cell transplant as a recognised therapy for the underlying condition. As transplant becomes an accepted treatment for a specific condition, patients with end organ disease undergo transplant, and some do not survive. Secondly, patients will need to undergo conditioning chemotherapy in order to prevent rejection of the graft and achieve donor chimerism and are therefore subject to the toxicities associated with conditioning. Thirdly, is the question of the optimum time to perform transplantation. Severe combined immunodeficiencies are usually fatal within the first 18 months of life, patients with other primary immunodeficiencies live longer and can survive into adulthood, albeit with a deteriorating quality of life as they accumulate end organ damage secondary to infection and inflammatory complications of the underlying disease. Registry data has provided good quality information about long term prognosis for some of these disorders, even when appropriate antimicrobial prophylaxis is available, enabling parents, patients and physicians to make informed choices about timing of transplantation[6, 11, 57]. Most non-SCID PIDs are life limiting in the medium to long term, rather than fatal in the first few months of life. As with SCID, results of transplant are better if patients are transplanted whilst younger, without end organ damage. For those with XLP, survival from transplant is significantly better for patients who do not experience haemophagocytic lymphohistiocytosis[58]. In a similar fashion, for patients with chronic granulomatous disease, outcome is better in those that have not had significant fungal infection or inflammatory sequalae[24]. Thus transplantation at an early age before onset of complications gives the best chance of survival and cure but some families may prefer not to put a healthy child through transplant, rather monitoring them closely for significant deterioration and transplant at the first sign of trouble. It is advisable that searches for an appropriate donor are made soon after diagnosis so that transplantation can proceed quickly once the decision to transplant has been made.

The amount of chimerism required to achieve cure depends on the underlying disease. Patients with CD40 ligand require only good T cell donor chimerism which will enable immunoglobulin class switching, whereas Wiskott Aldrich Syndrome patients with incomplete donor chimerism are much more likely to develop auto immune complications[20]. The cell lineage in which donor chimerism is required may not be

obvious. A patient with IPEX syndrome was cured although T cells appeared to be recipient, because on detailed study, the only donor cells were found to be the FOXP3+ cells[59], required to cure the condition. On the other hand, donor B cells are required in CgC or JAK-3 deficient SCID to achieve independence from immunoglobulin substitution[51, 60], but recipient B cells are functional in SCID due to defects in IL7Ra.

9. Complications post HSCT

Complications following transplantation for primary immunodeficiency are similar to those for patients with malignancy. Infection through the transplant period is a risk, particularly when patients are aplastic (Figure 4). Any primary immunodeficiency patients may carry pre-existing infection (particularly viral infection) into transplant, as the underlying condition means that they are unable to clear infections effectively. Careful assessment of
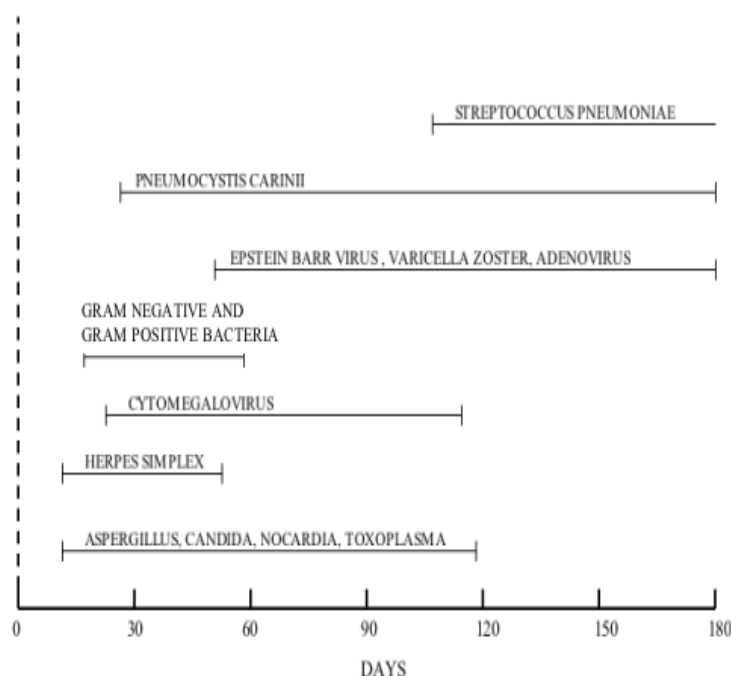

