Patterns of B Cell Repletion Following Rituximab Therapy in a Pediatric Rheumatology Cohort

Chace Mitchell, Courtney B. Crayne, and Randy Q. Cron

Objective. To investigate the association between demographic characteristics, disease characteristics, the number of rituximab (RTX) rounds, and concurrent immunosuppression on B cell level repletion following RTX therapy.

Methods. A retrospective chart review of 112 children who met inclusion criteria and were treated with RTX at a single institution was performed. Demographic, clinical, and laboratory data were extracted and compared. CD19 levels were reviewed at 6 and 12 months post-RTX with depletion defined as fewer than 10 cells/μL and complete repopulation to normal levels defined as 170 cells/μL or more.

Results. Among patients with CD19 levels, 48% of patients remained depleted at 6 months, 89% were repleted with 10 cells/μL or more by 12 months, and 46% remained below normal levels at 12 months following infusion. There was no significant association between the number of RTX rounds or underlying disease and persistent depletion below normal levels at 12 months following RTX infusion. Depletion at 6 months was associated with a 79% chance of persistent depletion below normal levels at 12 months. The association between concurrent cyclophosphamide (CYC) and repletion of 10 cells/μL or more at 6 ($P = 0.091$) and 12 months ($P = 0.087$) trended toward significance with no significant association between association between CYC and persistent depletion below normal levels.

Conclusion. RTX therapy for pediatric rheumatic diseases is well-tolerated and results in variable repletion and normalization of B cell numbers at 6 and 12 months. B cell repletion in children is variable and independent of underlying disease and of the number of RTX infusions.

INTRODUCTION

Rituximab (RTX) is a chimeric monoclonal antibody that binds to CD20 and is found predominantly on mature B cells, resulting in reduced B cell numbers and B cell activity and subsequent immune suppression. In the United States, it is approved for use in adults with B cell malignancies (ie, non-Hodgkin lymphoma and chronic lymphocytic leukemia), rheumatoid arthritis (RA), granulomatosis with polyangiitis (GPA) and microscopic polyangiitis, and pemphigus vulgaris. Additionally, it is commonly used off-label in children with the aforementioned diagnoses as well as in patients with antibody-mediated autoimmune diseases, including systemic lupus erythematosus (SLE), multiple sclerosis (MS), immune thrombocytopenic purpura (ITP), and IgG4-related disease (1–5). Depletion of B cells leads to clinical improvement in patients with RA with return of symptoms after B cell repopulation. Time to relapse ranges from immediately upon return of B cells and as early as 2 months in some patients to several months after B cell repopulation and as long as two years in other patients. At the time of clinical relapse, autoantibodies, specifically rheumatoid factor immunoglobulin M, were elevated in RA patients, further strengthening the cause and effect association between B cell repopulation and disease activity (6,7).

RTX is generally well-tolerated. The most common adverse event reported is infusion reactions, which are thought to be related to the rate of infusion (1,8,9), but another potential adverse event is infection. Progressive multifocal leukoencephalopathy (PML) is a rare and potentially fatal disease caused by the John Cunningham virus. Incidence is reported to be 1 in 32,000 of adults treated with RTX (10). There are no published cases of PML in children treated with RTX. Hypogammaglobulinemia can occur in the months following RTX infusion with a mean onset of about 8 months (9,11–13). Replacement with intravenous immunoglobulin (IVIg) can help reduce the risk of infection (8,9,11). Late-onset neutropenia has also been reported several weeks after RTX infusion in adult patients, more often in those treated for hematologic disease and less commonly reported in patients with rheumatic disease (14–19).

Chace Mitchell, BS, Courtney B. Crayne, MD, MSPH, Randy Q. Cron, MD, PhD: University of Alabama at Birmingham.

No potential conflicts of interest relevant to this article were reported.

Address correspondence to Randy Q. Cron, MD, PhD, Children’s of Alabama, Division of Rheumatology, 1600 7th Avenue South, CPPN #G10, Birmingham, AL 35233-1711 USA. E-mail: rcr@peds.uab.edu.

