

Controversies in IgG replacement therapy in patients with antibody deficiency diseases

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This Current perspectives article will review and highlight the importance of accurate diagnosis of patients who have failed to produce specific antibodies to naturally encountered foreign proteins or polysaccharides or after vaccination and the appropriate institution of immunoglobulin replacement therapy. The field of primary immunodeficiency disease (PID) has expanded remarkably since the early descriptions 6 decades ago. With greater recognition and advanced cellular and molecular diagnostic technology, new entities and single-gene defects in patients with PID are rapidly being defined. This, combined with treatment advances and newborn screening for severe combined immunodeficiency, has resulted in improved outcomes and survival and even permanent cures. Awareness of PID has also increased, but the guidelines for recognition remain to be validated. The zeal for registering and enrolling patients has potentially created a large body of “patients” treated with immunoglobulin replacement unnecessarily. The complexity, diversity, and availability of laboratory testing have brought awareness of PID to the forefront, but because of an absence of standardization of certain assays, concerns about the correct diagnosis and appropriate treatment have increased. We hope to refocus the discussion on identifying clear laboratory and clinical guidelines for the establishment of an accurate diagnosis of antibody deficiency, its rationale, and, where indicated, institution of safe treatment. (*J Allergy Clin Immunol* 2013;131:1001-5.)

Key words: Antibody deficiency, immunoglobulin replacement, common variable immunodeficiency, primary immunodeficiency, pneumococcal antibody concentration, specific antibody deficiency

Major advances in the definition, delineation, diagnosis, and definitive treatment of primary immunodeficiency disease (PID) have provided the clinician with the ability to identify holes in the armor of host defense and even restore what nature did not provide.¹ Despite these satisfying developments, which include identification of defective genes in patients with PID^{2,3}

Abbreviations used

CVID: Common variable immunodeficiency

PID: Primary immunodeficiency disease

and, in some cases, curative cellular therapy with hematopoietic stem cell reconstitution⁴⁻⁶ or gene therapy,^{7,8} there still remains a murky area of diagnosis and treatment in patients with the most common forms of PID: antibody deficiency disorders. These areas of concern center around the laboratory methods and interpretation of antibody test results,⁹⁻¹¹ which often leads to inappropriate placement of individuals on lifelong immunoglobulin replacement therapy. Here we do not address those antibody deficiency diseases with established molecular and genetic lesions causing absent or nonfunctional cellular factors. Our concern is with the more common situation of patients given the presumptive diagnosis of antibody deficiency based on imperfect laboratory tests and nonvalidated interpretation of the results of such assays.

Table 1 lists antibody deficiencies or combined cellular and antibody diagnoses, including proposed justifications for the use of immunoglobulin replacement therapy (A, established; F, not indicated). It is a very general approach that reflects, to a large degree, the opinion of the authors and is not meant to encompass all clinical situations. In several of the cellular and antibody deficiencies, immunoglobulin replacement has a limited effect because hematopoietic stem cell transplants are needed to restore T-cell numbers and function. We include this table to give readers a sense of the scope of immunoglobulin replacement in patients with PID and suggest where it might be considered effective therapy and where it is not.

A majority of patients with proved PID have some impairment of humoral or antibody-mediated immunity.^{12,13} There are currently a number of laboratory-based methods available for defining the extent of the antibody deficiency. With the advances in knowledge and technology, recognition of single-gene disorders has increased.^{3,14} Flow cytometry-based analyses of lymphocyte subpopulations, including B-cell phenotyping, have also become routine, together with quantitation of serum immunoglobulin levels. With a focus on the PIDs that constitute predominantly antibody deficiencies, combining the 3 approaches of serum immunoglobulin quantitation, evaluation of circulating B-cell numbers and phenotypes, and molecular analyses has revealed important defects leading to arrests of B-cell differentiation at the pre-B-cell or B-cell stage.¹⁵⁻¹⁷ These defects generally result in severe reductions in levels of all serum immunoglobulin isotypes with profoundly decreased or absent numbers of circulating B cells, severe reduction in at least 2 serum immunoglobulin isotypes with normal or low numbers of B cells, low serum IgG and IgA levels with normal to increased IgM levels and normal numbers of total circulating B cells but decreased numbers of memory and/or switched B cells, or rare light chain deficiencies with normal numbers of circulating B cells. Because these conditions are

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TABLE I. Indication of immunoglobulin replacement for antibody deficiency