Figure 4.

pre-existing infection needs to be made pre-transplant so that treatment can be optimised as patients begin transplant to reduce the risk of infectious complications. Pneumonitis, particularly at engraftment is a risk, especially in those with pre-existing viral pneumonitis at time of transplantation[61]. Nebulised steroids may improve the outcome in these patients. Acute and chronic graft versus host disease may be significant complications in primary immunodeficiency patients post transplant. Unlike patients with malignancy, graft versus host disease is not encouraged as there is no graft versus leukaemia benefit to be gained. Therefore HLA matching, T cell depletion where appropriate and Graft versus host disease prophylaxis with a calcineurine inhibitor (with or without additional MMF/steroids/Methotrexate) should be utilised. Staging and treatment of established graft versus host disease (Table 3) is as for other transplant patients - methylprednisoline

(2mg/kg) is standard first line treatment. If the patient fails to respond to this, then a variety of other agents may be tried including monoclonal antibodies such as infliximab, alemtuzimab, antithymocyteglobulin, mesenchymal stem cells and extra corporeal phototherapy but none have yet proven to be consistently effective. Veno occlusive disease is a risk factor particularly in those with osteopetrosis, with pre-existing liver disease or under going conditioning with busulphan or cyclophosphamide. Prophylaxis with defibrotide is effective at preventing veno-occlusive disease and treating if necessary[62]. Haemorrhagic cystitis is a rare complication following HSCT, associated with adenovirus or BK virus infection, associated with T cell depletion or prolonged immunosuppression. Treatment is as for patients undergoing HSCT for other indications[63].

Stage	Skin % body surface area	Liver Bilirubin µmol/l	Gut Diarrhoea vol. ml/kg/day (or if >50kg body weight)
1	Maculopapular <25%	34-50	10-19.9 (or 500-1000ml/day)
2	25-50%	51-100	20-30 (or 1000-1500ml/day)
3	Generalised erythema	101-255	>30 (or>1500ml/day)
4	Exfoliation/vesicles	>256	Severe abdo pain +/- ileus

Grade	Skin stage	Liver stage	Gut stage	Clinical performance
I	1-2	0	0	normal
II	1-3	1	1	mild decrease
III	2-3	2-3	2-3	marked decrease
IV	2-4	2-4	2-4	incapacitated

Adapted from Jacobsohn DA[69]

Table 3. Staging of acute GVHD

10. Post transplantation immuno-reconstitution

Full immunoreconstitution can take up to 2 years post transplant. Most patients remain on immunoglobulin replacement for around 6 months post transplant or until evidence of immunoglobulin production (gauged by measuring IgM production). Ongoing immunosuppression or graft versus host disease will delay assessment of antibody production. Thymopoiesis can be measured by documenting an increase in recent thymic emigrants measured by TRECS or surrogate markers such as CD27 or CD31 on T cells. Thymopoiesis normally occurs around 120 days post transplant[64]. Graft versus host disease may impair or abrogate this process[65].

Once immunoglobulin production is established, immunoglobulin replacement should be discontinued. After a wash out period of 3 months, primary vaccinations with non-live antigens can begin, having assessed base line specific antibody levels. The response to