Submitted for publication March 21, 2019; accepted in revised form July 24, 2019.
Although the short-term safety and efficacy of RTX in various diseases is well-established, the long-term effects of RTX, specifically on B cell repopulation, are variable. The mean time to B cell repopulation ranges from 6 to 9 months in adults with RA and connective tissue diseases (CTD; eg, SLE) with depletion defined as a CD19 count of fewer than 5 cells/\(\mu\)L to about 2 years in patients with GPA with depletion defined as a CD19 count 1 cell/\(\mu\)L or less (6,7,20,21). Among patients who received repeated infusions of RTX, the average time for B cell repopulation (ie, CD19 count greater than 5 cells/\(\mu\)L) remained closer to 8 to 9 months (6). Some patients take longer to repopulate and thus remain depleted for months to years with a maximum time reported to be 4 years in an adult with SLE (6,20). Failure to repopulate B cells can occur after repeated infusions as well as after only one round (6). Depletion responses are also variable, with reports of near depletion for up to 24 weeks in MS patients. CD19+ B cells returned to 30% of baseline value by 48 weeks (22). In a pediatric cohort of 19 children with ITP, CD19+ B cell numbers remained lower than normal for a median of 210 days (range 180-720 days) in patients still in remission and 180 days (range 120-290 days) for those who relapsed. Patients who relapsed had an average CD19 count of 172 \(\pm\) 159 cells/\(\mu\)L at 4 months and 232 \(\pm\) 164 cells/\(\mu\)L at 6 months (5).

The published literature reports B cell depletion and repopulation using a focused approach that is disease specific and is limited to adult patients. This study explores the association between various demographics and disease factors as well as the effects on B cell depletion and repopulation following RTX infusion in pediatric patients with various rheumatic diseases. The primary outcome was defined as persistent depletion below normal levels by 12 months following last RTX infusion. Secondary outcomes explored the relationships between the demographic variables, underlying disease, the number of RTX infusions, and concurrent immunosuppression (IS) and the effects on CD19 levels following RTX. Adverse events, including infections that require hospitalization and hypogammaglobulinemia, were also analyzed.

**MATERIALS AND METHODS**

A retrospective Institutional Review Board–approved chart review was performed on 118 patients who received at least one RTX infusion at Children’s of Alabama from September 2007 to May 2018. Patients who were 18 years of age or younger at the time of the first RTX infusion and those who were treated for autoimmune disease as diagnosed by a pediatric rheumatologist were included in the study. Five patients were excluded for age exceeding 18 years, and one patient was excluded for treatment of malignancy for a total of 112 patients who met inclusion criteria.

For the purpose of this study, B cell depletion was defined as a CD19 count lower than 10 cells/\(\mu\)L (23). Levels were reviewed at 6 and 12 months following any RTX infusion. Using the lower limit of normal for adolescents as reported by Valianthan et al (24), complete B cell repopulation to normal levels was defined as a CD19 level of 170 cells/\(\mu\)L or greater. Patients who had fewer than 170 cells/\(\mu\)L at 12 months or later following the last RTX infusion met criteria for persistent B cell depletion below normal levels. CD19 levels were obtained per provider discretion based on clinical disease activity prior to re-dosing RTX, and as such, CD19 levels were missing in 34 patients at 6 months and in 39 patients at 12 months. Ten patients had CD19 levels checked just prior to the 12-month mark and had a CD19 count greater than 10 but less than 170 cells/\(\mu\)L. These patients were labeled as repleted at 10 cells/\(\mu\)L or greater but missing for complete repopulation at 170 cells/\(\mu\)L or greater.

RTX was dosed per institutional practice and was given as two infusions separated by two weeks, each with a dose of 750 mg/m\(^2\) (maximum, 1 g) (8). The two doses combined comprised one round. Patients were all premedicated with intravenous methylprednisolone (30 mg/kg; maximum 1 g) to reduce the risk of an RTX infusion reaction and as treatment of the underlying disease. Determination for repeat RTX dosing was provider-dependent based on clinical disease activity and was routinely redosed if the patient had repopulation of B cells during a flare.

Diagnostic categories were labeled as CTD (ie, SLE, mixed CTD, Sjogren syndrome, and scleroderma), dermatomyositis, vasculitis (ie, GPA, Henoch-Schönlein purpura, and polyarteritis nodosa), and miscellaneous (ie, juvenile idiopathic arthritis [JIA], idiopathic pulmonary hemosiderosis, ITP, autoimmune hemolytic anemia, Castleman disease, idiopathic central nervous system vasculitis, and transverse myelitis).

