Haematopoietic stem cell transplantation

In patients with sickle cell anaemia

Françoise Bernaudin, Eliane Gluckman
1. Why is haematopoietic stem cell transplantation recommended in sickle cell anaemia

Sickle cell anaemia (SCA) is a severe recessive genetic disorder resulting from a single nucleotide substitution in codon 6 of the β-globin gene, which, in the homozygous state, produces abnormal haemoglobin that is prone to polymer formation under deoxygenated conditions. The polymerised haemoglobin leads to decreased red blood cell deformability and sickling within end arterioles, resulting in vaso-occlusion and pain (1). SCA is associated with high-risk complications, including strokes, acute chest syndrome and recurrent severe painful vaso-occlusive episodes with increased risk of early mortality and significant morbidity (2-4). To date, more than 250 children and young adults worldwide have undergone haematopoietic stem cell transplantation (HSCT) (5-12), a rather limited number of patients considering that hundreds of thousands of individuals worldwide are affected with SCA. Despite progress made in SCA management, such as the prevention of pneumococcal infections (13-16), introduction of hydroxyurea therapy (17-22), and early cerebral vasculopathy detection with transcranial Doppler ultrasound (24-26), SCA remains a disease with high risk of morbidity and early death. A recent report concerning the Dallas newborn SCA cohort (1983-2002) mentioned a rate of death of 0.59 per 100 patient-years with a probability of death before the age of 18 years of 6.4% (4). Hydroxyurea therapy, used in SCA children since 1992, has allowed a significant reduction in the rate of vaso-occlusive crises, acute chest syndrome and transfusion requirements in most patients (17-19); nevertheless, its effectiveness on cerebrovasculopathies has been debated (20, 22, 27). Cerebral vasculopathy is the most important complication affecting SCA children, with a risk of stroke occurrence of 11% before the age of 18 years (4, 28). Overt strokes are related to macrovasculopathy involving the large arteries of the circle of Willis that can be explored with MR angiography (MRA). However, the use of transcranial Doppler ultrasonography (TCD), which measures blood flow velocity in the large arteries of the circle of Willis, has come to be more valuable in young children because it can identify patients at risk of stroke before MRA can detect the occurrence of stenosis (24-26, 29, 30). Children with mean maximum cerebral blood flow velocities of 2m/sec or greater have a 40% risk of stroke within 36 months (24, 29), which can be reduced by 90% via a transfusion program, as shown by the Stroke Prevention Trial in Sickle Cell Anaemia (STOP 1) (1995-2000) (31). However, the STOP 2 study showed that discontinuation of transfusion resulted in a high rate of strokes and reversion to abnormal velocities, even in patients whose velocities normalised after a 30-month transfusion program and with normal MRA (32). In addition to overt strokes, microvasculopathy can result in silent infarcts, which are defined as the presence...
of ischaemic lesions on MRI in absence of clinical deficits and are thought to compromise cognitive functioning (33-36).

The team at the Créteil Reference Centre has been following from birth an SCA cohort (n=217, period: 1988-2007) comprised of patients who were studied annually with TCD from the age of 12-18 months, starting in 1992, and who received chronic transfusion as soon as velocities were detected as abnormal. In this cohort, the probability of strokes by the age of 18 years was markedly reduced from the previously published 11% to 1.9% (37). The number of patients at risk was considerable since the probability of having an abnormal TCD was 29.7%, with a “plateau” at 8 years of age (37). G6PD deficiency, absence of α-thalassaemia and the degree of haemolysis were significant and independent risk factors for abnormal TCD (38). In addition, despite very satisfactory results regarding the prevention of overt strokes, the probability of silent strokes before 18 years of age remained at 42.8% (36). Taken into account both macro- and micro-vasculopathy, the probability of exhibiting cerebrovascular disease (abnormal MRA and/or MRI) was around 50% by the age of 18 years. Thus, despite progress in management, SCA remains a very severe disease, justifying the use of more intensive therapies to preserve cognitive functioning.

