

Assessment of protein antibody response in patients with suspected immune deficiency

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INTRODUCTION

“Practical Aspects of Ambulatory Diagnosis and Management of Immunodeficiency Disorders” is intended to help allergists evaluate and manage patients with suspected immune deficiencies.¹⁻³ This article reviews the use of protein antibody response in the evaluation of immune competence.

Primary disorders of humoral immunity may be due to intrinsic defects in B cells or lack of signals necessary for B cell maturation and function. The latter may occur in T-cell deficiency or when T-cell signaling is impaired, as in X-linked hyper IgM syndrome. Antibody deficiency increases susceptibility to bacterial and viral infections and allows recurrences of infections normally experienced only once.^{4,5} Reduced immunoglobulin levels raise concerns for antibody deficiency; however, normal or elevated immunoglobulin levels can also conceal antibody deficiencies (eg, selective antibody deficiency, human immunodeficiency virus, multiple myeloma, and IgG subclass deficiency). In addition, some medications may reduce immunoglobulin levels without causing immune deficiency. Therefore, evaluation of such patients must include evaluation of responsiveness to protein and polysaccharide antigens.⁵⁻⁷

PROTEIN-INDUCED IMMUNE RESPONSE

Unlike polysaccharides, proteins can recruit T-helper cells and initiate a robust immune response. Thus, proteins are usually thymus-dependent antigens and are potent immunogens. Initial exposure to a protein antigen induces a predominantly IgM response. Subsequent exposures induce robust memory cell production with earlier, greater, and longer-lasting IgG. Primary and secondary responses are absent in patients with few B cells, such as those with Bruton agammaglobulinemia, and those with severe combined immune deficiency who have undergone stem cell transplantation but have poor B-cell engraftment. The secondary response is

impaired in hyper IgM syndromes (inability to class switch^{8,9}) and C4 deficiency (repeated primary responses without maturation to a secondary response).

ROLE OF PROTEIN ANTIBODY RESPONSE IN EVALUATION OF IMMUNE COMPETENCE

Identification of the underlying immune defect and determination of pathogens causing the patient's symptoms are the main goals of an immune workup. These can be achieved by measuring specific antibody titers against infections that have been documented in a particular patient. However, these are often not available, and a clinician will usually obtain antibody titers against vaccinations rather than documented infections.

WHICH ANTIBODY TITERS TO CHOOSE

High purity, potent immunogenicity, and nearly universal immunization make tetanus and diphtheria the ideal candidates for evaluation of protein antibody response. Measles-mumps-rubella, hepatitis B, pertussis, and polio offer additional choices. In general, the more different antigens are evaluated, the more valid the conclusions about the patient's ability to respond will be. Furthermore, in childbearing age females or overseas travelers, normal results to tetanus and diphtheria may suggest the absence of an immune defect, but the lack of antibodies against particular antigens (rubella or hepatitis A, respectively) may warrant reimmunization and evaluation of specific responses. To help clinicians choose age-appropriate antibody tests, the ages of first exposure to different protein vaccine antigens in the United States are summarized in Table 1.¹⁰

TESTING AND REPORTING OF PROTEIN ANTIBODY TITERS

Antibodies may be quantified by different methods. Enzyme-linked immunosorbent assays are used most commonly. A plastic bead or plate is coated with an antigen. Antibody in the patient's serum binds to the immobilized antigen and is detected by enzyme-conjugated antihuman IgG or IgM. A chromogenic substrate is added, and the amount of antibody present is proportional to enzyme activity detected. Neutralization assays measure inhibition of cytopathic effects of standard inocula of viruses in tissue cultures by antibodies in serum. Hemagglutination-inhibition assays are used for mea-

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Table 1. Ages of First Exposure to Protein Vaccines in the United States^a

Age of first exposure	Vaccine	Active antigen
<6 mo ^b	Hepatitis B	Recombinant viral antigen
	DTaP	Toxoids and inactivated bacterial components
	Rotavirus	Live attenuated virus
	IPV	Inactivated virus
6–12 mo ^b	Influenza	Inactivated viral components ^c
12–24 mo ^b	MMR	Live attenuated virus
	Varicella	Live attenuated virus
11–12 y ^b	Hepatitis A	Inactivated viral antigen
	HPV	Recombinant viral antigen

Abbreviations: DTaP, diphtheria and tetanus toxoids and acellular pertussis; HPV, human papilloma virus; IPV, inactivated poliovirus vaccine; MMR, measles-mumps-rubella vaccine.

^a Antibody titers should be obtained 4 to 6 weeks after receiving the vaccine. IgM antibody tests are commercially available for hepatitis A, pertussis, influenza, MMR, and varicella.

^b Age when the first dose of vaccine is recommended.

