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Targeted B-Cell Depletion Therapy in Childhood-Onset Systemic Lupus Erythematosus

Progress to Date

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Abstract

Childhood-onset systemic lupus erythematosus (SLE) is a multisystemic autoimmune disease associated with significant morbidity and mortality with lupus nephritis being a major prognostic factor. Children with SLE tend to have more severe hematologic and renal involvement compared with adults. Although the morbidity and mortality have greatly improved over the last 20 years, recent studies show that there are still associated major risks from under treatment (with resultant severe flares of disease activity) and over treatment (with additional medication adverse effects including risks of severe infection; many of these patients have inherent abnormal complement pathways).

Therapies used to treat children with SLE need to be individualized based on multiorgan involvement, severity of disease, history of disease flares, and knowledge of recent relevant clinical, hematologic, and immunologic parameters. These medications need to be the most effective treatments, allowing normal growth, development, fertility, and the avoidance of severe toxicity and future malignancies. Many toxic effects of current medications range from the well described Cushingoid features of corticosteroids to the gastrointestinal adverse effects of mycophenolate mofetil.

In vitro studies have shown that rituximab causes B-cell depletion by mechanisms involving antibody-dependent cell-mediated cytotoxicity, complement-dependent cytotoxicity, and direct signaling leading to apoptosis. As the adverse effect profile of B-cell depletion with rituximab has been well described in adults and children with oncologic and other autoimmune diseases, initial pilot studies using rituximab in patients with refractory SLE have been carried out according to different protocols. Evidence to date in open studies demonstrates that targeted B-cell depletion therapy can be safe and efficacious as an addition to standard immunosuppressant agents in refractory childhood-onset and adult-onset disease. Although there are positive outcomes in using this therapy, caution is necessary with respect to minimizing the number of doses and treatments given to reduce the incidence of developing human anti-chimeric antibodies.

The next phase for the clinical and research community are multicenter randomized controlled trials of rituximab in severe childhood SLE, such as a comparative trial of rituximab versus intravenous cyclophosphamide in patients both at presentation and with exacerbations of disease activity.

Systemic lupus erythematosus (SLE) is a multisystem disease of adults and children characterized by the production of different autoantibodies and associated with significant morbidity and mortality. Hematologic and renal involvement is more severe in childhood-onset SLE, which has an unpredictable natural history; renal involvement with lupus nephritis (LN) is one of the most important prognostic factors. Survival of children with SLE has improved during the last few decades with the use of superior immunosuppressant agents. However, despite the improvements, the morbidity remains high as a result of the disease itself and the associated adverse effects and toxicity of standard treatments.[1-7]

The etiopathogenesis of SLE is not fully understood with respect to the immunologic triggers of B-cell, T-cell, chemokine, and complement dysregulation and dysfunction, which produce a constellation of various clinical manifestations. Autoantibody formation and the presence of apoptotic cells result in immune complex and complement activation, leading to tissue injury and damage. Research in murine models of SLE has shown that B

lymphocytes play a role in this process through antibody-dependent and antibody-independent mechanisms,[8-10] and this is mirrored by the quantitative and functional B-cell abnormalities in humans (both adults and children) with SLE.[11-13] Autoantibodies contribute to autoimmunity by multiple mechanisms, including immune complex-mediated type III hypersensitivity reactions, type II antibody-dependent cytotoxicity, and by instructing innate immune cells to produce pathogenic cytokines, such as interferon-α, tumor necrosis factor (TNF), and interleukin-1.[14] Antigen presentation, T-cell activation and polarization, and dendritic-cell modulation may be autoantibody-independent B-cell functions mediated by B-cell production of immunoregulatory cytokines, chemokines, and lymphangiogenic growth factors. This basic science research emphasizing abnormal B-cell regulation has been put to the test with bench-to-bedside clinical studies targeting the B-cell compartment and thereby preventing renewal of autoantibodies and antigen presentation by pathogenic B cells.

