IMMUNE COMPLEX DISEASES*

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The essential feature of immune complex diseases is that they are caused by antigen–antibody complexes formed in the circulation or in interstitial fluids. Complexes so formed are for the most part taken up and catabolized to harmless end products by reticuloendothelial phagocytes and blood leukocytes, but a small fraction eludes these cells and tends to accumulate in filtering structures throughout the body such as glomeruli and blood vessels where they can cause injury. This accumulation is apparently determined by anatomic and physiologic, not immunologic, factors so that the sites of injury or disease need have no immunologic relationship to the causative antigen or antibody. In this respect, immune complex disease is quite different from the "antitissue antibody" type immunologic disease in which the subject makes antibody specifically reactive with an antigenic component of the target or diseased tissue and the distribution of the tissue-fixed antigen determines the location of the disease. Further, immune complex diseases may involve multiple sites or tissues depending upon physical and biologic characteristics of the complexes and local permeability factors in the filtering structures.

The pathogenicity of immune complexes is in large part determined by their antigen–antibody ratio, i.e., size, and the biologic properties of their antibody. Complexes formed in antibody excess tend to be insoluble and are rapidly phagocytosed, do not circulate, and have little opportunity to accumulate in filtering sites. Complexes formed in extreme antigen excess, i.e., antibody-2 antigen, are usually too small to become trapped in physiologic filters and, further, such antigen excess complexes do not contain an arrangement of immunoglobulin molecules capable of complement (C) activation. Also essential in C activation is the nature of the antibody in the complex since some antibodies are capable and others incapable of C activation. While the immunochemical character of the antigen does not seem to be an important factor in phlogogenicity, the size of the antigen may play a role since large protein molecules and even virions themselves acting as antigens would of necessity result in large nonfilterable antigen–antibody complexes regardless of antigen–antibody ratio.

Most of our knowledge of the pathogenic mechanisms in immune complex disease has derived from the study of experimental and clinical serum sickness. These diseases resulting from a single large injection of foreign serum protein are acute phlogogenic, multisystem disorders involving blood vessels, heart, kidneys, lymphoid tissue, skin, and joints, resembling in part acute glomerulonephritis, rheumatic fever, systemic lupus erythematosus, polyarteritis, and rheumatoid arthritis [1]. Through the use of isotope-labelled bovine serum albumin (BSA) as antigen in rabbits, it was possible to quantitate the various immunologic events involved in this disease [2]. As indicated in the Figure the immune response to BSA in the rabbit is indicated by the rapid terminal elimination of the circulating antigen beginning 11 days after injection. The fate of circulating serum protein antigen is characterized by an initial two-day period during which the antigen equilibrates between intravascular and extravascular spaces followed by a slower rate of loss, lasting a little more than a week, during which the antigen is catabolized nonimmunologically, and finally by a rapid terminal immune elimination. Measurement of circulating antigen–antibody complexes indicates their appearance on day 8 shortly before immune elimination begins and their increase during the following 2–3 days. Coincident with the increase of circulating complexes and the beginning of immune elimination is a fall in the serum C to levels half normal. Presumably, as antibody is formed and complexes increase in size and amount, they are capable of reacting with serum C which in turn increases the size of the complexes and makes them more readily phagocytosed, thereby hastening their elimination. The importance of immune complex size is illustrated by the fact that only rabbits with circulating complexes greater than 19s in size developed significant serum sickness [3]. Once the antigen is eliminated, free antibody appears in the circulation. Simultaneously with the appearance of circulating complexes the clinical and histologic manifestations of serum sickness develop. Following elimination of circulating complexes the serum sickness diminishes. If the immune complexes are truly the etiologic agent of this disease, one would expect to find them in the lesions themselves and this has been done utilizing the fluorescent antibody technique [2]. BSA, host immunoglobulin, and host C have all been detected in diseased glomeruli and in sites of arteritis.