Using the electronic medical record, demographic information, CD19 levels, the number of RTX infusions, diagnostic information, and adverse events were abstracted and analyzed using JMP Pro 14 statistical software (SAS Institute). Analysis of variance (ANOVA) and chi-squared tests were performed for continuous (ie, age and number of rounds) and nominal variables (ie, sex, race, B cell response, and concurrent IS), respectively, with a level of significance set at \(P < 0.05\).

**RESULTS**

Of the 112 patients who met inclusion criteria, most of the sample cohort were female (\(n = 87; 78\%\)) and nonwhite race (\(n = 79; 71\%\)). Average age at time of initial RTX infusion was 12.4 \(\pm\) 4.2 years with a range of 2 to 18 years. The mean number of RTX rounds was 2.7 \(\pm\) 2 with a range of 0.5 to 11 and a median of 2 rounds. A majority of patients received RTX for CTD (\(n = 74; 66\%\)), followed by vasculitis (\(n = 15; 13\%\)), dermatomyositis (\(n = 11; 10\%\)), and miscellaneous (\(n = 12; 11\%\)). Comparison of demographics across the five diagnostic categories showed a statistically significant difference among age means (\(P = 0.001\)), race (\(P = 0.016\)), and sex (\(P = 0.006\)). The number of RTX rounds was similar between the groups (\(P = 0.768\)) (Table 1).

At 6 months following RTX infusion, 38 of 78 (49\%) patients remained depleted with CD19 levels indicating fewer than 10
B CELL LEVELS FOLLOWING RTX

529

cells/μL and 40 of 78 (51%) patients with CD19 levels 10 cells/μL or greater. The majority of patients who remained depleted at 6 months had CTD (n = 25 of 38, 66%). By 12 months, 65 of 73 (89%) of patients repleted to levels of 10 cells/μL or greater. At 12 months, 29 of 63 (46%) patients remained depleted below normal levels with CD19 levels indicating fewer than 170 cells/μL (Table 1).

Among the patients with CTD, 25 of 53 (47%) patients remained depleted at 6 months. At 12 months, 47 (92%) of 51 patients with CTD were repleted by 12 months to CD19 levels 10 cells/μL or greater, but 23 of 44 (52%) did not repopulate to normal levels and had CD19 levels lower than 170 cells/μL. A majority of the patients with vasculitis remained depleted at 6 months (n = 7/11, 64%) but repopulated by 12 months, with 8 of 10 (80%) patients achieving levels of 10 cells/μL or greater and 5 of 7 patients (71%) gaining complete repopulation to normal levels of 170 cells/μL or greater. There was no statistical significance among patients who were depleted at 6 months (P = 0.490), 12 months (P = 0.596), or those who did not repopulate to normal levels by 12 months (P = 0.159) based on diagnosis (Table 1).

In terms of other variables, there was no association between age (P = 0.478), sex (P = 0.209), race (P = 0.295), or the number of rounds of RTX (P = 0.359) and CD19 levels below normal indicating fewer than 170 cells/μL at 12 months following RTX infusion (Table 2). Moreover, there was no association between age, sex, race, or the number of RTX rounds and B cell depletion fewer than 10 cells/μL at 6 months (Supplementary Table 1) or at 12 months (Supplementary Table 2) following infusion. The number of RTX rounds was similar among both sexes and all races (Supplementary Table 3).

Paired comparison of patients with CD19 levels available at 6 months and at 12 months showed a statistically significant agreement (P < 0.0001) with a κ of 0.64 and 95% confidence interval (0.43, 0.84) with respect to failure to replete at 6 months and persistent depletion below normal levels at 12 months. Twenty-two of 28 patients who remained depleted at 6 months