Rituximab is a mouse-human chimeric monoclonal antibody of the immunoglobulin (IgG)1-κ type (see figure 1) with murine anti-CD20 variable sequence regions and human constant sequence regions. In view of the B-lymphocyte dysregulation in SLE, rituximab is the ideal candiate to carry out B-lymphocyte depletion. Rituximab has been proven to be safe and efficacious as a treatment used in adults and children with lymphomas and post-transplant lymphoproliferative disorders, with data regarding its efficacy in adults with rheumatoid arthritis (RA) and other autoimmune diseases.[15-19] Rituximab binds specifically to the CD20 antigen, which is a non-glycosylated tetraspanning cell membrane-embedded phosphoprotein restricted to only B-cell lineage and, although not expressed on plasma cells, is expressed on pre-B cells, immature, mature naive, pre-germinal center and germinal center mature, and memory B cells. Rituximab causes B-cell depletion by mechanisms involving antibody-dependent cell-mediated cytotoxicity, complement-dependent cytotoxicity, and direct signaling leading to apoptosis *in vitro* and mediates B-cell lysis by reducing pre-B and B lymphocytes *in vivo*.

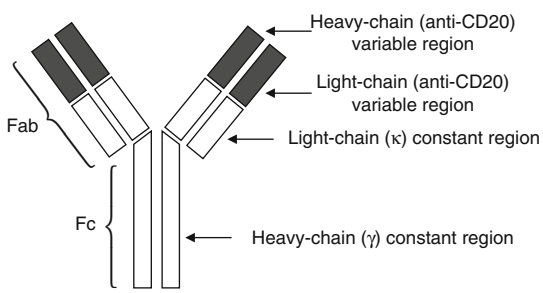

Fig. 1. Structure of rituximab: a mouse-human chimeric monoclonal antibody of the immunoglobulin (IgG) 1-κ type with murine anti-CD20 variable sequence regions (filled areas) and human constant sequence regions (open areas).

Due to the basic scientific investigations of B-cell dysregulation in SLE, as well as the safety and efficacy of B-cell depletion therapies in other disorders, initial pilot studies of rituximab have been undertaken in adults and children with refractory SLE. This article critically appraises the current available data on the use of B-cell depletion therapy with rituximab in the treatment of SLE in adults and children. Due to the paucity of data in children we initially focus on its use in adults. A literature search was performed using PubMed to identify articles on the treatment of SLE patients with rituximab up to December 2006. Papers were sought with the terms 'rituximab,' 'B cell,' 'B lymphocyte,' 'SLE,' 'systemic lupus erythematosus,' and 'lupus nephritis'. We excluded case reports, letters of one or two cases and abstracts from scientific meetings.

1. Rituximab Therapy in Adult Systemic Lupus Erythematosus (SLE)

There have been many reported case series of the use of B-cell depletion therapies in adults with SLE. We report here the evidence to date on the use of rituximab therapy in adult SLE.

There have been 105 adult patients with SLE treated with rituximab using different protocols from single infusions of 100 mg/m^2 in dose-escalation studies up to a maximum of four doses each of 500 mg/m^2 (see table I). A total of 31% (33 of 105) of patients continued other immunosuppressant agents. Some of the positive effects in those patients may have been related to either rituximab or the use of additional maintenance immunosuppressants as may the adverse effects that were reported in 33% (35 of 105) of patients. Two deaths have occurred, one due to invasive histoplasmosis, the other due to sepsis (with a total of 18 reported cases of infection). There were four reported cases of infusion-related and hypersensitivity reaction, the most severe being a serum sickness reaction in a patient who was previously exposed to a humanized monoclonal antibody (alemtuzumab). However, despite adverse reactions, these papers specify the improvement in adults with SLE treated with B-cell depletion with respect to individual systems, notably LN, cerebral lupus, arthritis, serositis, cutaneous vasculitis, mucositis, rashes, and fatigue.