These immunologic developments trigger a

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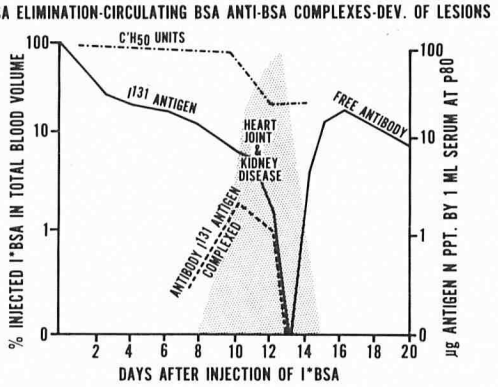

FIGURE. Dynamics of the immune response following injection of I^{131}-labelled BSA in rabbits.

number of secondary humoral and cellular events capable of mediating an inflammatory response. As the complexes form they act upon basophils probably via homocytotropic antibodies and, as a result of this, platelets, the major reservoir of histamine and serotonin in the rabbit, are clumped and otherwise affected to liberate these substances. This systemic liberation of vasoactive substances is essential for the subsequent localization of complexes in various parts of the vascular bed, presumably by increasing vascular permeability, and the localization can be blocked by antihistamines and antiserotonins. As the complexes begin to localize in glomeruli, arteries, etc., they also activate C, and the C-derived chemotactic factors attract polymorphonuclear (PMN) leukocytes which release their proteolytic enzymes and basic proteins causing local tissue destruction. Complexes also cause endothelial proliferation, a prominent aspect of the glomerular response in acute serum sickness. Since depletion of C and PMN in animals subjected to serum sickness will not completely suppress the associated glomerulonephritis, it seems that complexes can also cause damage in the sites in which they localize by C and PMN independent routes, the nature of which is not yet well understood [4].

A chronic form of serum sickness clinically more similar to the spontaneous human serum sickness-like diseases can be produced by daily injections of relatively small amounts of foreign serum proteins into rabbits [5]. Properly executed this procedure results in the presence of circulating complexes for at least several hours every day. Such rabbits develop a chronic membranous or necrotizing glomerulonephritis morphologically distinct from the acute proliferative glomerulonephritis occuring in one-shot serum sickness. The antigen-antibody system and the pathways of mediation of inflammation, etc. appear to be similar in both kinds of serum sickness so the difference in morphologic expression of disease is presumably related to the quantitative and temporal aspects of exposure to immune complexes, a fact worth remembering when trying to relate particular human disease patterns to certain types of antigen-antibody complexes.

Our understanding of the role of immune complexes in serum sickness is reasonably complete because the antigen is known and can be traced via isotopic and immunofluorescent techniques, thereby making possible detailed observation of formation and fate of the complexes. In most human diseases of suspected immune complex pathogenesis, the antigen is not known and the search for immune complexes is far from direct so suspicions are based on indirect and/or circumstantial evidence. The first suggestion of immune complex pathogenesis in human disease was based on the clinical and histopathologic similarities of serum sickness on one hand and glomerulonephritis, rheumatic fever, systemic lupus erythematosus, rheumatoid arthritis, and various vasculidites on the other. This relationship was postulated even before the pathogenesis of serum sickness was well understood. Immunologic and physical attempts to demonstrate circulating immune complexes have provided suggestive evidence of their presence in some patients with rheumatoid arthritis, systemic lupus erythematosus, hyperglobulinemic purpura, and Australia antigen infections. Among the more successful techniques employed in the demonstration of circulating immunoglobulin containing complexes possibly antigen-antibody in nature are: (1) *analytic ultracentrifugation*—a quantitative but relatively insensitive procedure which gives a measurement of size as well as amount of protein complexes, (2) *cryoprecipitation*—an easily performed, often quite sensitive means of separating complexes with temperature-related solubility, (3) *reaction with the C1q component of complement* —a reaction capable of detecting immunoglobulin containing complexes as well as other possible serum constituents such as DNA and endotoxin [6], and (4) *reaction with IgM rheumatoid factor* —selected rheumatoid factors will react with small amounts of immunoglobulin containing complexes, particularly those found in rheumatoid patients [7].

