Treatment of acquired aplastic anemia

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Aplastic anemia (AA) is a rare disease, with an incidence of only 1-2 per million population per year in the West. Consequently, there have been few prospective, controlled clinical trials for the different treatment options, namely immunosuppressive therapy and bone marrow transplantation, but those that have been performed are of major importance. Further assessment of treatment outcomes derives from retrospective studies from large registries such as the European Blood and Marrow Transplant (EBMT) and Centre for Blood and Marrow Transplant Research (CIBMTR) registries. Other important data derive from centers with a specialist interest in AA. Over the last 10 years, major improvements have occurred in the treatment of acquired AA. The impact of improved supportive care on outcome after both bone marrow transplantation (BMT) and immunosuppressive therapy during this time is likely to have been of major importance although difficult to quantify.

Prior to formulating a management plan for a newly diagnosed patient, careful definition of the disease is required. Other causes of pancytopenia with a hypocellular bone marrow must be excluded, especially hypocellular myelodysplastic syndrome (MDS).[1] Inherited bone marrow failure syndromes such as Fanconi's anemia and dyskeratosis congenita may be obvious clinically but not in all cases. Patients with so called *cryptic* dyskeratosis congenita characterized by mutations in telomerase genes, *TERC* or *TERT*, lack the somatic changes of classical dyskeratosis congenita and invariably fail to respond to immunosuppressive therapy.[2,3] All patients should be screened for cytogenetic and paroxysmal nocturnal hemoglobinuria clones at diagnosis;[4,5] an abnormal cytogenetic clone does not necessarily mean a diagnosis of MDS and some patients with a clone show a similar response to immunosuppressive therapy as do AA patients without a clone.[5] Assessment of disease severity according to standard criteria[1] is important for management decision making and also has prognostic significance. During this phase of disease definition, it is vital that patients receive adequate transfusional support to maintain safe blood counts. Prophylactic antibiotics and antifungal agents should be given, and fever should be treated promptly with systemic antibiotics and early introduction of systemic antifungal drugs. Hematopoietic growth factors such as granulocyte colony-stimulating factor (G-CSF) should only be used to treat a severe infection that is not responding to antibiotics, although most patients with severe AA will predictably show no increase in neutrophil count. G-CSF should not be used to *treat* the disease which would otherwise delay definitive treatment with BMT or IST, as there is no rationale for using G-CSF in this manner.[6]

Patients with severe AA, if they are <40 years old and have an HLA compatible sibling, should be transplanted up front, and should not receive prior immunosuppressive therapy[1] (Figure 1). Delay in transplantation also increases the risk of life-threatening infections. Despite this, recent studies show that up to half the patients have been treated with immunosuppressive therapy before BMT, and this, along with a longer period from diagnosis to BMT, has an adverse impact on outcome.[7-9] Standard conditioning is cyclophosphamide 200 mg/kg and antithymocyte globulin (ATG) with cyclosporine A (CSA) and methotrexate as graft-versus-host disease (GVHD) prophylaxis. There is no role for irradiation as this increases GVHD, pneumonitis, second tumors, and infertility and impairs growth and development in children. Overall survival is 80-95% with best results in younger patients.[8,10,11] In contrast, the survival rate is only 50% for those >40 years old.[12] Graft failure occurs in 5-15%; severe acute GVHD appears to occur less often. However, chronic GVHD remains a major problem in 30 - 40% of patients, and has an impact on quality of life, second tumors and mortality.[8,10,13] A recent retrospective, combined CIBMTR/EBMT study showed worse survival and more chronic GVHD when peripheral blood stem cells were used instead of bone marrow. Although neutrophil and platelet recovery occurred earlier after the peripheral blood stem cell grafts, the use of these stem cells did not reduce graft failure[14] (*personal communication from M. Eapen 2006*). Although a temporal association between the addition of ATG to cyclophosphamide and improved engraftment has been seen, the true contribution of ATG to reducing graft failure is unclear. A recent prospective, randomized CIBMTR study comparing cyclophosphamide alone or with ATG showed no difference in graft failure, although the study was under-powered.[15] Campath monoclonal antibodies have been used instead of ATG resulting in a low incidence of GVHD, especially chronic GVHD, but the graft failure rate was high at 24%, although only 14% when peri-transplant administration of Campath was avoided.[16] Donor-recipient sex mismatching affects GVHD (female donor/male recipient) and graft failure (male donor/female recipient), but the use of ATG abrogates the negative effect of sex mismatching.[17] A high degree