Score	PIDD entity	Immune defect	Expected response to IVIG/SCIG
A1	Agammaglobulinemia (X-linked, AR) HIgM caused by AID and UNG deficiency	Lack of B cells Abnormal B-cell signaling resulting in defective CSR and SHM	Effective Effective
A2	HIgM caused by CD40L and CD40 deficiency	Defective T/B-cell interaction resulting in abnormal CSR and SHM; defective macrophage activation	Effective, susceptibility to opportunistic infections not reduced
A3	CVID with normal T-cell function (including deficiencies of CD19, CD20, CD21, CD80, ICOS, TACI, or BAFF-R)	Hypogammaglobulinemia, antibody deficiency, often with CSR defect	Effective
B1	CVID with complications (splenomegaly, granuloma formation, autoimmunity, lymphoma)	Hypogammaglobulinemia, antibody deficiency, CSR and SHM affected, often associated with a T-cell defect (abnormal CD40L expression, decreased CD4/CD8 ratio)	Effective in reducing infections but not granuloma formation, autoimmunity or incidence of malignancy
B2	Thymoma with immune deficiency (Good syndrome)	B- and T-cell defects	Effective in reducing infections
B3	XLP with EBV-induced loss of B cells	Antibody deficiency caused by reduced number of B cells; defective cytotoxic T cells, NK cells	Effective in reducing infections, no effect on EBV-related pathology
B4	SCID after HSCT without B-cell engraftment	Mixed chimera with donor T cells and host B cells	Effective
C1	Selective antibody deficiency	Defective CSR reported; anti-PPS antibodies measured by ELISA do not reflect functionality	Antibiotic prophylaxis might be equally effective
C2	Clinically and genetically well-described syndromes with variable antibody deficiency (WAS, DiGeorge syndrome, STAT3 deficiency, VODI, DKC, ICF, AT, Netherton syndrome)	Abnormal antibody responses associated with other immune defects; characteristic syndromic defects might predominate	Partially effective; other disease-specific strategies required
D1	CID (eg, mutations in <i>PNP</i> , <i>ZAP70</i> , and genes controlling MHC class I and II expression)	Hypogammaglobulinemia; B- and T-cell defects	Limited benefit; HSCT should be considered
D2	Hypomorphic mutations in <i>RAG1/2</i> , <i>IL2RG</i> , <i>ADA</i> , <i>RMRP</i> , <i>Artemis</i> , and <i>DNA ligase IV</i>	Hypogammaglobulinemia, combined immune deficiency, often normal (but oligoclonal) T-cell numbers, low B-cell numbers (Omenn syndrome)	Limited benefit; HSCT indicated
D3	SCID	Severe B- and T-cell deficiency, lymphopenia	Limited temporary benefit while waiting for and during HSCT
E1	Complement deficiencies (C3, C4, and C5-9), properdin deficiency	Abnormal antibody responses have been described	Might be beneficial; other prophylactic strategies include hyperimmunization, prophylactic antibiotics
E2	Transient hypogammaglobulinemia of infancy with severe recurrent infections	Hypogammaglobulinemia, generally normal antibody production	Immunoglobulin replacement not indicated except if antibody production is demonstrated to be temporarily defective
E3	IgG subclass deficiency	≥1 IgG subclass affected	Immunoglobulin replacement only if a significant antibody deficiency is demonstrated
F	Asymptomatic hypogammaglobulinemia and normal antibody responses; selective immunoglobulin deficiencies	Normal B- and T-cell numbers, normal antibody responses; selective IgM, IgA, and IgE deficiency	Immunoglobulin replacement not indicated

IVIG/SCIG is effective in entities scored A and in most that are scored B. Those entities scored C and D might obtain limited benefit, and those scored E and F are not expected to benefit from immunoglobulin replacement.