| Table 1. | Sample demographics by diagnosis (N = 112) |
|----------|------------------------------------------|
| Variable | Total (n = 112) | Connective tissue disease (n = 74) | Dermatomyositis (n = 11) | Vasculitis (n = 15) | Miscellaneous (n = 9) | P value |
| Age (years) | Mean ± SD | 12.375 ± 4.16 | 13.34 ± 3.39 | 8.64 ± 3.67 | 11.4 ± 5.38 | 11.08 ± 5.11 | 0.001a |
| Median, range | 13, 2 to 18 | 14, 2 to 18 | 7, 5 to 15 | 13, 2 to 17 | 12.5, 3 to 18 |
| Sex | | | | | | |
| Male | 25 (22.3) | 13 (17.6) | 1 (9.1) | 9 (60) | 2 (16.7) | 0.006a |
| Female | 87 (77.7) | 61 (82.4) | 10 (90.9) | 6 (40) | 10 (83.3) |
| Race | | | | | | |
| White | 33 (29.5) | 15 (20.3) | 4 (36.4) | 9 (60) | 5 (41.7) | 0.016a |
| Nonwhite | 79 (70.5) | 59 (79.7) | 7 (63.6) | 6 (40) | 7 (58.3) |
| CD19 <10 cells/μL at 6 months following RTX | | | | | | |
| Yes | 38 (48.7) | 25 (47.2) | 4 (57.1) | 3 (42.9) | 2 (28.6) | 0.490 |
| No | 40 (51.3) | 28 (52.8) | 3 (42.9) | 4 (36.4) | 5 (71.4) |
| CD19 <10 cells/μL at 12 months following RTX | | | | | | |
| Yes | 8 (11) | 4 (7.8) | 1 (25) | 2 (20) | 1 (12.5) | 0.596 |
| No | 65 (89) | 47 (92.2) | 3 (75) | 8 (80) | 7 (87.5) |
| CD19 <170 cells/μL at 12 months following RTX | | | | | | |
| Yes | 29 (46) | 23 (52.3) | 3 (60) | 2 (28.6) | 1 (14.3) | 0.159 |
| No | 34 (54) | 21 (47.7) | 2 (40) | 5 (71.4) | 6 (85.7) |
| # Rounds | Mean ± SD | 2.66 ± 1.96 | 2.80 ± 1.99 | 2.45 ± 2.50 | 2.33 ± 1.62 | 2.38 ± 1.75 | 0.768 |
| Median, range | 2, 0.5 to 11 | 2.1 to 11 | 1, 1 to 9 | 2, 0.5 to 7 | 1.75, 1 to 6 |

Abbreviation: RTX, rituximab.

*Statistical significance P < 0.05.

| Table 2. | Persistent depletion below normal levels (CD19 < 170 cells/μL) at 12 months following rituximab |
|----------|------------------------------------------|
| Demographics and # rounds of rituximab | CD19 <170 cells/μL n (%) | No (N = 34) | Yes (N = 29) | P value |
| Age | Mean ± SD | 11.7 ± 4.5 | 12.3 ± 3.3 | 0.478 |
| Median, range | 12, 2 to 18 | 13, 5 to 17 |
| Sex | Male | 9 (69) | 4 (31) | 0.209 |
| Female | 25 (50) | 25 (50) |
| Race | White | 11 (65) | 6 (35) | 0.295 |
| Nonwhite | 23 (50) | 23 (50) |
| # Rounds | Mean ± SD | 3.2 ± 2.4 | 3.7 ± 1.9 | 0.359 |
| Median, range | 2, 2.5 to 11 | 3, 0 to 9 |
with CD19 levels lower than 10 cells/μL did not repopulate to normal levels of CD19 count greater than 170 cells/μL by 12 months following RTX. Twenty-three of 28 patients who were repleted at 10 cells/μL or more at 6 months also repopulated to normal levels by 12 months following RTX (Table 3).

There were 106 of 112 (95%) patients who received concurrent IS. Overall, 62 of 112 (55%) received cyclophosphamide (CYC), 45 of 112 (40%) received mycophenolate mofetil (MMF), and 39 of 112 (35%) received methotrexate (MTX). Of the patients who received CYC, 27 of 48 (56%) patients remained depleted at 6 months ($P = 0.091$). Seven of the eight patients who failed to replete (CD19 count less than 10 cells/μL) at 12 months also received CYC ($P = 0.087$). There was no statistically significant association between concurrent CYC and repopulation to normal levels by 12 months following RTX ($P = 0.172$).