vaccine antigens can be assessed once the primary vaccination schedule is complete. Vaccination with live vaccines such as MMR should only be considered once normal T cell proliferation has been demonstrated and an antibody response to the primary vaccination schedule has been confirmed. Prophylaxis against polysaccharide coated organisms should be continued for at least 2 years post transplant[66]. Patients should however have received the conjugated pneumococcal and meningococcal vaccines. Once the response to polysaccharide organisms has been demonstrated in those with normal splenic function, antibiotic prophylaxis can be discontinued. Patients should be monitored for evidence of endocrine dysfunction - particularly thyroid dysfunction[67]. Thyroid dysfunction occurs in up to 10% of post transplant patients. Usually this is hypothyroidism but more rarely hyperthyroidism has been described and each should be managed appropriately. In the long term there may be issues with fertility due to the conditioning regimens and patients should be counselled accordingly – referral to fertility clinics or endocrine specialists may be necessary. Growth is usually normal, and growth retardation due to the underlying illness may be reversed post transplant. Ongoing care of previous end organ damage such as bronchiectasis may require specialist input with regular monitoring of lung function and radiological changes. The quality of life post transplant has not been extensively assessed in primary immune deficiency patients. One study looking at the outcome of patients transplanted for severe primary immune deficiency demonstrated an increased risk of long term cognitive difficulties with associated emotional and behavioural difficulties. Specific genetic diagnosis and a severe clinical course were specifically associated with poor outcome[68]. Conversely a recent study looking at patients with CGD found significantly better quality of life skills in those who had undergone transplantation compared to those who were not transplanted, with the post transplant patients score similar to normal controls. As more patients survive the transplant procedure and a longer follow up is achieved further work will be needed in this area to determine quality of life.

11. Future prospects

Transplantation for primary immunodeficiency is a successful treatment leading to cure of disease and normal life quality for the majority of patients. Future work will need to address optimal timing of transplantation, which may be gauged as future registry data becomes available. Less damaging conditioning regimens, particularly for newborns identified with the newborn screening programmes will become important. Treosulfan, fludarabine containing regimens are less toxic than busulphan containing regimens but targeting conditioning using radiotherapy or monoclonal antibodies may play a great role in the future. Accelerating thymopoiesis and immuno-reconstitution will be important particularly when pre-existing viral infection is present. Agents including exogenous interleukin 7 or keratinous growth factor may have a role to play. Graft manipulation may improve outcomes for some patients - better use of T cell depleted donors when no matched donors are available and particularly improving immune reconstitution through TCR alpha beta depletion may be appropriate. Expansion of stem cells in cord blood transplantation, the use

of multiple cords or the combined use of haplo-identical stem cells with an umbilical cord blood stem cell unit may also speed of immune reconstitution and lessen the risk from viral infection. Increased use of viral specific cytotoxic T cells will have a role to play. Finally better and more effective treatment of steroid resistant Graft versus host disease is needed and the use of extra corporeal phototherapy, the role of regulatory T cells and of mesenchymal stem cells needs to be explored.

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12. References

[1] Gatti RA, Meuwissen HJ, Allen HD, Hong R, Good RA. Immunological reconstitution of sex-linked lymphopenic immunological deficiency. Lancet. 1968;2(7583):1366-9. Epub 1968/12/28.

[2] Bach FH, Albertini RJ, Joo P, Anderson JL, Bortin MM. Bone-marrow transplantation in a patient with the Wiskott-Aldrich syndrome. Lancet. 1968;2(7583):1364-6. Epub 1968/12/28.

[3] Rao K, Amrolia PJ, Jones A, Cale CM, Naik P, King D, et al. Improved survival after unrelated donor bone marrow transplantation in children with primary immunodeficiency using a reduced-intensity conditioning regimen. Blood. 2005;105(2):879-85. Epub 2004/09/16.

[4] Lion T, Baumgartinger R, Watzinger F, Matthes-Martin S, Suda M, Preuner S, et al. Molecular monitoring of adenovirus in peripheral blood after allogeneic bone marrow transplantation permits early diagnosis of disseminated disease. Blood. 2003;102(3):1114-20. Epub 2003/04/19.

[5] Brown L, Xu-Bayford J, Allwood Z, Slatter M, Cant A, Davies EG, et al. Neonatal diagnosis of severe combined immunodeficiency leads to significantly improved survival outcome: the case for newborn screening. Blood. 2011;117(11):3243-6. Epub 2011/01/29.