Anolik et al.[21] first described the potential variability of B-cell depletion by rituximab in the treatment of 12 adults with SLE due to differing genotypes of FcγRIIIa thereby suggesting the importance of antibody-dependent cell-mediated cytotoxicity and/or apoptosis induction via FcγRIIIa-expressing effector cells in the mechanism of B-cell depletion. Further publications on this cohort identified that compared with healthy controls, SLE patients displayed several abnormalities in peripheral B-cell homeostasis prior to B-cell depletion (including naive lymphopenia, expansion of a CD27−, IgD− [double negative] population, and expansion of circulating plasmablasts), which resolved after effective B-cell depletion with rituximab and immune reconstitution.[22]

Table I. Case series of adult systemic lupus erythematosus (SLE) patients (pts) treated with rituximab

Reference	No. of pts	Rituximab regimen	Continued immunosuppression, including corticosteroid doses[a]	B-cell depletion achieved [% of pts]	Adverse effects (no. of pts)	Outcomes
Looney et al.,[20] Anolik et al.[21,22]	18 (including 3 pts who withdrew)	100 mg/m^2 × 1 (6 pts) or 375 mg/m^2 × 1 (7) or 375 mg/m^2 × 4 (5)	AZA/MMF/CsA (n = 5 pts), average daily prednisone dose reduced from 13mg to 10mg	65 (11 of 17)	Infusion-related reaction (1), infections (3), HACAs (5), unrelated (3, including TIA)	Improved SLAM scores at 2mo in 16 pts (p = 0.05)
Leandro et al.,[23,24] Cambridge et al.[25]	24 (6 in the initial pilot)	500mg × 2 + CYC (6), 1g × 2 + CYC (18; 2 pts did not receive CYC)	AZA (2), maintenance prednisolone	96 (23 of 24)	Death from sepsis (1), pancytopenia (1)	Mean BILAG global score decreased from 13.9 at baseline to 6.8 at 3mo and 5.0 at 6mo (p < 0.00001)
Vigna-Perez et al.[26]	22 with LN	500–1000mg × 2	AZA/MTx/MMF/CYC (21), unchanged corticosteroid therapy (73% [16 of 22 pts] were on corticosteroid at baseline)	91 (20 of 22)	Death from invasive histoplasmosis (1)	MEX-SLEDAI index significantly decreased in 90% of pts at 60 days (p < 0.05)
Smith et al.[27]	11 (excluding 11 with AAV)	375 mg/m^2 × 4 + CYC	MMF (4), median daily prednisolone dose decreased from 10mg to 7mg at 6mo (p = 0.003) and 5mg at 12mo (p = 0.0005)	100 (11 of 11)	Infusion-related reactions (common: mild-moderate; 1: severe), SSR (1), infection (5 SLE and AAV patients), HACAs (3)	BILAG scores decreased from a median of 14 at baseline to 3 at 6mo and 2 at 12mo (p < 0.0001)
Vallerskog et al.[28]	10 (excluding 9 with RA)	375 mg/m^2 × 4 + CYC	Maintenance corticosteroids	100 (10 of 10)	None reported	SLAM score reduced from 4–25 to 0–16 at depletion and 0–11 at 6mo post-treatment
Tokunaga et al.[29]	10 with NPSLE	375 mg/m^2 × 4 (6), 500 mg/m^2 × 4 (2), 375 mg/m^2 × 1 (1), 1g × 2 (1)	AZA (1), maintenance daily low to moderate (15–40mg) prednisolone dose	100 (10 of 10)	Infection (3: herpes/ varicella zoster virus, decubitus ulceration), pneumonia (2)	SLEDAI decreased from a median of 19.9 at baseline to 6.2 after treatment (p = 0.013)
Sfikakis et al.[30]	10 with LN	375 mg/m^2 × 4	No post-rituximab corticosteroid dose data	100 (10 of 10)	Mild infections (3: URTI/ cystitis); meningitis (1), hypersensitivity reaction (1)	Partial remission achieved in 80% (8 of 10) within 1–4mo with 5 pts achieving complete remission

a Cortocosteroid dose documented where published with titration to the patient's disease activity.