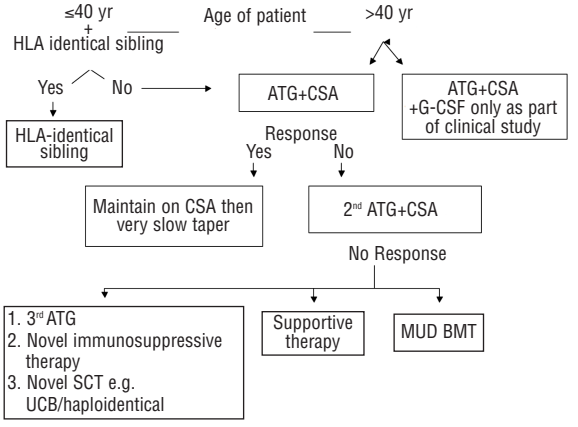

Figure 1. Algorithm for management of severe aplastic anemia. ATG: antithymocyte globulin; CSA: cyclosporin A; UCB: umbilical cord blood; MUD: matched unrelated donor; SCT: stem cell transplantation.

of mixed chimerism occurs in AA BMT.[18,19] Progressive mixed chimerism is associated with a high risk of graft failure but stable mixed chimerism results in the absence of chronic GVHD and excellent survival.[19]

In view of the higher transplant-related mortality in patients >40 years of age, new approaches to BMT are needed for this group. The use of fludarabine-based, lower dose cyclophosphamide regimens, similar to those for BMT from matched unrelated donors, is currently being explored.

Immunosuppressive therapy is indicated for (i) patients with severe AA who are >40 years old (ii) those <40 years and without an HLA-matched sibling donor and (iii) patients with non-severe AA. The standard regimen is ATG and CSA. Overall survival at 5 years is around 80%,[12] but the event-free survival rate is lower because of relapse, non-response and later clonal disorders such as MDS, acute myeloid leukemia (AML) and paroxysmal nocturnal hemoglobinuria and solid tumors.[20] Relapse occurs in up to 30% of patients.[21] The full impact of later MDS/AML requires long follow-up to 10 years.[22] In Europe, as well as in USA, it has become common practice to use G-CSF with ATG and CSA, despite lack of proven benefit. A previous EBMT prospective randomized study showed earlier neutrophil recovery and fewer serious infections when G-CSF was used with ATG and CSA, but no difference in survival or response.[23] There are concerns about an increased risk of MDS/AML when using G-CSF with ATG and CSA. Earlier studies from Japan showed a very high incidence of MDS and monosomy 7, which was associated with very high doses and prolonged duration of G-CSF therapy.[24] A large retrospective study from Italy showed no increase in MDS or abnormal cytogenetic clones when G-CSF was used at a standard dose and on average for 4 months.[25] In contrast, a recent retrospective EBMT study of more than 800 patients treated with ATG and CSA showed a non-significant increase in the cumulative incidence of MDS (5.8% vs 3.2%, p=0.17) and of AML (6.4% vs 3.2%, p=0.14) for G-CSF vs non-G-CSF-treated patients, respectively, but on Cox multivariate analysis, a hazard ratio of 2.5 for AML among patients given G-CSF (p=0.003).[26] This study emphasizes that the standard immunosuppressive therapy should be ATG and CSA alone, and that G-CSF should only be used in the context of prospective clinical trials. A second EBMT randomized study, comparing ATG and CSA with or without G-CSF, is currently ongoing. This will be a larger study than the previous one, and should answer important questions about potential advantages and disadvantages of using G-CSF.