Furthermore, there was no statistically significant association between concurrent MMF or MTX and CD19 levels 10 cells/μL or greater at 6 or 12 months (Table 4).

Sixteen of 112 (14%) patients were hospitalized for non-life-threatening infections. One patient was hospitalized twice, and two patients were hospitalized three times. Of the 16 patients hospitalized for infection, 7 (44%) were depleted with CD19 counts lower than 10 cells/μL, 3 (19%) were repleted above 10 but less than 170 cells/μL, and 3 (19%) had completely repopulated to normal levels at 170 cells/μL or greater. CD19 levels were not obtained in three of the hospitalized patients. There were no other reported adverse events related to the RTX infusion or CD19 level. Unrelated to the CD19 counts, 23 of 112 (21%) patients were treated with IVg for reasons other than disease management. Three patients had confirmed hypogammaglobulinemia. In the remaining 20 patients, IgG levels were obtained and were within normal limits.

## DISCUSSION

Although current literature reports variable times to B cell repletion following RTX (5–7,20,22), this is the first study to explore the relationship between demographic variables and B cell depletion and complete repopulation frequency among pediatric patients with various rheumatic diseases. Much of the knowledge about the effects of RTX on B cell depletion is limited to data from adult patients and reports the short-term effects. Popa et al (6) reported on adults with RA who had persistent B cell depletion 12 months following RTX. Thiel et al (21) reported prolonged B cell depletion in a cohort of adults with RA, CTD, and antineutrophil cytoplasmic antibody (ANCA)-associated vasculitis over 12+ months, defining complete B cell repopulation as CD19 counts greater than 70 cells/μL. A significantly larger percentage of patients with vasculitis (more than 90%) failed to completely repopulate at 12 months compared with only 7% with RA and 12% with CTD. The median time of depletion with CD19 levels 1 cell/μL or less was 21 to 26 months in vasculitis, and with CD19 levels 5 cells/μL or less, 8 to 9 months in CTD and RA (6,7,20,21).

We report a relatively large cohort of 112 pediatric patients treated with RTX for various autoimmune diseases, including JIA, SLE, mixed CTD, dermatomyositis, vasculitis, and ITP, and the long-term effects on B cell depletion and repopulation. Of the patients with CD19 levels available, approximately half remained depleted at fewer than 10 cells/μL at 6 months, and although nearly 90% were replete at 10 cells/μL or greater at 12 months, just under half did not completely repopulate to normal levels at CD19 counts 170 cells/μL or greater at 12 months following the last RTX infusion. A majority of patients within this cohort had CTD; however, there was no statistically significant difference in patients who remained depleted at 6

### Table 3. Paired depletion (CD19 <10 cells/μL) at 6 months and persistent depletion below normal levels (CD19 <170 cells/μL) at 12 months following rituximab

| CD19 <10 cells/μL at 6 months | CD19 <170 cells/μL |
|-------------------------------|-------------------|
| Yes                           | Yes               |
| No                            | No                |
| Yes                           | 23                |
| No                            | 5                 |

*Bowker test of symmetry $P < 0.0001$.

### Table 4. Concurrent immunosuppression and CD19 levels

| CD19 <10 cells/μL at 6 months following RTX | Cyclophosphamide | Mycophenolate mofetil | Methotrexate |
|--------------------------------------------|------------------|-----------------------|-------------|
| Yes                                        | Yes (n = 62)     | Yes (n = 45)          | Yes (n = 39) |
| No                                         | No (n = 50)      | No (n = 67)           | No (n = 73) |
| P value                                    | 0.091            | 0.374                 | 0.438       |

| CD19 <10 cells/μL at 12 months following RTX | Cyclophosphamide | Mycophenolate mofetil | Methotrexate |
|-----------------------------------------------|------------------|-----------------------|-------------|
| Yes                                           | Yes (n = 62)     | Yes (n = 45)          | Yes (n = 39) |
| No                                            | No (n = 50)      | No (n = 67)           | No (n = 73) |
| P value                                       | 0.087            | 0.227                 | 0.1615      |

| CD19 <170 cells/μL at 12 months following RTX | Cyclophosphamide | Mycophenolate mofetil | Methotrexate |
|-----------------------------------------------|------------------|-----------------------|-------------|
| Yes                                           | Yes (n = 62)     | Yes (n = 45)          | Yes (n = 39) |
| No                                            | No (n = 50)      | No (n = 67)           | No (n = 73) |
| P value                                       | 0.172            | 0.297                 | 0.911       |