[6] Jones LB, McGrogan P, Flood TJ, Gennery AR, Morton L, Thrasher A, et al. Special article: chronic granulomatous disease in the United Kingdom and Ireland: a comprehensive national patient-based registry. Clinical and experimental immunology. 2008;152(2):211-8. Epub 2008/04/16.

[7] Wynn RF, Boelens JJ. Bone-marrow transplantation in non-malignant disease. Lancet. 2009;374(9693):856-8. Epub 2009/09/05.

[8] Gennery AR, Slatter MA, Grandin L, Taupin P, Cant AJ, Veys P, et al. Transplantation of hematopoietic stem cells and long-term survival for primary immunodeficiencies in Europe: entering a new century, do we do better? The Journal of allergy and clinical immunology. 2010;126(3):602-10 e1-11. Epub 2010/08/03.

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[*] Corresponding Author

[9] Griffith LM, Cowan MJ, Kohn DB, Notarangelo LD, Puck JM, Schultz KR, et al. Allogeneic hematopoietic cell transplantation for primary immune deficiency diseases: current status and critical needs. The Journal of allergy and clinical immunology. 2008;122(6):1087-96. Epub 2008/11/11.

[10] Laffort C, Le Deist F, Favre M, Caillat-Zucman S, Radford-Weiss I, Debre M, et al. Severe cutaneous papillomavirus disease after haemopoietic stem-cell transplantation in patients with severe combined immune deficiency caused by common gammac cytokine receptor subunit or JAK-3 deficiency. Lancet. 2004;363(9426):2051-4. Epub 2004/06/23.

[11] Zhang Q, Davis JC, Lamborn IT, Freeman AF, Jing H, Favreau AJ, et al. Combined immunodeficiency associated with DOCK8 mutations. The New England journal of medicine. 2009;361(21):2046-55. Epub 2009/09/25.

[12] McDonald DR, Massaad MJ, Johnston A, Keles S, Chatila T, Geha RS, et al. Successful engraftment of donor marrow after allogeneic hematopoietic cell transplantation in autosomal-recessive hyper-IgE syndrome caused by dedicator of cytokinesis 8 deficiency. The Journal of allergy and clinical immunology. 2010;126(6):1304-5 e3. Epub 2010/09/03.

[13] Barlogis V, Galambrun C, Chambost H, Lamoureux-Toth S, Petit P, Stephan JL, et al. Successful allogeneic hematopoietic stem cell transplantation for DOCK8 deficiency. The Journal of allergy and clinical immunology. 2011;128(2):420-22 e2. Epub 2011/05/07.

[14] Gatz SA, Benninghoff U, Schutz C, Schulz A, Honig M, Pannicke U, et al. Curative treatment of autosomal-recessive hyper-IgE syndrome by hematopoietic cell transplantation. Bone marrow transplantation. 2011;46(4):552-6. Epub 2010/07/14.

[15] Bittner TC, Pannicke U, Renner ED, Notheis G, Hoffmann F, Belohradsky BH, et al. Successful long-term correction of autosomal recessive hyper-IgE syndrome due to DOCK8 deficiency by hematopoietic stem cell transplantation. Klinische Padiatrie. 2010;222(6):351-5. Epub 2010/11/09.

[16] Lagresle-Peyrou C, Six EM, Picard C, Rieux-Laucat F, Michel V, Ditadi A, et al. Human adenylate kinase 2 deficiency causes a profound hematopoietic defect associated with sensorineural deafness. Nature genetics. 2009;41(1):106-11. Epub 2008/12/02.

[17] Verbsky J, Thakar M, Routes J. The Wisconsin approach to newborn screening for severe combined immunodeficiency. The Journal of allergy and clinical immunology. 2012;129(3):622-7. Epub 2012/01/17.