A second course of immunosuppressive therapy is given for non-response or relapse after the first course. Response rates vary between 30-70% when immunosuppressive therapy is given for non-response[27-29] and between 60-65% when it is given for relapse.[27,28] If there is no response to two courses, it is unlikely that a third course will confer benefit, but a third course is useful when patients have previously responded to immunosuppressive therapy.[30]

Patients with non-severe AA who are transfusion-dependent have the best chance of response using ATG and CSA compared with CSA alone (74% vs 46%, respectively) and event-free survival (80% vs 51%, respectively) based on a prospective randomized EBMT study.[31] Treatment should not be delayed as recent data from EBMT, reported in this issue of the journal, shows no improvement in outcome of non-severe AA patients with time.[11] Patients should be referred promptly for consideration for combined ATG and CSA treatment.

Outcomes after unrelated BMT for severe AA have improved in the last 5 years, from the previously reported survival of around 40%.[32] Two different approaches have been employed. Deeg and colleagues have pioneered the use of low dose total body irradiation (TBI), having previously shown that cyclophosphamide with ATG is insufficient for successful engraftment but that standard dose TBI resulted in high mortality and high rates of GVHD and pneumonitis.[33] Using cyclophosphamide 200 mg/kg, 200 cGy TBI and ATG, an overall survival rate of 60% was reported for matched unrelated donor BMT, with excellent engraftment but high rates of GVHD and pulmonary toxicity. In contrast, Bacigalupo and colleagues used a non-irradiation, fludarabine-based regimen, with ATG and low dose cyclophosphamide (1200 mg/m^2), with an overall survival of 73%, a lower incidence of GVHD but a higher graft failure rate of 18%.[34] In both series, younger patients (< 16 years of age) did better. Additional maneuvers, such as alemtuzumab (campath-1H), are being explored to try to reduce GVHD and maintain

good engraftment.[35] A recent French national study has highlighted the importance of high resolution HLA matching between donor and recipient in improved outcome over time after unrelated donor BMT.[36]

The management of patients with severe AA who have no HLA matched sibling or unrelated bone marrow donor, and who have failed to respond to one (for children) or two (for adults) courses of immunosuppressive therapy, is difficult. Mycophenolate mofetil has been shown to be ineffective (i) as a single agent in refractory severe AA in an EBMT study of 17 patients[37] and (ii) as an additional agent when used with ATG and CSA; the response rate at 6 months was 30% (65% for patients with relapsed AA), similar to that recorded in the past using ATG and CSA, and the relapse rate was still 30% despite continuing mycophenolate mofetil for 18 months after ATG.[38] High dose cyclophosphamide (200 mg/kg) given without stem cell support has been advocated by one group in USA, but its use is associated with unacceptably prolonged cytopenias and severe infections including invasive fungal infections, contributing to early mortality.[39,40] Alemtuzumab is currently undergoing clinical trials at the National Institutes of Health, and will also be evaluated by the EBMT SAA Working Party. Novel transplant procedures such as haploidentical BMT[41] and double umbilical cord blood transplants warrant further investigation.[42,43]

In this issue of the journal,[11] Locasciulli and colleagues, from the EBMT SAA Working Party, report important changes in outcome after BMT and immunosuppressive therapy. Gratifyingly, outcomes after both sibling and unrelated donor BMT continue to improve with time. The authors confirm the negative effect of using irradiation in BMT for acquired severe AA. They also confirm that early transplant is an important factor in improved outcome. In contrast, for immunosuppressive therapy, although there has been an improvement in outcome in children, especially those with very severe AA (in keeping with a recent studies),[44] patients with non-severe AA showed a worse outcome with time (70% survival from 1991-1996 compared with 56% from 1997-2002) and a longer period between diagnosis to treatment. This observation is of concern and emphasizes the importance of referring new patients for at least an assessment to a center with expertise in AA. A further finding was that 80% of patients treated with immunosuppressive therapy also received G-CSF, despite the lack of evidence of benefit from this drug and the concern about risk of later MDS and AML. Over the last three decades, the EBMT SAA Working Party has uniquely contributed important data on very large numbers of patients with AA. This latest report has important new messages for all physicians who see adult or pediatric AA patients either infrequently or frequently.

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