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Problems of methodology

Although a rapidly growing body of literature exists on this topic, clinical decision-making often remains difficult. The rationale for using the anti-CD20 antibody is not uniform, since the autoantibodies are produced by reactive, polyclonal B-lymphocytes in some autoimmune cytopenias⁹⁻¹⁰ and by monoclonal cells in others.¹¹⁻¹² Differences in abnormalities of T-cell subsets and antigen presentation may also contribute to the heterogeneity.¹³⁻¹⁴

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Rituximab for the treatment of autoimmune cytopenias

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By 2003, 23 cases had been published altogether and responses had been observed in 21; the two non-responders were reported in the only prospective study.\(^1,3,7,21\) The two larger prospective trials and the large retrospective series, however, showed response rates between 45 and 60\%.\(^18-20\) In many case reports, remissions were classified as complete responses (CR).\(^14,15,21\) whereas the systematic studies showed that CR to rituximab as single agent therapy are uncommon.\(^3,4\) The obvious explanation for these major discrepancies is that response rates estimated from pooled case reports are highly likely to be influenced by publication bias and heterogeneous or poorly defined response criteria.

For most autoimmune cytopenias, it will be unrealistic to require more than two or three well-performed phase 2 trials in order to consider a given therapy modality as evidence-based. If safety and efficacy of two or more treatment modalities have been documented in a given condition, it will often be impossible to put forward evidence-based guidelines on which therapy should be preferred. Response rates derived from pooled case reports should not be accepted as a basis for clinical practice.

**Autoimmune hemolytic anemia (AIHA)**

Warm-antibody AIHA is idiopathic in about 50% of patients, whereas an underlying or associated disorder can be identified in the remaining 50%.\(^22\) Even in warm-AIHA complicating clonal lymphoproliferative diseases such as chronic lymphocytic leukemia (CLL), the hemolysis is caused by high-affinity IgG antibodies produced by reactive, polyclonal B cells that have undergone somatic hypermutation.\(^3,7,24\) Warm-antibody AIHA is frequently associated with other autoimmune diseases, suggesting a failure of regulatory mechanisms of the immune system.\(^3,4,24\) An immune derangement is assumed to be present even in CLL.\(^3,24\) Reactive, polyclonal B-lymphocytes should, therefore, be considered the primary target cells for anti-CD20 therapy in warm-type AIHA, although killing of clonal B cells may also play a role when an underlying lymphoproliferative disease is present.\(^3,4\)

Case reports have indicated favorable responses to rituximab in refractory AIHA.\(^29-31\) In a retrospective series of five patients with a variety of underlying clonal lymphoproliferative disorders, improvement was observed in all patients and responses were classified as complete in three.\(^32\) In another retrospective study, Narat and co-authors reported on response to rituximab monotherapy in seven of 11 AIHA patients.\(^33\) Gupta and co-workers observed good responses to combination therapy with rituximab, dexamethasone and cyclophosphamide in a retrospective study of eight patients with refractory AIHA complicating CLL.\(^34\)

D’Arena and colleagues analyzed retrospectively the outcome after rituximab as single agent therapy in 14 CLL patients with AIHA and observed increased hemoglobin levels and transfusion independence in ten. Conversion to a negative direct antiglobulin test was achieved in five patients.\(^29\) The same group performed a retrospective study of 11 steroid-refractory adults with idiopathic AIHA.\(^35\) After rituximab monotherapy, all patients achieved transfusion independence and significantly increased hemoglobin levels with a mean increment of 3.3 g/dL, and remissions were classified as CR in eight patients. Favorable response rates in corticosteroid-refractory pediatric patients were observed in two retrospective\(^27,28\) and one prospective series; in the latter, Zecca and co-authors reported responses in 13 of 15 children.\(^36\) High response rates to rituximab monotherapy in childhood- and adult-type AIHA have also been reported from mixed retrospective series of autoimmune cytopenias.\(^39-41\)

Primary chronic CAD accounts for 13-15% of the cases of AIHA,\(^22,24,42\) making it an uncommon condition with a prevalence of 16 cases per million inhabitants and an incidence rate of 1 case per million per year according to a population-based study from Norway.\(^20\) Comprehensive review articles have been published recently.\(^44-46\)

In addition to complement-mediated immune hemolysis, most patients suffer from cold-induced circulatory symptoms, and exacerbations associated with conditions of acute phase reaction occur in more than 70% of the patients.\(^20,47-49\) Transfusion dependence is a frequent feature.\(^48\) Counseling on cold avoidance has been considered the mainstay of therapy, but a relatively large study showed that in more than 70% of cases, the patient and/or physician had not perceived this measure as sufficient.\(^20\) The results of treatment with corticosteroids, azathioprine, alkylating agents, interferon-α monotherapy and cladribine monotherapy have been poor.\(^20,50-52\) CAD is not associated with other autoimmune diseases.\(^20,48\) The existence of clonal B cells that produce monoclonal cold agglutinins, usually IgMκ, has been postulated for decades based on electrophoretic findings\(^11\) and such agglutinins have been verified more directly during the last decade by flow cytometric and immunohistochemical studies.\(^12,20\)