AAV = antineutrophil cytoplasmic antibody (ANCA)-associated vasculitis; **AZA** = azathioprine; **BILAG** = British Isles Lupus Assessment Group; **CsA** = ciclosporin; **CYC** = cyclophosphamide; **HACAs** = human anti-chimeric antibodies; **LN** = lupus nephritis; **MEX-SLEDAI** = Mexican Systemic Lupus Erythematosus Disease Activity; **MMF** = mycophenolate mofetil; **MTx** = methotrexate; **NPSLE** = neuropsychiatric SLE; **RA** = rheumatoid arthritis; **SLAM** = systemic lupus activity measure; **SSR** = serum sickness reaction (previously exposed to a humanized monoclonal antibody [alemtuzumab]); **TIA** = transient ischemic attack; **URTI** = upper respiratory tract infections.

Looney et al.[20] built on the findings from these initial 12 patients as part of the first phase I/II dose-escalation trial of rituximab added to ongoing therapy in 18 adults with SLE. They showed that rituximab was safe and effective for the treatment of SLE at least for the majority of patients experiencing profound B-cell depletion. Rituximab was administered as a single infusion of 100 mg/m^2 (low dose), a single infusion of 375 mg/m^2 (intermediate dose), or as four infusions (1 week apart) of 375 mg/m^2 (high dose) and was well tolerated with most patients in the cohort experiencing no significant adverse effects. There were dose-related responses with no adverse effects in the low dose treated group who had lower human antichimeric antibodies (HACAs) but required addition of other immunosuppressant agents. However, the three serious adverse events were thought to be unrelated to rituximab administration. A total of 65% (11 of 17) of patients had profound B-cell depletion (to <5 CD19+ B cells/μL). In these patients, the Systemic Lupus Activity Measure (SLAM) score, which was used as the primary outcome for clinical efficacy, was significantly improved at 2 and 3 months compared with baseline ($p = 0.0016$ and 0.0022, respectively) and persisted for 12 months, despite the absence of a significant change in anti-double-stranded DNA antibody and complement levels. In total, 33% (6 of 18) of patients developed HACAs at a level of >100 ng/mL; this was associated with African-American ancestry, higher baseline SLAM scores, reduced B-cell depletion, and lower levels of rituximab at 2 months after initial infusion.

After Leandro et al.[23] reported preliminary evidence for the safety and efficacy of B-cell depletion therapy in six female adult patients with refractory SLE, they went on to describe their longitudinal analysis of the largest studied cohort (24 patients, 6 of whom were in the pilot study).[24] Patients in the pilot study received two 500mg intravenous infusions of rituximab with the remaining patients receiving 1000mg 2 weeks apart accompanied by two 750mg intravenous cyclophosphamide infusions and two methylprednisolone infusions of 250mg each. All but two patients stopped concomitant immunosuppression. Follow-up at 3–51 (mean 23) months showed that the global British Isles Lupus Assessment Group (BILAG) global score ($p < 0.00001$ with improvements in each of the eight organs or systems), complement C3 levels ($p < 0.0005$), and anti-double-stranded DNA (dsDNA) levels ($p < 0.002$) all improved from the time of B-cell depletion to 6 months after treatment. The period of B-cell depletion ranged from 3 to 8 months (except for one patient who failed to achieve depletion and another who remained depleted after >4 years). One patient with very aggressive disease with major skin, renal, joint, and neuropsychiatric disease died from sepsis approximately 5 months after B-cell depletion; another patient had a severe reaction to the second rituximab infusion, with thrombocytopenia followed by pancytopenia, from which he recovered completely. Subsequent immunologic work by Cambridge et al.[25] on the same patients showed that B-cell clones committed to producing antinucleosome and anti-dsDNA antibodies have a relatively rapid turnover compared with B-cell clones producing other antibodies. There was also a trend toward a greater and more sustained decrease in anti-dsDNA antibodies in patients with clinical benefit lasting >1 year.