The best understood of the various immune complex diseases is systemic lupus erythematosus [8]. In this disease there are multiple antibodies reactive with a variety of cellular antigens of the host. Best known are the diagnostic antinuclear and particularly anti-DNA antibodies. In the course of lupus some of these cellular antigens are detectable in the serum (antigen excess) and at other times free antibodies to these antigens are present (antibody excess). During shifts from antigen to antibody excess and vice versa, immune complexes would of necessity be present in the serum, a situation very similar to that in serum sickness. These complexes may be evidenced by cryoprecipitates or positive C1q reactions. As would be expected, while immune complexes cir-

culate, the manifestations of disease worsen. These complexes deposit in glomeruli and larger blood vessels associated with the most serious manifestations of this disease, glomerulonephritis and vasculitis. Renal biopsy has been extremely valuable in the study of this kidney disease. Nuclear (DNA) antigens have been found in diseased glomeruli as well as host immunoglobulin and complement. In addition, antinuclear (DNA) antibodies have been eluted from the kidneys of lupus patients. Thus, all of the components of immune complexes have been identified in the diseased organs.

The other disease with a reasonably well demonstrated immune complex pathogenesis is glomerulonephritis. About 95 per cent of glomerulonephritis appears to be associated with immune complex deposition in the glomeruli and 5 per cent is caused by antibodies formed against antigens fixed in the glomerular basement membrane. Immune complexes deposit in a characteristic pattern in glomeruli, irregular granular to lumpy agregates along the glomerular basement membranes. Thus, when host immunoglobulin and complement are found in this distribution, it is strong presumptive evidence for immune complexes even if the nature of the antigen is not known. In immune complex glomerulonephritis a number of different antigens have been found capable of forming nephritogenic complexes. In addition to the nuclear antigens in systemic lupus there are: streptococcal antigens in acute post streptococcal glomerulonephritis, malarial antigens in the nephrosis accompanying quartan malaria, staphylococcal antigens in the nephritis accompanying infected ventriculoatrial shunts for hydrocephalus, Australia antigen in nephritis and vasculitis accompanying infection with this agent, and thyroglobulin in nephritis associated with autoimmune thyroiditis. However, taken together these recognized antigens account for a small proportion of all immune complex glomerulonephritis. The great variety of antigens capable of causing this single disease via a single pathogenetic mechanism illustrates the antigenic nonspecificity of immune complex disease.

In a sense, our study of glomerulonephritis has turned up many more instances of immune complex disease than we have antigens to account for them and has prompted a search for other etiologic agents. Examination of spontaneous immune complex diseases of animals finds many, perhaps most, of them associated with chronic viral infections. In these chronic infections viremia is the rule, and commonly the viruses themselves and perhaps soluble viral products serve as antigens to form circulating immune complexes. This situation has been found in chronic infections with the following viruses: lymphochoriomeningitis, lacticdehydrogenase, Gross, Rauscher, Friend, Aleutian, and the agent of equine infectious anemia. Indeed, in the well-studied NZBxW mice with a lupus-like disease, not only are there nuclear antigens involved in immune complex formation but also antigens derived from the Gross-like virus which infects them. Thus, in these animal models of human lupus at least two kinds of antigen-antibody systems are simulataneously forming immune complexes which are depositing in glomeruli and causing nephritis. The search for similar viruses which may be involved in human immune complex disease has just begun, and one virus, Australia antigen, has been shown to form circulating complexes with immunoglobulin and to deposit in glomeruli associated with the development of nephritis.

From this discussion it is apparent that immune complexes are a common means by which immunologic diseases are mediated. The facts that a single kind of antigen-antibody complex may induce disease in a variety of tissues and organs, as in serum sickness; that a large number of different antigens may be able to form complexes all capable of causing a similar disease, as with the multiple agents involved in immune complex glomerulonephritis; and that multiple antigen-antibody systems may be found participating simultaneously in immune complex formation, as in NZBxW mice and patients with systemic lupus erythematosus, indicate the pathogenetic complexity of these disorders. However, now that some of the immunologic characteristics of these diseases are being recognized, the search for causal agents should yield results and then some rational immunotherapy and perhaps even immunoprophylaxis may be possible.

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