ADA, Adenosine deaminase; *AID*, activation-induced cytokine deaminase; *AR*, autosomal recessive; *AT*, ataxia telangiectasia; *BAFF-R*, B cell-activating factor of the TNF family receptor; *CD40L*, CD40 ligand; *CID*, combined immunodeficiency; *CSR*, class-switch recombination; *DKC*, dyskeratosis congenita; *HIgM*, hyper-IgM syndrome; *HSCT*, hematopoietic stem cell transplantation; *ICF*, immunodeficiency with centromeric instability and facial anomalies; *ICOS*, inducible costimulator; *IL2RG*, IL-2 receptor γ ; *IVIG*, intravenous immunoglobulin; *NK*, natural killer; *PNP*, purine nucleoside phosphorylase; *PPS*, pneumococcal polysaccharides; *RAG*, recombination-activating gene; *RMRP*, RnaseMRP RNA; *SCID*, severe combined immunodeficiency; *SCIG*, subcutaneous immunoglobulin; *SHM*, somatic hypermutation; *STAT3*, signal transducer and activator of transcription 3; *TACI*, transmembrane activator and calcium modulator and cyclophilin ligand; *UNG*, uracil-N-glycosylase; *VODI*, hepatic veno-occlusive disease with immunodeficiency; *WAS*, Wiskott-Aldrich syndrome; *XLP*, X-linked lymphoproliferative syndrome; *ZAP70*, ζ chain-associated protein of 70 kDa.

associated with a global failure of normal antibody production, immunoglobulin replacement is generally indicated.

The present approach used to determine whether subjects with suspected antibody deficiency require immunoglobulin replacement therapy relies heavily on the performance of antibody tests to both

protein and polysaccharide vaccines.¹⁸ A common belief is that these antibody responses to polysaccharide antigens are more meaningful than those to protein antigens, and the use of a 23-valent pneumococcal polysaccharide vaccine has been considered by some as the most informative. There are several problems with the

interpretation of current laboratory measurements of antibody levels to the 23-valent pneumococcal vaccine: What constitutes a satisfactory increase in these antibody levels (is it a 2- or a 4-fold increase)? What defines a protective antibody concentration (currently 1.3 $\mu\text{g/mL}$)? The response to how many serotypes is considered normal? Finally, there is the lack of a functional assay. Importantly, these failures of response often are associated with a clinical picture that generally includes borderline immunoglobulin levels, a history of poorly documented pneumonias in which offending organisms are not defined, and a preponderance of chronic rhinosinusitis (often in atopic patients), and chronic fatigue is a common complaint. Because the decision to treat or not to treat such patients long-term with immunoglobulin replacement rests on nonfunctional laboratory assessments, subjects with normal B-cell immunity are often being treated unnecessarily. Moreover, these decisions to treat have been coupled with a trend toward use of larger and as yet nonvalidated doses of IgG (up to 800 mg/kg/mo) for immunoglobulin replacement, as reviewed in a meta-analysis of 12 open-label prospective clinical trials and 1 open-label retrospective clinical trial that included 482 patients with various forms of reported antibody deficiency.¹⁹ Doses of up to 1.2 g/kg/mo have been given to patients with B-cell deficiency in a similar study of 151 patients.²⁰ Without the benefit of carefully controlled clinical trials with higher doses, this might result in significant and inappropriate expenditures.

An important area of controversy is the use of immunoglobulin replacement therapy for subjects with IgG subclass deficiency with or without associated IgA deficiency.²¹⁻²³ Initial studies linked IgG₂ deficiency to impaired responses to polysaccharide antigens,²⁴ but this entity has been challenged at several levels.²⁵ First, and perhaps most importantly, genetic deficiencies of any of the IgG subclasses were not associated with susceptibility to infection.²⁶ Second, normal laboratory values are not well defined, especially in children, and therefore absolute values of a particular subclass are less meaningful than looking at percentage distribution. The latter provides a "check" on the laboratory by determining whether the sum of the 4 subclasses is similar to the "total" serum IgG value. If the difference is greater than 10%, it may be assumed there is an error. As a general rule, the subject IgG subclass distribution has been reported to be an IgG₁ level of greater than 60%, an IgG₂ level of greater than 10% to 15%, and an IgG₃ level of greater than 5%; IgG₄ might be absent in a significant number of subjects. Using the percentage range for assessing IgG subclass is preferable to judging quantitative immunoglobulin subclasses as abnormal in subjects with borderline total serum IgG. The third concern is that certain drugs, such as corticosteroids, lead to hypercatabolism of IgG, with perhaps the largest effect on IgG₂ levels. Immunoglobulin replacement is not recommended in this latter group because antibody responses are usually normal.²⁷