Vigna-Perez et al.[26] reported on 22 patients with active refractory SLE and LN (mainly Class III and IV LN according to the WHO classification) whose immunosuppressive regimens were augmented with two doses of rituximab 500–1000mg. They found a significant reduction in disease activity and proteinuria (both $p < 0.05$) at 60 and 90 days after rituximab therapy with 91% (20 of 22) of patients demonstrating B-cell depletion; one patient died at day 70 due to invasive histoplasmosis. These investigators assessed the function of regulatory T lymphocytes and apoptosis of immune cells and demonstrated a significant enhancement with sustained levels of different CD4+ regulatory cells (Treg, Th3 and Tr1), but not CD8+ T lymphocytes. This was accompanied by an improvement in their regulatory function as well as an unexpected increase in the apoptosis of T cells at day 30. Interestingly, the enhancement in the suppressive function of Treg cells was not observed in the two patients who showed the poorest clinical response to rituximab.

Vallerskog et al.[28] investigated the interaction between levels of BAFF (B-cell activation factor of the TNF family) and APRIL (a proliferation-inducing ligand) and B-cell frequencies in ten adult SLE patients treated with B-cell depletion with rituximab in combination with cyclophosphamide and corticosteroids. Nine patients with RA were also treated in the same way and patients were followed longitudinally for up to 6 months after B-cell repopulation. As the primary aim was to assess immunologic function, there was a paucity of clinical data, outcomes, and adverse reactions. However, the authors demonstrated increased BAFF levels during the absence of circulating B-cells in the SLE cohort with data suggesting that BAFF and APRIL are differently affected by rituximab treatment and differentially regulated in SLE compared with RA patients.

Tokunaga et al.[29] describe the clinical and laboratory findings of ten adult patients with neuropsychiatric involvement of SLE (NPSLE) treated with rituximab (administered at different doses). Rapid improvement of central nervous system-related manifestations, particularly acute confusional state, cognitive dysfunction, psychosis, and seizures were observed with lasting effects for >1 year in 50% of patients. The reported adverse effects were infections (such as pneumonia and herpes and varicella zoster viral infections) in 50% of patients who received multiple immunosuppressive therapies, including plasmapheresis.

Sfikakis et al.[30] investigated the clinical efficacy of rituximab therapy in ten adult patients with SLE and active proliferative LN. B-cell depletion lasted for 1–7 months and was well tolerated.

Table II. Case series of pediatric systemic lupus erythematosus patients (pts) treated with rituximab

Reference	No. of pts	Rituximab regimen	Continued immunosuppression, including corticosteroid doses[a]	B-cell depletion achieved [% of pts]	Adverse effects (no. of pts)	Outcomes
Marks et al.[33,34]	7	750 mg/m^2 × 2 + CYC (if none previously)	AZA/MMF (n = 5 pts), median daily prednisolone dose of 0.35 (0.06–1.95) mg/kg reduced to 0.14 (0.05–0.39) mg/kg within 6mo (p = 0.0003) and maintained at 0.13 (0.05–0.25) mg/kg at follow-up at 12mos (p = 0.0014)	100 (7 of 7)	None reported	Median BILAG scores decreased from 22 at baseline to 6 at follow-up (p = 0.002)
Willems et al.[35]	11	350–450 mg/m^2 × 2–12 infusions + CYC (2)	AZA/MTx/MMF/CYC (6), 25–50% baseline prednisolone dose	88 (7 of 8 tested pts)	Septicemia (2), lymphopenia ± neutropenia ± thrombocytopenia (6) with rash (2), impetigo (1)	Complete hematologic remission in 100% (2 of 2 pts); complete renal remission in 25% (2 of 8 pts) and partial remission in 50% (4 of 8 pts)

a Corticosteroid dose documented where published with titration to the patient's disease activity.

AZA = azathioprine; **BILAG** = British Isles Lupus Assessment Group; **CYC** = cyclophosphamide; **MMF** = mycophenolate mofetil; **MTx** = methotrexate.