[18] Hassan A, Booth C, Brightwell A, Veys P, Rao K, Gennery A, Slatter M, Bredius R, Finocchi A, Aiuti A, Porta F, Ridella M, Steward C, Filipovich A, Bordon V, Almuhsen S, Al-Mousa H, Speckmann C, Fischer A, Mahlaoui N, Nichols K, Grunebaum E, Honig M, Friedrich W, Boelens J, Davies EG, Roifman C, Cavazzana-Calvo M, Notarangelo L, Gaspar HB. Outcome of Haematopoietic Stem Cell Transplantation for Adenosine Deaminase Deficient Severe Combined Immunodeficiency. Blood 2012 (in press).

[19] Filipovich AH, Stone JV, Tomany SC, Ireland M, Kollman C, Pelz CJ, et al. Impact of donor type on outcome of bone marrow transplantation for Wiskott-Aldrich syndrome: collaborative study of the International Bone Marrow Transplant Registry and the National Marrow Donor Program. Blood. 2001;97(6):1598-603. Epub 2001/03/10.

[20] Moratto D, Giliani S, Bonfim C, Mazzolari E, Fischer A, Ochs HD, et al. Long-term outcome and lineage-specific chimerism in 194 patients with Wiskott-Aldrich syndrome

treated by hematopoietic cell transplantation in the period 1980-2009: an international collaborative study. Blood. 2011;118(6):1675-84. Epub 2011/06/11.

[21] Shin CR, Kim MO, Li D, Bleesing JJ, Harris R, Mehta P, et al. Outcomes following hematopoietic cell transplantation for Wiskott-Aldrich syndrome. Bone marrow transplantation. 2012. Epub 2012/03/20.

[22] Khawaja K, Gennery AR, Flood TJ, Abinun M, Cant AJ. Bone marrow transplantation for CD40 ligand deficiency: a single centre experience. Archives of disease in childhood. 2001;84(6):508-11. Epub 2001/05/23.

[23] Gennery AR, Khawaja K, Veys P, Bredius RG, Notarangelo LD, Mazzolari E, et al. Treatment of CD40 ligand deficiency by hematopoietic stem cell transplantation: a survey of the European experience, 1993-2002. Blood. 2004;103(3):1152-7. Epub 2003/10/04.

[24] Seger RA, Gungor T, Belohradsky BH, Blanche S, Bordigoni P, Di Bartolomeo P, et al. Treatment of chronic granulomatous disease with myeloablative conditioning and an unmodified hemopoietic allograft: a survey of the European experience, 1985-2000. Blood. 2002;100(13):4344-50. Epub 2002/10/24.

[25] Soncini E, Slatter MA, Jones LB, Hughes S, Hodges S, Flood TJ, et al. Unrelated donor and HLA-identical sibling haematopoietic stem cell transplantation cure chronic granulomatous disease with good long-term outcome and growth. British journal of haematology. 2009;145(1):73-83. Epub 2009/02/19.

[26] Chiesa R, Gilmour K, Qasim W, Adams S, Worth AJ, Zhan H, et al. Omission of in vivo T-cell depletion promotes rapid expansion of naive CD4+ cord blood lymphocytes and restores adaptive immunity within 2 months after unrelated cord blood transplant. British journal of haematology. 2012;156(5):656-66. Epub 2012/01/10.

[27] Fernandes JF, Rocha V, Labopin M, Neven B, Moshous D, Gennery AR, et al. Transplantation in patients with SCID: mismatched related stem cells or unrelated cord blood? Blood. 2012;119(12):2949-55. Epub 2012/02/07.

[28] Ouachee-Chardin M, Elie C, de Saint Basile G, Le Deist F, Mahlaoui N, Picard C, et al. Hematopoietic stem cell transplantation in hemophagocytic lymphohistiocytosis: a single-center report of 48 patients. Pediatrics. 2006;117(4):e743-50. Epub 2006/03/22.

[29] Leiding JW, Freeman AF, Marciano BE, Anderson VL, Uzel G, Malech HL, et al. Corticosteroid therapy for liver abscess in chronic granulomatous disease. Clinical infectious diseases : an official publication of the Infectious Diseases Society of America. 2012;54(5):694-700. Epub 2011/12/14.