Lymphoplasmacytic lymphoma in the bone marrow is a frequent finding even in patients otherwise classified as having primary CAD.\(^12,13,20\) Although a role of polyclonal, reactive B-lymphocytes has been demonstrated in Waldenström’s macroglobulinemia\(^39\) and has not been excluded in primary CAD, the clonal B-lymphocytes should probably be considered the primary target cells for novel therapies.\(^45\)

The efficiency of rituximab as single agent therapy in primary CAD has been outlined in the methodolo-
Autoimmune thrombocytopenia (ITP)

ITP is the most common autoimmune cytopenia.\(^7\) Although the pathogenetic role of polyclonal anti-platelet IgG-antibodies and removal of antibody-labeled platelets by the spleen has been well documented, autoantibodies are not detectable in up to 50% of the patients.\(^5,6\) Abnormalities of T-cell subsets have also been demonstrated.\(^1,10\) The American as well as the British guidelines for therapy have recently been critically discussed.\(^11,12\) As pointed out by Godeau and co-authors, both sets of guidelines are based mainly on expert opinion rather than on outcomes derived from clinical trials and should not be used as the sole basis for decisions.\(^13\) Most authors agree that adult patients do not require treatment unless they either have a platelet count below 30×10^9/L, have bleeding symptoms, or are undergoing procedures likely to cause bleeding.\(^13,14\) Whereas ITP tends run a chronic course in adults, childhood ITP usually resolves spontaneously within weeks or months; however, chronic disease of the adult type is encountered in 10-20% of pediatric ITP patients.\(^4,6\)

A beneficial effect of rituximab therapy in adult and childhood ITP has been shown in retrospective series\(^15,18-20\) and prospective, single-arm trials.\(^26,75\) The largest prospective study described in a definitive publication was done by Cooper and colleagues.\(^4\) They assessed the safety and efficacy of rituximab therapy in 57 adults with ITP, all of whom had failed to respond to two or more previous treatment modalities. Thirty-one patients (54 %) responded; 18 had a CR and 13 a PR. Fifteen of 18 patients who achieved a CR maintained their response for more than 1 year, while response duration was generally shorter in patients achieving a PR. A preliminary report of an equally sized, prospective single-arm trial of rituximab in adult ITP has been provided by Godeau and co-authors, who observed very good responses in 24 of 60 patients (40%) and therapy failure in 34 (57%).\(^7\) At least 18 patients (30%) maintained a good response for more than 1 year. Several case reports have described response to re-treatment with rituximab in ITP patients who relapsed after previous rituximab therapy.\(^16\)

Pre-defined criteria for CR and PR were used in most reports; however, there was some variation in the platelet count thresholds.\(^7,9,10,17\) The most commonly used criteria are listed in Table 1. In a recent, comprehensive literature analysis, Arnold and colleagues calculated that according to pooled results of studies enrolling at least five patients, responses were achieved in 63% of the patients, with a 95% confidence interval (CI) of 53-73%; the responses were complete in 46% (CI, 30-58%); and partial in 24% (CI, 15-38%).\(^7\)

While these studies clearly document the therapeutic efficacy of rituximab in chronic, refractory ITP in...
both adults and children, we still do not know which ITP patients should receive rituximab therapy. In contrast to CAD, in which the relatively efficient treatment with rituximab can be compared to several ineffective conventional therapies even in the absence of comparative phase 3 studies, further trials are definitively needed in order to determine the place of rituximab in the treatment of ITP. Corticosteroids (or in some cases high-dose immunoglobulin) still hold their position as first-line therapy, and splenectomy remains a relatively efficient and sufficiently safe second-line treatment. Numerous options exist or may be proposed for third-line treatment, including rituximab and the new thrombopoietin receptor agonists. As compared to splenectomy, however, rituximab therapy is a non-surgical and reversible measure with a favorable safety profile and reasonably high response rates.

The question has, therefore, been raised as to whether infusions of rituximab should be preferred to splenectomy as a second-line treatment, possibly reducing the need for splenectomy. Clearly, randomized studies are warranted in order to try to answer this question, and it is hoped that the relatively high incidence rate of ITP will permit the completion of well-designed trials. Such studies are currently being performed.

**Evans’ syndrome**

Evans’ syndrome is a rare condition defined by the combination of AIHA and ITP in the absence of any underlying disease. Typically, the disease runs a chronic course in both adults and children and is characterized by frequent exacerbations and remissions. Although direct antiglobulin test is almost invariably positive, most often showing IgG on the erythrocyte surface, T-cell abnormalities have also been a consistent finding. Management remains difficult. Corticosteroids are still the mainstay of first-line treatment, but relapses during tapering are common. As for second-line therapy, there are many options but few systematic studies to support their use. Rituximab therapy has previously been described in at least 19 cases.