^c Intranasal influenza vaccine is a live attenuated viral vaccine and is not recommended for those younger than 5 years.

sles, and nephelometry is used for purified antigens. Of these, only enzyme-linked immunosorbent assays can distinguish between IgG and IgM.

Enzyme-linked immunosorbent assay and nephelometry results are reported in micrograms or international units per milliliter. Neutralization and hemagglutination-inhibition assays are expressed as dilutions of serum or “titers.” The “reference range” indicates protective, equivocal, or nonprotective antibody levels determined by comparison to standard serum samples known to protect animals, or to the mean or minimal titers in recipients of clinically successful immunizations.

INTERPRETATION OF PROTEIN ANTIBODY TITERS

Age-specific ranges for protein antibody titers may not be available, and patients may have missed recommended immunizations. Thus, nonprotective levels do not necessarily indicate poor immune responses. Patients with nonprotective antibody titers should be reimmunized with the respective vaccines, and postimmunization titers should be drawn 4 to 6 weeks later. A 4-fold or greater increase in the titers confirms specific responsiveness, provided the postimmunization titers are protective.^{5–7} For example, 0.1 IU/mL or greater is usually considered protective for tetanus. A patient with preimmunization and postimmunization titers of 0.01 and 0.04 IU/mL, respectively, despite the 4-fold increase, is not protected. Preimmunization and postimmunization titers of 0.08 and 0.40 IU/mL indicate a normal response. However, patients with high preimmunization titers may not show a 4-fold increase but are still considered immunocompetent.

VARIABLES IN INTERPRETATION OF PROTEIN ANTIBODY RESPONSES

The following 4 variables are most commonly encountered in everyday practice: vaccine efficacy, infants, aging, and patients receiving plasma products.

Vaccine Efficacy

The efficacy of vaccines varies. Seroconversion rates are available from the *Red Book*¹¹ and from prescribing information given with the vaccines. Protective titers can persist for 35 and 15 years with tetanus and hepatitis B, respectively, whereas pertussis titers may wane in 3 to 5 years.

Infants

IgG can be transferred transplacentally, so antibody titers may be protective despite an infant’s inability to produce them. However, the presence of specific IgM (not transplacentally transported) in infants suggests immunocompetence.

Aging

Several studies^{12–14} suggest that protein antibody titers from childhood immunization decline with age in healthy adults older than 50 years. Thus, measuring the response to reimmunization is more useful for assessing immunocompetence than measuring antibody titers at a single point in time only.

Patients Receiving Plasma Products

Recipients of IgG, plasma, or platelet infusions can acquire antibodies passively and may have protective titers despite an inability to produce them. We recommend that the infusions be discontinued 3 months before evaluation of antibody responses.

If discontinuation is not possible, immunization with neoantigens may be considered. Because the general population is not exposed to neoantigens, such as keyhole limpet hemocyanin or bacteriophage ϕ X174, plasma products lack antibodies against them. Usually, 2 doses are given 6 weeks apart and titers are measured 4 weeks later.^{15,16} Although these antigens are commercially available, special arrangements (with institutional review board approval) must be made for their administration and titers are only measured in a few research laboratories.

SECONDARY CAUSES OF REDUCED PROTEIN ANTIBODY RESPONSES

Malnutrition, vitamin deficiencies (A and B₁₂), corticosteroids, gold, and chemotherapy may cause variable antibody deficiencies.^{17–19}

Protein-losing conditions (nephrotic syndrome and enteropathies) can reduce immunoglobulin levels without affecting antibody responses. Uremia can cause functional antibody deficiency.

Human immunodeficiency virus infection can cause polyclonal B-cell activation and impair production of specific antibodies. In most adults, B-cell memory antedates their acquisition of human immunodeficiency virus, but infants may be helped by intravenous immunoglobulin if highly active antiretroviral therapy is not available. Most patients receiving highly active antiretroviral therapy for 1 year respond to immunization with tetanus, hepatitis A, and keyhole limpet hemocyanin.¹⁶

Rituximab, an anti-CD20 monoclonal antibody frequently used in B-cell malignancies and autoimmune diseases, may impair antibody responses to new antigens, but memory responses are usually preserved.²⁰

INITIATION OF γ -GLOBULIN INFUSIONS

The decision to initiate IgG replacement involves many factors. In addition to IgG levels and poor responses to vaccines, other considerations include severity and frequency of infections, effect on the patient's general health, job or school attendance, quality of life, and risks of IgG replacement. Some patients with partial antibody deficiencies may be maintained with prophylactic antibiotics with close monitoring.

CONCLUSIONS

Protein antigens are potent immunogens that recruit T-cell help. Measuring antibodies to protein antigens helps determine immunocompetence. In patients with nonprotective titers, reimmunization and measurement of postimmunization titers provide a more complete evaluation than measurements made at only one point in time. Measuring specific antibodies to multiple proteins facilitates accurate evaluation of immunocompetence.

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