Perhaps the largest controversy revolves around the diagnosis of common variable immunodeficiency (CVID).²⁸⁻³² In a small percentage of cases, the diagnosis of CVID can be linked to a single-gene defect and thus should no longer be classified as CVID but rather as a specific form of antibody deficiency, such as mutations in inducible costimulator (*ICOS*), transmembrane activator and calcium modulator and cyclophilin ligand (*TACI*), B cell-activating factor of the TNF family receptor (*BAFFR*), *CD19*, *CD20*, and *CD81* and immunoglobulin light and heavy chain deficiencies. The CVID phenotype is heterogeneous, complex, often associated with hepatosplenomegaly, granulomatous interstitial lung disease, autoimmunity, and increased incidence

of cancer and lymphoma.³³ To qualify as having CVID, patients have to present with hypogammaglobulinemia (significant reduction in ≥ 2 isotypes of serum immunoglobulin (less than 50% lower limit of normal) and not simply borderline values) and defective antibody production. In addition, flow cytometry analysis in CVID should show abnormalities in B cells, such as alterations in memory B cells or isotype switched B cells. Abnormal flow cytometry data are particularly important to confirm a questionable diagnosis.³⁴ The problem with the diagnosis of CVID lies not with the patients who exhibit these features (true CVID) but with those presenting with the more common presentation of "recurrent infections" that are often poorly defined, fatigue, and borderline IgG levels. The latter is often seen in older patients, although rare CVID with typical clinical features can occur in young children as well. In the absence of a genetic diagnosis and without the sentinel clinical features of CVID, overinterpretation of B-cell phenotyping and the findings of borderline IgG levels can prompt the diagnosis of CVID and initiation of immunoglobulin replacement therapy. As described above, the diagnosis has often been supported by the overinterpretation of responses to vaccination, particularly responses to pneumococcal polysaccharide vaccination, although responses to other protein antigens might be normal.³²

In fact, if there is an aspect directly related to the diagnosis and treatment of PID with predominantly antibody deficiency that requires the most in-depth re-evaluation, it is the laboratory assessment of the response to pneumococcal polysaccharide vaccination. We suggest that a significant number of subjects in the United States and elsewhere have been started on immunoglobulin replacement therapy erroneously based on the absence of a robust response to pneumococcal polysaccharide vaccine. The most important aspect related to treatment of antibody deficiency is to establish diagnostic criteria. One would think that after so many decades of investigating antibody responses, the criteria for the diagnosis of antibody deficiency would be more clearly defined and experts would have established a consensus opinion. Unfortunately, that is not the case, with the result that too many healthy subjects are being administered an expensive treatment for which there is no documented need. Without a consensus opinion,³⁵⁻⁴¹ physicians are confronted by patients with perceived recurrent infections, many of them troublesome but not serious, to provide them with immunoglobulin replacement therapy. Erroneously, this "passive" immunization with immunoglobulin at regular intervals might be credited as a miraculous recovery, a reminder to physicians that the placebo effect is hard to argue against and the result is difficulty in stopping unnecessary treatment.

To justify immunoglobulin replacement therapy, physicians in fellowship training and physicians participating in continuing medical education lectures are taught to practice evidence-based medicine and to use the clinical immunology laboratory for support of their diagnosis. Thus the presumed surrogate markers of antibody deficiency should be known and can be readily obtained. Tests that can be ordered to confirm a suspected diagnosis of antibody deficiency include measuring antibody titers to common protein and polysaccharide antigens. Protein antigens include those present in diphtheria, tetanus, pertussis, and conjugated pneumococcal or *Haemophilus influenzae* vaccines. Although the antibody responses to these proteins are useful,¹⁸ as they certainly are in cases of panhypogammaglobulinemia, it is the antibody response to the 23-valent pneumococcal polysaccharide vaccine that currently is considered by many physicians to be the most definitive

test of B-cell function.^{42,43} Most clinicians attempt to assess adequate B-cell function by measuring the production of antibodies to the 23-valent pneumococcal polysaccharide vaccine, comparing preimmunization and postimmunization antibody titers. However, this assay, as described above, is fraught with many problems, notwithstanding the many variations in laboratory-to-laboratory assays, standards, and reliability. Every clinical investigator struggles with these criteria because of their imprecise nature.