 Abbreviation: RTX, rituximab.
months versus 12 months across the diagnostic categories. Paired analysis at 6 months and at 12 months following RTX showed a statistically significant association between depletion at 6 months and failure to normalize at 12 months. There is a 79% chance of persistent depletion below normal CD19 levels (fewer than 170 cells/μL) if a patient fails to become replete at 10 cells/μL or greater at 6 months compared with an 18% chance of persistent depletion below normal at 12 months if repletion at 10 cells/μL or greater occurs at 6 months. This suggests that depletion at 6 months might be associated with long-term failure to completely repopulate to normal CD19 levels at 12 months. The clinical significance of complete repopulation versus partial repletion remains unclear.

There was no statistically significant association between age and race and B cell repletion. There was also no significant difference in the number of RTX rounds received among the disease groups. Furthermore, the mean number of rounds did not significantly differ between those who remained depleted with CD19 counts of fewer than 10 cells/μL both at 6 months and at 12 months following infusion or those who remained depleted below normal with CD19 levels of fewer than 170 cells/μL at 12 months following RTX infusion. This argues for a patient-dependent contribution to B cell normalization, suggesting underlying patient genetics or disease manifestations regulating B cell numbers following RTX treatment. Alternatively, additional therapeutics may influence B cell repopulation.

Although CYC is thought to reduce circulating B cells, Thiel et al and Cupps et al reported an independent association between B cell depletion and total cumulative CYC dose (21,25). The results herein for this pediatric cohort show that irrespective of underlying disease, a vast majority of patients were replete at 10 cells/μL or greater by 12 months. Just over half (55%) of this cohort received RTX in combination with CYC. There was a trend toward statistical significance in persistent B cell depletion (CD19 counts of 10 cells/μL or greater) and concurrent CYC at both 6 and 12 months, but no apparent association between CYC and incidence of complete repopulation to normal levels of B cell numbers. These results suggest that RTX and CYC together may synergistically suppress B cell numbers and result in longer durations of B cell depletion.

RTX was well-tolerated in this cohort of pediatric patients. Only 14% of the cohort were hospitalized for non-life-threatening infection while receiving RTX, which is lower than the reported infection incidence of 30% in patients receiving RTX as monotherapy (26). Of these, about 44% had CD19 levels that were lower than 10 cells/μL. There were comparable rates of patients who were replete with 10 cells/μL or more but remained below normal with CD19 levels lower than 170 cells/μL and those who did completely repopulate to normal at 12 months, suggesting that the absolute level is not as important as the presence of circulating mature B cells, and likely plasma cells, in the degree of infection susceptibility. These data are limited in that they measure only one point in time. CD19 levels were not checked regularly or consistently. It is unclear how long prior to infection these patients were depleted. Longer duration of B cell depletion may increase infection susceptibility.

Hypogammaglobulnia requiring IVIg replacement was rare in this cohort; however, immunoglobulin levels were not consistently checked, and in many patients IVIg was given as a prophylaxis prior to laboratory-confirmed hypogammaglobulnia. In the setting of prolonged B cell depletion, patients are at higher risk for serious infection. B cell levels and immunoglobulin levels should be monitored in patients treated with RTX. Some patients may require IVIg replacement therapy, particularly in the setting of prolonged B cell depletion.

The results of our study support previously published data showing that the time to B cell repopulation varies among patients. There is no strong evidence to support that the number of RTX rounds or underlying disease process predispose patients to prolonged B cell depletion; however, concurrent use of CYC may lead to prolonged B cell depletion. As CYC is cytotoxic to lymphocytes, it makes sense that a combination of RTX and CYC delays normalization of B cell numbers following treatment. This is the first study to compare demographic variables to B cell repopulation responses. There does not appear to be an association between other demographic variables, including underlying diagnosis, and B cell depletion and complete repopulation. Larger studies are needed to further explore the relationship between demographic factors and B cell repopulation.