[30] McDonald GB, Slattery JT, Bouvier ME, Ren S, Batchelder AL, Kalhorn TF, et al. Cyclophosphamide metabolism, liver toxicity, and mortality following hematopoietic stem cell transplantation. Blood. 2003;101(5):2043-8. Epub 2002/10/31.

[31] Kroger N, Schetelig J, Zabelina T, Kruger W, Renges H, Stute N, et al. A fludarabine-based dose-reduced conditioning regimen followed by allogeneic stem cell transplantation from related or unrelated donors in patients with myelodysplastic syndrome. Bone marrow transplantation. 2001;28(7):643-7. Epub 2001/11/13.

[32] Slavin S, Nagler A, Naparstek E, Kapelushnik Y, Aker M, Cividalli G, et al. Nonmyeloablative stem cell transplantation and cell therapy as an alternative to conventional bone marrow transplantation with lethal cytoreduction for the treatment

of malignant and nonmalignant hematologic diseases. Blood. 1998;91(3):756-63. Epub 1998/02/03.

[33] Malar R, Sjoo F, Rentsch K, Hassan M, Gungor T. Therapeutic drug monitoring is essential for intravenous busulfan therapy in pediatric hematopoietic stem cell recipients. Pediatric transplantation. 2011;15(6):580-8. Epub 2011/07/09.

[34] Straathof KC, Rao K, Eyrich M, Hale G, Bird P, Berrie E, et al. Haemopoietic stem-cell transplantation with antibody-based minimal-intensity conditioning: a phase 1/2 study. Lancet. 2009;374(9693):912-20. Epub 2009/09/05.

[35] Ritchie DS, Seymour JF, Roberts AW, Szer J, Grigg AP. Acute left ventricular failure following melphalan and fludarabine conditioning. Bone marrow transplantation. 2001;28(1):101-3. Epub 2001/08/11.

[36] Burroughs LM, Storb R, Leisenring WM, Pulsipher MA, Loken MR, Torgerson TR, et al. Intensive postgrafting immune suppression combined with nonmyeloablative conditioning for transplantation of HLA-identical hematopoietic cell grafts: results of a pilot study for treatment of primary immunodeficiency disorders. Bone marrow transplantation. 2007;40(7):633-42. Epub 2007/07/31.

[37] Sjoo F, Hassan Z, Abedi-Valugerdi M, Griskevicius L, Nilsson C, Remberger M, et al. Myeloablative and immunosuppressive properties of treosulfan in mice. Experimental hematology. 2006;34(1):115-21. Epub 2006/01/18.

[38] Bacher U, Klyuchnikov E, Wiedemann B, Kroeger N, Zander AR. Safety of conditioning agents for allogeneic haematopoietic transplantation. Expert opinion on drug safety. 2009;8(3):305-15. Epub 2009/06/10.

[39] Akasheh MS, Freytes CO, Vesole DH. Melphalan-associated pulmonary toxicity following high-dose therapy with autologous hematopoietic stem cell transplantation. Bone marrow transplantation. 2000;26(10):1107-9. Epub 2000/12/07.

[40] Bruno B, Souillet G, Bertrand Y, Werck-Gallois MC, So Satta A, Bellon G. Effects of allogeneic bone marrow transplantation on pulmonary function in 80 children in a single paediatric centre. Bone marrow transplantation. 2004;34(2):143-7. Epub 2004/06/01.

[41] Horn B, Baxter-Lowe LA, Englert L, McMillan A, Quinn M, Desantes K, et al. Reduced intensity conditioning using intravenous busulfan, fludarabine and rabbit ATG for children with nonmalignant disorders and CML. Bone marrow transplantation. 2006;37(3):263-9. Epub 2005/12/06.

[42] Jacobsohn DA, Duerst R, Tse W, Kletzel M. Reduced intensity haemopoietic stem-cell transplantation for treatment of non-malignant diseases in children. Lancet. 2004;364(9429):156-62. Epub 2004/07/13.