2. HSCT in SCA: Procedure and results

Allogeneic HSCT is the only curative treatment for SCA; nevertheless, its use has been limited so far by reports of transplant-related mortality (TRM) risks. The first transplants for SCA began in 1984 (5-7). Worldwide experience with pre-transplant myeloablative conditioning (Table 1) showed similar results with an overall TRM risk for the US (n=50), Belgium (n=50) and French teams (n=87) of 7% and an event-free survival of 82-86% (8-12). As the worldwide experience with transplantation for SCA has expanded, the use of HSCT has changed from an experimental intervention reserved for the most severely affected patients to a treatment that is offered to increasingly younger children with early signs of SCA-related morbidity.

The recently reported French experience (12) showed that the outcome of the procedure improved significantly with time as shown by the event-free survival among patients transplanted after January 2000 (Figure 1) with a conditioning regimen consisting of intravenous busulfan, cyclophosphamide (total dose: 200 mg/kg) and rabbit antithymocyte globulin (ATG; total dose:20 mg/kg) was 95.3%. This conditioning regimen was well tolerated, with no case of vaso-occlusive disease and only one death during aplasia (12). The significant improvement was correlated with the use of ATG, which significantly reduced the graft rejection rate from 22.6% to 2.9%, the use of cord blood, which reduced the rate of graft-versus-host disease...
(GvHD), and earlier implementation of HSCT, before severe organ damage had occurred. These results show that HSCT does not expose children to a higher risk of death than SCA itself, while offering radically improved quality of life, given that a recent study of SCA children in the US showed that 6.4% of SCA patients still die before the age of 18 years (4). Contrary to HSCT for haematologic malignancies, the risk of mixed chimerism, rejection and non-engraftment is high in SCA because of the patients’ normal immunocompetence, highly proliferative bone marrow, and immunisation by multiple transfusions. Several reports have shown that mixed chimerism, even with a minority of donor cells, can have a significant beneficial effect (9, 11, 12) if chimerism remains stable; however, previous experience with non-myeloablative conditioning showed that chimerism progressively decreases, leading to rejection (39-41). Considering the very high risk of rejection after non-myeloablative procedures (42), only myeloablative transplantations should be considered in children with no organ failure.

<table>
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<tr>
<th>Table 1: Worldwide experience of HSCT for SCA</th>
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<tr>
<td><strong>Belgium</strong></td>
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<td><strong>Period</strong></td>
</tr>
<tr>
<td><strong>Patients (n)</strong></td>
</tr>
<tr>
<td><strong>Age (range)</strong></td>
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<tr>
<td><strong>Strokes n (%)</strong></td>
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<td><strong>Ferritin</strong></td>
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<td><strong>mean ng/mL range</strong></td>
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<tr>
<td><strong>ATG source</strong></td>
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<tr>
<td><strong>dose</strong></td>
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<tr>
<td><strong>Follow up (range)</strong></td>
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<tr>
<td><strong>&gt; 2 years FU</strong></td>
</tr>
<tr>
<td><strong>Rejections</strong></td>
</tr>
<tr>
<td><strong>TRM</strong></td>
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<tr>
<td><strong>EFS</strong></td>
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<td><strong>cGVH</strong></td>
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Despite preventive measures such as anticonvulsant prophylaxis, strict control of hypertension, swift magnesium replacement, and an increase in the red blood cell and platelet transfusion thresholds to 9 g/dL and 50,000/mm$^3$ (43), respectively, seizures and posterior leukoencephalopathy, albeit reversible, remained a particularly frequent adverse effect of cyclosporin and steroid therapy (12). Replacing cyclosporine in 2002 by mycophenolate mofetil in case of GvHD requiring steroid therapy resulted in significant reduction of the rate of these complications in the Créteil cohort (12).