This retrospective study has several limitations, most notably the lack of a literature consensus on the definition of depletion and repopulation. The cutoff definitions for CD19 levels for this study were chosen based on clinical practice and isolated literature (8,23). Thiel et al set the repopulation level lower at CD19 counts 70 cells/μL or higher and defined depletion as fewer than 1 cell/μL (21). Lowering the CD19 cutoff levels would likely increase sensitivity; however, the clinical significance remains unclear.

Furthermore, CD19 levels were not routinely checked within this cohort, which resulted in a large amount of missing data. Levels were most commonly obtained prior to another round of RTX. As such, the absence of CD19 levels may indicate disease remission. Charles et al reported no difference in the frequency of disease flares in adults with ANCA-associated vasculitis treated with timed RTX compared with tailored RTX infusions based on CD19 levels (27). Although regular monitoring of CD19 levels may be beneficial, the clinical significance with respect to absolute value and disease activity is unclear.

B cell response to RTX is likely multifactorial, and the physi-}

ogy of B cell depletion and repopulation is unknown. Perhaps, a CD20+ population is beneficial in helping B cells regenerate in the marrow. A larger cohort is needed to better evaluate the potential risk factors for prolonged B cell depletion.

In summary, RTX therapy for pediatric rheumatic diseases is well-tolerated and results in variable repletion and normalization of B cell numbers at 6 and 12 months. B cell repletion appears to be independent of the number of RTX rounds and of underlying disease. In contrast, co-administration of other IS thera-
pies, notably CYC, with RTX may delay B cell repopulation, thus increasing the risk for infection and resulting in hospitalization.

ACKNOWLEDGMENTS

The authors thank their co-physicians and staff for outstanding care of this patient population.

AUTHOR CONTRIBUTIONS

All authors were responsible for drafting the article or revising it critically for important intellectual content, and all authors gave final approval of the version of the article to be published.

Study conception and design. Mitchell, Crayne, Cron.

Acquisition of data. Mitchell, Crayne.

Analysis and interpretation of data. Mitchell, Crayne, Cron.

REFERENCES

1. Bernhardt MB, De Guzman MM, Grimes A, Kirk S, Nelson S, Bergsbaken J, et al. Rapid infusion of rituximab is well tolerated in children with hematologic, oncologic, and rheumatologic disorders. Pediatr Blood Cancer 2018;65.

2. Perugino CA, Stone JH. Treatment of IgG4-related disease: current and future approaches. Z Rheumatol 2016;75:881–6.

3. Mahmoud I, Jellouli M, Boukhris I, Charfi R, Ben Tekaya A, Saidane O, et al. Efficacy and safety of rituximab in the management of pediatric systemic lupus erythematosus: a systematic review. J Pediatr 2017;187:213–9.e2.

4. Brito-Zerón P, Bosch X, Ramos-Casals M, Stone JH. IgG4-related disease: advances in the diagnosis and treatment. Best Pract Res Clin Rheumatol 2016;30:261–78.

5. Popa C, Leandro MJ, Cambridge G, Edwards JC. Repeated B lymphocyte depletion with rituximab in rheumatoid arthritis over 7 yrs. Rheumatology (Oxford) 2007;46:626–30.

6. Parodi E, Nobili B, Perrotta S, Rosaria Matarese SM, Russo G, Licciardello M, et al. Rituximab (anti-CD20 monoclonal antibody) in children with chronic refractory symptomatic immune thrombocytopenic purpura: efficacy and safety of treatment. Int J Hematol 2006;84:48–53.

7. Vinod SS, Reed AB, Maxwell J, Cron RO, Stoll ML. Pediatric rheumatology infusion center: report on therapeutic protocols and infusions given over 4 years with focus on adverse events over 1 year. Pediatr Rheumatol Online J 2018;16:16.

8. Tambralli A, Beukelman T, Cron RO, Stoll ML. Safety and efficacy of rituximab in childhood-onset systemic lupus erythematosus and other rheumatic diseases. J Rheumatol 2015;42:541–6.

9. Bohra C, Sokol L, Dalia S. Progressive multifocal leukoencephalopathy and monoclonal antibodies: a review. Cancer Control 2017;24:1073274817729901.