[43] Seger RA. Modern management of chronic granulomatous disease. British journal of haematology. 2008;140(3):255-66. Epub 2008/01/26.

[44] Slatter MA, Rao K, Amrolia P, Flood T, Abinun M, Hambleton S, et al. Treosulfan-based conditioning regimens for hematopoietic stem cell transplantation in children with primary immunodeficiency: United Kingdom experience. Blood. 2011;117(16):4367-75. Epub 2011/02/18.

[45] Schulz AS, Glatting G, Hoenig M, Schuetz C, Gatz SA, Grewendorf S, et al. Radioimmunotherapy-based conditioning for hematopoietic cell transplantation in

children with malignant and nonmalignant diseases. Blood. 2011;117(17):4642-50. Epub 2011/02/18.

[46] Czechowicz A, Kraft D, Weissman IL, Bhattacharya D. Efficient transplantation via antibody-based clearance of hematopoietic stem cell niches. Science. 2007;318(5854):1296-9. Epub 2007/11/24.

[47] Chen J, Larochelle A, Fricker S, Bridger G, Dunbar CE, Abkowitz JL. Mobilization as a preparative regimen for hematopoietic stem cell transplantation. Blood. 2006;107(9):3764-71. Epub 2006/01/28.

[48] Laurenti L, Sora F, Piccirillo N, Chiusolo P, Cicconi S, Rutella S, et al. Immune reconstitution after autologous selected peripheral blood progenitor cell transplantation: comparison of two CD34+ cell-selection systems. Transfusion. 2001;41(6):783-9. Epub 2001/06/16.

[49] Schumm M, Depletion of TcRab+ and CD19-positive cells from leukapheresis products with the CliniMACS device. Bone Marrow Transplant. 2011;46: 339 Suppl 1:P1093.

[50] Neven B, Leroy S, Decaluwe H, Le Deist F, Picard C, Moshous D, et al. Long-term outcome after hematopoietic stem cell transplantation of a single-center cohort of 90 patients with severe combined immunodeficiency. Blood. 2009;113(17):4114-24. Epub 2009/01/27.

[51] Recher M, Berglund LJ, Avery DT, Cowan MJ, Gennery AR, Smart J, et al. IL-21 is the primary common gamma chain-binding cytokine required for human B-cell differentiation in vivo. Blood. 2011;118(26):6824-35. Epub 2011/11/01.

[52] Borghans JA, Bredius RG, Hazenberg MD, Roelofs H, Jol-van der Zijde EC, Heidt J, et al. Early determinants of long-term T-cell reconstitution after hematopoietic stem cell transplantation for severe combined immunodeficiency. Blood. 2006;108(2):763-9. Epub 2006/07/11.

[53] Kane L, Gennery AR, Crooks BN, Flood TJ, Abinun M, Cant AJ. Neonatal bone marrow transplantation for severe combined immunodeficiency. Archives of disease in childhood Fetal and neonatal edition. 2001;85(2):F110-3. Epub 2001/08/23.

[54] Myers LA, Patel DD, Puck JM, Buckley RH. Hematopoietic stem cell transplantation for severe combined immunodeficiency in the neonatal period leads to superior thymic output and improved survival. Blood. 2002;99(3):872-8. Epub 2002/01/25.

[55] Qasim W, Derniame S, Gilmour K, Chiesa R, Weber M, Adams S, et al. Third-party virus-specific T cells eradicate adenoviraemia but trigger bystander graft-versus-host disease. British journal of haematology. 2011;154(1):150-3. Epub 2011/04/20.

[56] Cole TS, Johnstone IC, Pearce MS, Fulton B, Cant AJ, Gennery AR, et al. Outcome of children requiring intensive care following haematopoietic SCT for primary immunodeficiency and other non-malignant disorders. Bone marrow transplantation. 2012;47(1):40-5. Epub 2011/03/02.