In contrast to the early neurological toxicity observed after HSCT, the outcome of pre-existing cerebrovascular disorders was highly favorable after transplantation (11, 12). In particular, in the patients successfully engrafted, no new ischaemic lesions were observed by MRI after transplantation, even in patients with cerebral arteriopathy, showing that the most important risk factor for stroke is SCA itself and not the associated macrovascular disease (12). Moreover, rapid normalisation of arterial velocities was observed in two patients whose TCD values had remained abnormal despite a 3-year transfusion program, suggesting that HSCT is more effective than transfusion for cerebrovascular disorders and supporting that it be offered to patients as soon as velocities fail to normalise on transfusion programs (12). The possibility of diagnosing cerebrovascular disorders by means of TCD has
improved the early detection of stenosis and primary stroke prevention by transfusion programs (24, 31). Therefore, it seems reasonable to recommend HSCT as soon as stenosis is detected and before a stroke occurs, especially as it is not safe to discontinue transfusion programs in patients with a history of abnormal TCD, even in those whose velocities and MRA had normalised (Table 2).

The principal early complication in our patients was GvHD, which was responsible for the majority of observed deaths (12). There was a significant relationship between GvHD and older age, suggesting that HSCT should be offered before the age of 15 years; however, more recent data show that patients over 15 years of age can be successfully transplanted (12). Although this experience is limited, it suggests that stem cell grafting could also be proposed to young adults who had non-severe disease during childhood but developed severity criteria during adulthood, such as a tricuspid regurgitant jet velocity of at least 2.5 m per second (44). The risk of GvHD was significantly reduced when using cord blood for transplantation, which should be encouraged whenever possible (45, 46).

The principal long-term deleterious effect of transplantation was the risk of infertility, indicating that cryopreservation of ovaries should be recommended in girls. All boys developed normal spontaneous puberty after transplantation. Most of the girls required oestrogen and progesterone treatment to induce puberty, although girls undergoing HSCT at a younger age are more likely to develop spontaneous puberty (47).

The use of unrelated cord blood for SCA patients without related donors would be a logical approach (48). However, a review of US studies where unrelated 4/6 HLA-

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<th>Table 2: Current indications for HSCT in patients with SCA (includes SS and Sβ0 patients)</th>
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<td><strong>Young SCA patients with genoidentical donor and:</strong></td>
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<tr>
<td><strong>1 or more of the following complications</strong></td>
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<tr>
<td>• Stroke without severe cognitive disabilities</td>
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<td>• Stenoses or occlusions on cerebral MRA (magnetic resonance angiography)</td>
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<tr>
<td>• Ischaemic lesions demonstrated by cerebral MRI (magnetic resonance imaging)</td>
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<tr>
<td>• Recurrent vaso-occlusive crises and/or acute chest syndromes and/or priapism despite hydroxyurea therapy</td>
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<tr>
<td>• Osteonecrosis of multiple joints</td>
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<tr>
<td>• Red cell immunisation with 2 or more antibodies</td>
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<tr>
<td><strong>Or with 1 or more severe risk factors</strong></td>
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<tr>
<td>• Abnormal high velocities on transcranial Doppler</td>
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<tr>
<td>• Severe chronic anaemia (Hb &lt; 7g/dL)</td>
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<tr>
<td>• Tricuspid jet regurgitation &gt; 2.5 m/sec on cardiac Doppler</td>
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matched cord bloods were used reports rather disappointing results in terms of GvHD and rejection rates (49). Cord blood banking must therefore be encouraged in this setting to increase the possibility of finding a well-matched unrelated donor, which is currently very low (50). Young patients qualifying for transplantation but lacking an HLA-identical donor are likely to be the first candidates for gene therapy in the near future (51).