Ten patients were reported as single case observations, four were described within a mixed retrospective series of autoimmune cytopenias, and a prospective series of childhood AIHA included five patients with Evans’ syndrome. Thus, all previously published reports should be regarded as case observations that do not allow calculation of response rates. Nevertheless, remission after infusions of rituximab was achieved in many of these cases. The brief report in this issue of Haematologica by Bader-Meunier and co-authors, describing a French retrospective multicenter series of rituximab therapy in 17 children with refractory Evans’ syndrome is, therefore, of considerable interest. The retrospective design may have posed some methodological problems. Furthermore, it was probably not easy to define adequate response criteria, since response definitions for AIHA as well as ITP had to be taken into consideration and might be a matter of discussion. Anyway, this is a large study of therapy in such a rare disease. CR or PR of at least one cytopenia was achieved in 13 patients (76%), with no obvious difference in responses between the AIHA and ITP components. Among ten patients who presented with AIHA and ITP simultaneously, remission of both cytopenias was observed in six. As the authors conclude, rituximab therapy should be considered at an early stage in children with steroid-refractory Evans’ syndrome. Further prospective trials should be performed at a multi-national level, if possible.

**Other autoimmune cytopenias**

All original publications on rituximab therapy for autoimmune neutropenia (AIN) and pure red cell aplasia (PRCA) are case reports or descriptions of small numbers of patients within mixed series of autoimmune cytopenias. Success has been reported in single cases of PRCA complicating CLL, AIN in immunodeficient children, and AIN combined with ITP. Results of rituximab therapy have been disappointing, however, in several other cases of AIN and PRCA. A more important role of T-cell abnormalities as compared to humoral autoimmunity has been proposed as an explanation, although such statements are uncertain in these heterogeneous and rare conditions.

**Tolerability and dosage**

The favorable safety profile of rituximab therapy has been clearly documented in lymphoma patients, despite the well-known, rather frequent infusion-related adverse effects and the less frequent delayed toxicity. The amount of B-lymphocytes killed after administration of rituximab is less in autoimmune dis-
orders than in lymphoma. In our experience, rituximab toxicity is a small problem in patients with CAD. Conflicting results have been published regarding the frequency and severity of adverse events in polyclonal autoimmune cytopenias, possibly reflecting more frequent side effects in children. Generally, however, literature and experience indicate that adverse effects are tolerable, reversible and probably less frequent than in lymphoma patients. Rapid (90-minute) infusion of rituximab has been shown to be a safe option in corticosteroid-pretreated lymphoma patients receiving their second or later infusion, and trials exploring the safety of this regimen may be justified in patients with autoimmune cytopenias as well. Since response duration may be relatively short and re-treatment with rituximab may be effective even several times, severe B-cell immunodeficiency might be a possible concern. In follicular lymphoma, however, prolonged rituximab therapy has not resulted in a significantly increased incidence of adverse events. Furthermore, although peripheral B-lymphocytes are virtually absent after weekly administration of four doses, naïve and memory B cells can usually be detected again after 4-6 months. In my experience with CAD patients, even repeated treatment with rituximab has been well tolerated.

Dose-finding trials of rituximab therapy have hardly been performed in autoimmune cytopenias, and the dosage as well as treatment schedule used in follicular lymphoma has been adopted almost uniformly. In this issue of the journal, the question of dosage is addressed by Provan and colleagues. They analyzed retrospectively the data of 11 consecutive patients with autoimmune cytopenias who received a fixed dose of 100 mg rituximab weekly for 4 weeks. All patients had failed to respond to conventional therapies. The authors observed sustained CR in four of seven patients with ITP and in one patient with pancytopenia, and concluded that low dose rituximab offers a further treatment option in autoimmune cytopenias. Larger, confirmatory studies are required. If the results can be confirmed, rituximab therapy for autoimmune cytopenias will become substantially cheaper and may be associated with even less toxicity than today.

Conclusions
The literature on rituximab therapy for autoimmune cytopenias is growing rapidly. Nevertheless, the low prevalence and incidence rate of most of these diseases makes it difficult to perform large, prospective studies that can underpin evidence-based guidelines. The most convincing documentation has been provided in CAD and ITP, and a reasonable amount of evidence has been provided in warm-antibody AIHA. In CAD, rituximab may currently be recommended as first-line treatment in patients who require medical therapy, but further trials are warranted in order to improve on response rate and duration. In ITP and warm-antibody AIHA, further studies are needed in order to put forward recommendations on which patients should receive rituximab therapy. The recent data on rituximab for the treatment of Evans’ syndrome are promising. Preliminary results seem less encouraging in AIN and PRCA. In these two conditions, more established therapy modalities should be preferred in clinical practice until more data have been provided.

References