Clinical investigators must take a step back from the dilemma of whether to initiate immunoglobulin replacement therapy by questioning the very nature and importance of anti-pneumococcal antibody responses not in isolation but in the context of a clinical picture with evidence for documented pneumococcal disease. It must be remembered that antibody values have to reflect functional antibody responses rather than what is currently measured (ie, antibody protein concentration) to be meaningful.^{9,10} There should be a validation assay that measures killing of organisms by vaccine-elicited anti-pneumococcal antibodies and not simply an assessment of the quantity of antibody measured above baseline levels. Use of the opsonophagocytic assay would provide clinicians with more valuable information on which a diagnosis of true antibody deficiency could be established.⁴⁴⁻⁴⁷ This more stringent requirement of functional antibody assessment would be even more important for the popular diagnoses of "selective antibody deficiency," IgG subclass deficiency, and CVID in patients with serum IgG concentrations that are marginally less than the 95% CIs.

Another aspect of treatment of immunodeficient patients with immunoglobulin replacement therapy concerns the amount of IgG that should be provided.⁴⁸ Over the decades that saw the explosion of monogenic defects resulting in antibody deficiency, there has been a growing perception that when it comes to the amount of IgG administered, "more is better." For example, clinicians were introduced to the use of immunoglobulin replacement therapy with the recommendation of providing 200 to 400 mg/kg every 3 to 4 weeks; subsequently, the recommendation was increased to 400 to 600 mg/kg, and now the dose has crept up close to 800 mg/kg or greater every month.^{19,20,49-52} Some have advocated a "biologic" level that is the dose that keeps the patient infection free.⁵³ These dose escalations require serious scrutiny and clinical proof before becoming routine. As reported, antibody levels with immunoglobulin replacement therapy might not correlate well with serum IgG levels.⁵⁴ Furthermore, different subjects catabolize administered immunoglobulin at different rates,⁵⁵ and therefore dose titration to keep a subject infection free can vary. By and large, US and European immunologists treat immunodeficient patients with approximately the same amount and frequency of immunoglobulin replacement, although some significant differences exist between the 2 groups.⁵⁶ The high doses of immunoglobulin replacement therapy now being prescribed for antibody-deficient patients is approaching immunomodulatory doses of IgG, thus possibly modifying immune responses that might prove harmful to the patient.⁵⁷

Assessment of humoral immunity is an essential component in any subject suspected of having a PID. However, equally important, interpretation of antibody response testing must rely on clinical correlations. The use of so-called "warning" signs has had little, if any, verification.⁵⁸ A diagnosis of PID is linked to susceptibility to infectious diseases and, in the case of antibody deficiency, to a specific set of bacterial microorganisms. This is clearly seen in patients with X-linked agammaglobulinemia in which susceptibility to encapsulated organisms, such as *Streptococcus pneumoniae*, is intertwined with the failure to make

specific antibodies. Because the diagnosis and treatment of CVID and, more recently, specific antibody deficiency has almost solely relied on the response to pneumococcal polysaccharide vaccination by many practitioners, this assumption must be questioned and more thoroughly investigated by careful interpretation of laboratory test results (hopefully to include functional antibody testing in the future), as well as the clinical context and history from the patients.

In principle, the consideration of an antibody deficiency is triggered by a patient presenting with the main complaint of repeated infections. The first objective findings are often questionable serum immunoglobulin levels or a low serum IgG level. The next step should be a careful evaluation of the clinical presentation, particularly documentation of the types and severity of infections; evidence for autoimmunity, lymphadenopathy, or splenomegaly; and findings of significant chronic lung or gastrointestinal disease. If warranted, determination of antibody responses to administered protein and polysaccharide antigens is indicated but should be coupled with rigorous evaluation of the responses in context with the clinical picture. The basis for the diagnosis of antibody deficiency beyond the obvious and well-documented disorders has largely evolved to an interpretation of antibody titers in the blood of previously vaccinated subjects, particularly with antibodies elicited by the unconjugated 23-valent pneumococcal polysaccharide vaccine. As currently measured, these anti-pneumococcal antibodies do not represent critical functional antibodies and, in the present state of interpretation, do not have a coherent and consensus opinion or clinical correlation. Because many subjects are being started on immunoglobulin replacement therapy (intravenous or subcutaneous) on uncertain grounds, there is a critical need for (1) better laboratory assessment of functional antibody responses, (2) development of a consensus as to what constitutes a normal antibody response, and (3) better clinical and microbiological evaluation of the types, extent, and severity of infections with which the patients present relative to those seen in true antibody-deficient patients. This is especially important when a diagnosis of CVID, subclass deficiency, or selective antibody deficiency is considered. Finally, controlled clinical trials free of commercial bias should be performed to better define the amount of immunoglobulin replacement that is sufficient or optimal for the prevention of infections in those defined properly as antibody deficient.

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