Partial remission of LN was achieved in 80% of patients within a median of 2 (range 1–4) months; in five of these patients, complete remission was subsequently established (at a median of 3 months from baseline) and it was sustained at 12 months in 40% of patients. The authors demonstrated that clinical remission of LN was preceded by down-regulation of the T cell co-stimulatory molecule CD40 ligand and associated with a decrease in T helper cell activation, suggesting an additional role for B cells, independent of autoantibody production in promoting disease.

We excluded papers that report on the use of rituximab in adults with different autoimmune diseases, where SLE patients are a subgroup, because delineating treatment courses and/or adverse events related specifically to SLE patients is difficult.[31,32] However, Smith et al.[27] described 11 adult patients with active or refractory SLE in the same paper as 11 patients with active or refractory antineutrophil cytoplasmic antibody (ANCA)-associated vasculitis treated with rituximab with a single dose of intravenous cyclophosphamide. Six and five SLE patients achieved a complete and partial response, respectively. All six patients with LN had a renal response with a reduction in hematuria (p = 0.008) and proteinuria from 4.6 g/day to 0.45 g/day in 12 months (p = 0.06).[27] Clinical improvement was accompanied by significant reductions in the daily dose of prednisolone although relapse occurred in 64% of the patients with SLE. There were few infective complications although infusion reactions were common.

2. Rituximab Therapy in Pediatric SLE

In contrast to adult studies (see section 1), B-cell depletion with rituximab has been studied only in small case series in the pediatric population. The use of rituximab has been reported in a total of 18 children treated with intravenous infusions of 350–750 mg/m^2; adverse effects increased with increasing treatments up to 12 infusions (see table II).

We have now treated 21 patients with refractory childhood-onset SLE with B-cell depletion therapy in our institution and have reported on the safety and efficacy of this treatment in our initial cohort of seven (four female) patients, aged 7.7–16.1 (median 14.8) years with active SLE resistant to standard immunosuppressant agents.[33,34] Patients were treated during a 2-week period with two 750 mg/m^2 infusions of intravenous rituximab (on days 1 and 15) with intravenous cyclophosphamide (on days 2 and 16 at doses of 750mg [or 500mg if the patient's dry weight was estimated to be <50kg] if they had not received this treatment previously). Patients were treated with high-dose oral corticosteroids at doses of 30mg, 20mg and 10mg (at days 2, 3, and 4, respectively, as well as at days 16, 17, and 18, respectively). Patients continued all their other regular maintenance therapies and, after receiving the higher prednisolone doses, continued with their usual maintenance prednisolone dose.

Our standard protocol included pre-medicating with chlorphenamine and paracetamol 1 hour prior to the rituximab infusion with a dose of intravenous methylprednisolone 100mg given immediately prior to the rituximab infusion. Rituximab was diluted to the required dose with 0.9% sodium chloride or 5% glucose to a final concentration of 1–4 mg/mL and infused at 25 mg/hour and increased by increments of 25 mg/hour every 30 minutes to a maximum of 200 mg/hour as tolerated.

There were no serious adverse effects noted during the follow-up of 0.6–1.7 (median 1.0) years. The clinical manifestations of disease for which rituximab therapy was initiated were improved in all patients with significant improvement in the BILAG global scores from a median of 22 (range 14–37) at baseline to a median

of 6 (range 4–11) at follow-up (p = 0.002) [see figure 2]. In this cohort, two patients with severe multisystemic and life-threatening disease who were unresponsive to standard therapy (including plasma exchange) had renal replacement therapy successfully withdrawn following B-cell depletion and have subsequently shown further significant improvement in renal function and proteinuria.

In our cohort, there were statistically significant improvements in the patients' hematologic profiles after rituximab therapy with an increase in hemoglobin levels from a median of 8.8 (range 5.4–12.1) to 11.0 (range 9.2–13.3) g/dL (p = 0.02) and platelet count from a median of 84 (range 20–451) to 250 (range 129–471) $\times 10^9$/L (p = 0.02). There was also a trend toward an increase in complement C3 and C4 levels with a reduction in anti-dsDNA antibodies (p = 0.14, 0.3, and 0.1, respectively). All patients were screened for hypogammaglobulinemia and no patients had infections or immunoglobulin levels (e.g. IgG < 0.2 g/L) requiring intravenous replacement.