3. Geno-identical HSCT in SCA: Why not more?
Since allogeneic HSCT is a curative therapy for patients with SCA, one can wonder why only a small fraction of patients receive this treatment, especially considering that the TRM risk is no greater than the risk of SCA-related mortality. In a paper published in 1996 (52), it was reported that only 6.5% of SCA patients had criteria for SCT and that 14% of those meeting entry criteria for SCT had an HLA-identical sibling but a wide variation was observed among the institutions (0.9-36%), suggesting that other barriers such as parental and/or physician refusal, lack of financial or psychosocial support are operative.
In order to increase the chance of having HLA-identical donors, sibling cord blood cryopreservation should be systematically offered to families and pre-implantation genetic diagnosis coupled with HLA selection discussed (53) with the parents. In Créteil, where cord blood cryopreservation has been systematically proposed to families with a SCA patient since 1998, the probability of having an indication for HSCT before the age of 18 years and an HLA-identical sibling was 35%. Considering this experience from one centre and the fact that 250 babies are born with SCA each year in France, about 80 geno-identical HSCT for this disease could be performed per year in France.
Considering the hope of curing 95% of SCA children with geno-identical HSCT, thereby preventing end-organ failure and preserving cognitive functioning that favours the future socio-professional insertion, this therapeutic approach should be discussed early with families and regarded as standard of care for children who have a suitable sibling-matched donor and SCA-related disabling complications such as cerebral vasculopathy or frequent vaso-occlusive crises.

References
20. Ware RE, Zimmerman SA, Schultz WH. Hydroxyurea as an alternative to blood transfusions.


Multiple Choice Questionnaire

To find the correct answer, go to http://www.esh.org/iron-handbook2009answers.htm

1. Among the following sentences concerning the occurrence of abnormal TCD in children with SCD, which are true?
   a) The risk is related to age with a 2% increase each year from 1 to 16 yrs
   b) The risk is increased when a G6PD deficiency is associated with SCD
   c) The risk is increased when alpha-thalassaemia is associated with SCD
   d) A transfusion program is necessary when there is an abnormal TCD
   e) The STOP2 study has shown that transfusion can be stopped if TCD returns to normal

   1) a+b+c+d+e ................................................................. [□]
   2) a+b+d ................................................................[□]
   3) b+d .......................................................................[□]
   4) b+c+d ...................................................................[□]

2. What is the major cause of death in young patients transplanted for sickle cell anaemia with a geno-identical donor?
   a) Rejection of the graft (linked to absence of previous chemotherapy and immunisation by transfusion) ........................................... [□]
   b) Veno-Occlusive Disease, due to the toxicity of the myeloablative conditioning regimen .......................................................... [□]
   c) Graft-versus-host disease, either acute or chronic ........................................ [□]
   d) Unusual and severe CNS toxicity of cyclosporine A observed in SCD patients .......................................................... [□]

3. Event-free survival for young patients transplanted for sickle cell anaemia with a geno-identical donor has improved with time because of:
   a) Use of non-myeloablative conditioning regimens
   b) Increased use of cord blood as stem cell source
   c) Decreasing rejection rate after addition of ATG
   d) Earlier HSCT implementation before severe organ damage has occurred

   1) a+b+c+d ................................................................. [□]
   2) a+c .......................................................................[□]
   3) b+c+d ...................................................................[□]
   4) a+c+d ...................................................................[□]
4. The risk of rejection is high in HSCT for sickle cell anaemia because of:
   a) Young age of patients at transplant
   b) Absence of previous chemotherapy
   c) Highly proliferative bone marrow
   d) High rate of transfusions before SCT

   1) a+b+c+d ..............................................................
   2) b+c+d ..............................................................
   3) a+c ..............................................................
   4) a+c+d ..............................................................

5. The cerebral vasculopathy outcome after SCT for SCA has shown:
   a) A good prevention of stroke recurrence (6%)
   b) Absence of new cerebral ischaemic lesions in successfully engrafted patients
   c) Frequent seizures
   d) Disappearance of all stenoses

   1) a+b+c+d ..............................................................
   2) b+c+d ..............................................................
   3) a+b+c ..............................................................
   4) b+c ..............................................................