[57] Engelhardt KR, McGhee S, Winkler S, Sassi A, Woellner C, Lopez-Herrera G, et al. Large deletions and point mutations involving the dedicator of cytokinesis 8 (DOCK8) in the autosomal-recessive form of hyper-IgE syndrome. The Journal of allergy and clinical immunology. 2009;124(6):1289-302 e4. Epub 2009/12/17.

[58] Booth C, Gilmour KC, Veys P, Gennery AR, Slatter MA, Chapel H, et al. X-linked lymphoproliferative disease due to SAP/SH2D1A deficiency: a multicenter study on the

manifestations, management and outcome of the disease. Blood. 2011;117(1):53-62. Epub 2010/10/12.

[59] Seidel MG, Fritsch G, Lion T, Jurgens B, Heitger A, Bacchetta R, et al. Selective engraftment of donor CD4+25high FOXP3-positive T cells in IPEX syndrome after nonmyeloablative hematopoietic stem cell transplantation. Blood. 2009;113(22):5689-91. Epub 2009/05/30.

[60] White H, Thrasher A, Veys P, Kinnon C, Gaspar HB. Intrinsic defects of B cell function in X-linked severe combined immunodeficiency. European journal of immunology. 2000;30(3):732-7. Epub 2000/03/31.

[61] Crooks BN, Taylor CE, Turner AJ, Osman HK, Abinun M, Flood TJ, et al. Respiratory viral infections in primary immune deficiencies: significance and relevance to clinical outcome in a single BMT unit. Bone marrow transplantation. 2000;26(10):1097-102. Epub 2000/12/07.

[62] Corbacioglu S, Cesaro S, Faraci M, Valteau-Couanet D, Gruhn B, Rovelli A, et al. Defibrotide for prophylaxis of hepatic veno-occlusive disease in paediatric haemopoietic stem-cell transplantation: an open-label, phase 3, randomised controlled trial. Lancet. 2012;379(9823):1301-9. Epub 2012/03/01.

[63] Harkensee C, Vasdev N, Gennery AR, Willetts IE, Taylor C. Prevention and management of BK-virus associated haemorrhagic cystitis in children following haematopoietic stem cell transplantation--a systematic review and evidence-based guidance for clinical management. British journal of haematology. 2008;142(5):717-31. Epub 2008/06/11.

[64] Muller SM, Kohn T, Schulz AS, Debatin KM, Friedrich W. Similar pattern of thymic-dependent T-cell reconstitution in infants with severe combined immunodeficiency after human leukocyte antigen (HLA)-identical and HLA-nonidentical stem cell transplantation. Blood. 2000;96(13):4344-9. Epub 2000/12/09.

[65] Fallen PR, McGreavey L, Madrigal JA, Potter M, Ethell M, Prentice HG, et al. Factors affecting reconstitution of the T cell compartment in allogeneic haematopoietic cell transplant recipients. Bone marrow transplantation. 2003;32(10):1001-14. Epub 2003/11/05.

[66] Slatter MA, Bhattacharya A, Flood TJ, Spickett GP, Cant AJ, Abinun M, et al. Polysaccharide antibody responses are impaired post bone marrow transplantation for severe combined immunodeficiency, but not other primary immunodeficiencies. Bone marrow transplantation. 2003;32(2):225-9. Epub 2003/07/03.

[67] Slatter MA, Gennery AR, Cheetham TD, Bhattacharya A, Crooks BN, Flood TJ, et al. Thyroid dysfunction after bone marrow transplantation for primary immunodeficiency without the use of total body irradiation in conditioning. Bone marrow transplantation. 2004;33(9):949-53. Epub 2004/03/09.

[68] Titman P, Pink E, Skucek E, O'Hanlon K, Cole TJ, Gaspar J, et al. Cognitive and behavioral abnormalities in children after hematopoietic stem cell transplantation for severe congenital immunodeficiencies. Blood. 2008;112(9):3907-13. Epub 2008/07/23.

[69] Jacobsohn DA. Acute GVHD in children. Bone Marrow Transplant 2008;41:215-221