This report has been followed by a report of the largest cohort (n = 11) of children with SLE treated with different strategies of B-cell depletion (from 2 to 12 intravenous infusions of rituximab [350–450 mg/m^2/infusion]) with corticosteroids and other immunosuppressant agents (two of six patients also received cyclophosphamide); this cohort was part of a French multicenter retrospective study.[35] The authors showed that remission was achieved in six of eight patients with LN and in two patients with autoimmune cytopenia. Clinical remission lasted in all patients who responded to treatment during the follow-up of 6–26 (mean 13.2) months, except for one patient who was successfully retreated with a second course of rituximab during the follow-up period.

Although positive outcomes were reported, 'severe' adverse events were documented in 45% of the patients. We would not advocate retreatment using repeated doses of rituximab unless there was evidence of flare of disease activity after return of B cells to the peripheral circulation. In the above cohort,[35] some patients received 12 doses of rituximab with increased infective

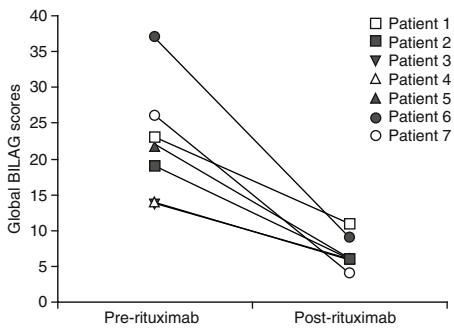

Fig. 2. British Isles Lupus Assessment Group (BILAG) global scores at baseline and end of the follow-up period in seven children with active systemic lupus erythematosus resistant to standard immunosuppressant agents treated with a rituximab regimen (p = 0.002).[33]

complications. Although there were hematologic adverse effects including lymphopenia, neutropenia, and thrombocytopenia reported in 55% (6 of 11) of patients, detection of these adverse events may vary in the literature due to different protocols of hematologic monitoring after rituximab infusions; these changes may also be due to disease activity as opposed to therapy itself. However, there was an increased incidence of infective complications with one patient treated with 11 infusions of rituximab developing septicemia on two occasions caused by *Streptococcus* spp. and then *Streptococcus bovis* with oral herpes and candida infections, and *Escherichia coli* septicemia and impetigo were reported in two other patients.

3. Future Studies

There has been ongoing drug development to discover a humanized monoclonal antibody with B-cell depletion properties. Multicenter controlled studies are being conducted as initial trials investigating B-cell depletion using the humanized anti-CD22 agent, epratuzumab, have shown that in mild to moderately active lupus, this agent was well tolerated with evidence of clinical improvement after the first infusion and durable clinical benefit across most body systems. Ocrelizumab is a second-generation humanized anti-CD20 monoclonal antibody, which is currently undergoing phase II and phase III studies in rheumatoid arthritis with future plans to recruit lupus nephritis patients.[36]

Unfortunately, blockade of the CD40-CD40 ligand pathway has resulted in only modest clinical benefit with thromboembolic complications. Other investigators are looking at blocking co-stimulatory interactions between T and B cells and inhibition of the B7 pathway; cytotoxic T lymphocyte antigen-4 immunoglobulin (CTLA-4Ig) is currently under early investigation in SLE clinical trials.