10. Kado R, Sanders G, McCune WJ. Diagnostic and therapeutic considerations in patients with hypogammaglobulinemia after rituximab therapy. Curr Opin Rheumatol 2017;29:228–33.

11. Lim E, Tao Y, White AJ, French AR, Cooper MA. Hypogammaglobulinemia in pediatric systemic lupus erythematosus. Lupus 2013;22:1382–7.

12. Short J, Spence A. Adjuvant rituximab causes prolonged hypogammaglobulinaemia following autologous stem cell transplant for non-Hodgkin’s lymphoma. Bone Marrow Transplant 2006;38:433–6.

13. Caironi R, Grillo G, Tedeschi A, D’Avanzo G, Marenco P, Morra E. High incidence of neutropenia in patients treated with rituximab after autologous stem cell transplantation. Haematologica 2004;89:361–3.

14. Chaowatanarat K, Lee N, Grigg A, Fishie R, Firkin F. Delayed-onset neutropenia associated with rituximab therapy. Br J Haematol 2003;121:913–8.

15. Lemiéux B, Tartas S, Traulé C, Espinouse D, Theiblement C, Bouafia F, et al. Rituximab-related late-onset neutropenia after autologous stem cell transplantation for aggressive non-Hodgkin’s lymphoma. Bone Marrow Transplant 2004;33:921–3.

16. Nitta E, Itzutsu K, Sato T, Ota Y, Takeuchi K, Kamijo A, et al. A high incidence of late-onset neutropenia following rituximab-containing chemotherapy as a primary treatment of CD20-positive B-cell lymphoma: a single-institution study. Ann Oncol 2007;18:364–9.

17. Rios-Fernández R, Gutierrez-Salmerón MT, Callejas-Rubio JL, Fernández-Pugnaire M, Ortego-Centeno N. Late-onset neutropenia following rituximab treatment in patients with autoimmune diseases. Br J Dermatol 2007;157:1271–3.

18. Voog E, Morschhauser F, Solal-Celigny P. Neutropenia in patients treated with rituximab. N Engl J Med 2003;348:2691–4.

19. Thiel J, Rizzi M, Engesser M, Dufner AK, Troilo A, Lorenzetti R, et al. B cell repopulation kinetics after rituximab treatment in ANCA-associated vasculitides compared to rheumatoid arthritis, and connective tissue diseases: a longitudinal observational study on 120 patients. Arthritis Res Ther 2017;19:101.

20. Auwerx DL, Debaet EA, De Baetenaer D, De Baets D, De Baets S, De Baets B, et al. B lymphocyte depletion with rituximab in rheumatoid arthritis. Arthritis Rheum 2006;54:613–20.

21. Vinod SS, Reed AB, Maxwell J, Cron RO, Stoll ML. Pediatric rheumatology infusion center: report on therapeutic protocols and infusions given over 4 years with focus on adverse events over 1 year. Pediatr Rheumatol Online J 2018;16:16.

22. Tambralli A, Beukelman T, Cron RO, Stoll ML. Safety and efficacy of rituximab in childhood-onset systemic lupus erythematosus and other rheumatic diseases. J Rheumatol 2015;42:541–6.

23. Bohra C, Sokol L, Dalia S. Progressive multifocal leukoencephalopathy and monoclonal antibodies: a review. Cancer Control 2017;24:1073274817729901.

24. Kado R, Sanders G, McCune WJ. Diagnostic and therapeutic considerations in patients with hypogammaglobulinemia after rituximab therapy. Curr Opin Rheumatol 2017;29:228–33.

25. Lim E, Tao Y, White AJ, French AR, Cooper MA. Hypogammaglobulinemia in pediatric systemic lupus erythematosus. Lupus 2013;22:1382–7.

26. Short J, Spence A. Adjuvant rituximab causes prolonged hypogammaglobulinaemia following autologous stem cell transplant for non-Hodgkin’s lymphoma. Bone Marrow Transplant 2006;38:433–6.

27. Caironi R, Grillo G, Tedeschi A, D’Avanzo G, Marenco P, Morra E. High incidence of neutropenia in patients treated with rituximab after autologous stem cell transplantation. Haematologica 2004;89:361–3.