Immune cells can also be manipulated indirectly through cytokine effects, such as the promising therapeutic approach of blocking B-cell activation factor (BAFF or B-lymphocyte stimulator [BLyS]) of the tumor necrosis family. Preliminary data on the treatment of SLE with belimumab, a fully human monoclonal antibody that specifically binds to and neutralizes BAFF, showed that it is well tolerated but reportedly did not meet primary efficacy endpoints in a phase II double-blind, placebo-controlled trial.[14]

4. Conclusions

The open-label studies reviewed in this article on the use of rituximab in refractory SLE show the safety and efficacy of B-cell depletion. Targeting B cells appears to be promising in the treatment of childhood SLE, providing additional evidence for the importance and diversity of B cells in disease pathogenesis. However, there needs to be ongoing B-cell depletion to ensure no

further flares of disease activity caused by the production of abnormal B-cell clones. This required B-cell depletion may differ in individual patients due to pharmacokinetic and pharmacodynamic variation in attaining sustained therapeutic serum rituximab levels, including factors affecting drug clearance, such as HACAs. The incidence of HACAs and infusion reactions that may be attributed to antibody reactions to the chimeric drug may be reduced by the addition of other immunosuppressant agents with rituximab infusions.

Rituximab appears to have the potential to induce clinical remission of disease activity in adults and children with severe, refractory SLE who have demonstrable B-cell depletion with the additional benefit of possible clinical responses in the various organ manifestations of their disease. The mechanism of action is not fully understood but rituximab interferes with B-cell function and inhibits autoantibody production. However, autoantibody-independent functions of B cells are probably more dominant as clinical improvement correlates with B-cell depletion and precedes any decline in serum levels of relevant autoantibodies by several months.[37] The selection of patients with SLE treated with varying regimens of rituximab and other immunosuppressant agents results in variable degrees of hypogammaglobulinemia (although replacement intravenous immunoglobulin is rarely given) and may therefore result in differing incidences of infectious complications.

The use of rituximab for other conditions has been associated with adverse effects including the cytokine release syndrome, which is a well documented adverse effect in the treatment of leukemias and lymphomas due to the great tumor cell numbers.[38] Rituximab can cause stimulation of a massive release of cellular cytokines, which can have profound effects on blood pressure, vascular integrity, and myocardial, lung, liver, and kidney functions,[39-41] and can be difficult to distinguish from severe anaphylaxis and acute respiratory distress syndrome.

The US FDA issued an alert on 19 December 2006 regarding safety issues related to the use of rituximab in patients with SLE. This was because two adult SLE patients who had previously been treated with multiple immunosuppressant agents, including rituximab, died from progressive multifocal leucoencephalopathy (PML) caused by reactivation of the polyoma JC virus.[42,43] There was also a report of one adult vasculitis patient treated with multiple immunosuppressive agents, including rituximab, latterly diagnosed with PML.[43] Since rituximab is not a licensed treatment for either SLE or vasculitis, caution was advised to physicians regarding patients presenting with new-onset neurologic manifestations. The first cases of PML developing in patients treated with rituximab were in 2002 after autologous peripheral blood stem cell transplantation,[44] allogeneic stem cell transplantation,[45] and treatment for lymphoma (both PML[46] and polyoma BK virus-induced leucoencephalopathy.[47]) However, the use of rituximab after high-dose therapy with hematopoetic stem cell transplantation delays the onset of PML in HIV-negative adults with lymphoproliferative disorders.[48] As PML is lethal but rare, longer term studies are required to look at its incidence after administration of rituximab, which in the setting of SLE may cause reactivation of JC virus and the development of PML.

There has been a significant improvement in the morbidity and mortality of children with SLE in the last decade. Rituximab may be of benefit to those children who do not respond to or experience the adverse effects of conventional steroid therapy with cyclophosphamide and other immunosuppressive agents.[49-54]

Future clinical trials will ascertain the immunologic determinants of treatment response to B-cell depletion with remission and how to determine individualized therapies using other immunosuppressant agents longer term with or without further repeated courses of rituximab. There is now a need to confirm these data with multicenter randomized controlled trials of B-cell depletion against conventional therapies such as intravenous cyclophosphamide and oral mycophenolate mofetil in childhood and adult-onset SLE. Additional longer term outcome studies need to be carried out to assess the growth, development, fertility and malignant potentials in children with SLE treated with B-cell depletion therapies.

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