Experimental Models of SLE: Induction in Chimeric States and Pathogenic Role of Immunoglobulin Interactions

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Experimental model of SLE represent an important tool for the study of the pathogenesis of autoimmune disorders. Thus, the importance of the genetic background has been documented in strains of mice spontaneously developing SLE-like diseases such as the NZB/W, MRL/l or BXSB mice. Immunopathological features characteristic of SLE were also found in mice undergoing graft- versus-host disease whereas a possible role for polyclonal B cell activation has been suggested by the observation of pathological changes following injection of B cell mitogens.

1. Generation of pathogenic immune complexes through immunoglobulin interactions

Immune complex (IC) lesions are a major feature of human and murine SLE. Mechanisms leading to the generation of those complexes include autoimmune responses against tissue antigens, e.g. DNA, or against antigens coded by retroviral sequences, e.g. anti-gp70. In some of these models, interactions between immunoglobulin molecules appear to play a major role in the formation of pathogenic complexes. Two types of interactions should be considered: those involving anti-Fc antibodies and those related to anti-idiotypic antibodies.

Recently, high levels of rheumatoid factor (RF) have been found in a murine strain, MRL-lpr/lpr. These mice, in addition, to lupus-like syndrome, spontaneously develop serologic and pathologic abnormalities that closely resemble those seen in patients with rheumatoid arthritis (RA). Their sera contain not only RF but also intermediate-sized IC similar to those found in the sera and synovial fluids of patients with RA. Further, MRL-lpr/lpr mice develop extremely large amounts of cryoglobulins which parallel the production of RF. The availability of such a murine strain provides a unique opportunity to analyze the role of RF for the formation of IC and cryoglobulins and eventually for the development of tissue lesions.

Indeed, although IgM RF could be relatively easily induced in various strains of mice as a result of polyclonal B cell activation or of a secondary immune response against IgG-containing IC, MRL-lpr/lpr mice have also been shown to produce high titers of IgG RF as well as IgM RF. Since only MRL-lpr/lpr mice among the SLE-
prone mice develop arthritic changes resembling RA\(^1\). IgG RF may be related to the development of arthritic lesions.

The presence of intermediate-sized IgG RF-containing IC in sera from MRL-\(lpr/lpr\) mice was first suggested by the presence of a unique interaction between two different individual MRL mouse sera: one serum forms a precipitin line in gels with another serum from a different mouse\(^2\). Intermediate-sized complexes of IgG, sedimenting between the normal 7S and 19S serum component and presumably containing IgG RF, were found to be the reactants causing these precipitin interactions. Such IgG RF IC were isolated from sera from MRL-\(lpr/lpr\) mice by affinity chromatography with agarose-mouse IgG columns. Apparently, IgG RF isolated from MRL-\(lpr/lpr\) mice underwent concentration-dependent self-association similar to those from patients with RA\(^3\). Further, C3H and B6 mice without apparent background for autoimmune diseases were shown to produce high titers of RF when the \(lpr\) gene was transferred from the MRL strain to these strains of mice.

Several strains of mice which spontaneously develop SLE-like syndrome form significant amounts of cryoglobulins and the most elevated concentrations of cryoglobulins have been found in MRL-\(lpr/lpr\) mice\(^4\) and in C3H and B6 mice bearing the \(lpr\) gene. The major components of cryoglobulins from \(lpr\) mice are immunoglobulins and the concentration of IgG in cryoglobulins was found to be about 30 times greater than the concentration of IgM. It was striking that there was a marked enrichment of IgG3 subclass in cryoglobulins as compared to other IgG subclasses. Of interest, we have recently found that 16 monoclonal IgG RF derived from MRL-\(lpr/lpr\) mice were all IgG3. BALB/c mice injected with some hybridomas secreting IgG3 RF developed substantial amounts of IgG3-rich cryoglobulins in ascites and sera. These results suggest that the IgG3 RF represents the major source of cryoglobulins occurring in \(lpr\) mice. In addition, it was seen that normal mice bearing one of the IgG3 RF hybridoma developed extensive pathologic manifestations within a few days, including peripheral vasculitis and glomerulonephritis, reminiscent of those characterizing some patients with acute cryoglobulinemia. The second type of immunoglobulin interactions which can be involved in the pathogenesis of SLE is related to idiotypes. Indeed, interactions between immunoglobulin molecules occurring through complementary sites in hypervariable regions have been observed during natural immune responses and polyclonal B cell activation and have been shown to contribute to the generation of soluble or cryoprecipitable complexes.

There is evidence that auto-anti-idiotypic antibodies may occur during the course of an immune response, with a specificity for the major idiotypic determinants of the antibodies resulting from this immune response. The reaction of anti-idiotypic antibodies with immunoglobulin molecules bearing corresponding idiotypes leads in such situations, to the formation of idiotype-anti-idiotypic immune complexes\(^5\). Some conditions are characterized by repeated antigenic stimulations. It is possible that, in these situations, the frequent reinduction of antigen-specific antibodies and of their corresponding anti-idiotypic antibodies could result in the formation of pathogenic idiotype-anti-idiotype complexes. The occurrence of circulating idiotype-anti-idiotype immune complexes does not depend directly on the presence in the circulation of a given antigen and thus idiotypic interactions leading to the formation of immune complexes may persist after elimination of the antigen inducing the initial immune response.
The possible role of idiotypic interactions in the pathogenesis of IC diseases was particularly studied in experimental situations where pathogenic IC are formed in association with polyclonal B cell activation. Indeed, the non-specific triggering of the B cell repertoire induced by B cell mitogens such as bacterial lipopolysaccharides or naturally occurring in the course of certain parasitic diseases (African trypanosomiasis, malaria) is often associated with the generation of circulating immune complexes and with the development of immune complex-mediated tissue lesions. In these situations, a wide variety of antibodies is produced, including antibodies directed against certain haptens, heterologous proteins, and also certain autoantigens. Thus, we have investigated whether polyclonal B cell activation could trigger the production of auto-anti-idiotypic antibodies and, if so, whether the reaction of these antibodies with immunoglobulins bearing corresponding idiotypic specificities could lead to the formation of idiotype-anti-idiotype immune complexes. To test this hypothesis, polyclonal B cell activation was induced in BALB/c mice by injection of bacterial LPS, and the spleens of these mice were assayed for antibody-producing cells to phosphorylcholine (PC), to TEPC-15 myeloma protein (anti-PC), and to other myeloma proteins. We found that LPS injection results in a triggering of both anti-PC and anti-TEPC-15 antibody producing cells. Anti-PC antibodies could be detected in the circulation and the majority of these antibodies was found to bear the T15 idiotype. The simultaneous production of anti-T15 idiotype antibodies were shown to lead to the formation of idiotype-anti-idiotype immune complexes. It is clear that T15 idiotype immune complexes represent only a fraction of the immune complexes formed in BALB/c mice after LPS injection or during experimental trypanosomiasis. However, polyclonal B cell activation is known to stimulate a wide variety of B cell clones and one may consider that a certain proportion of these clones produced antibodies able to react with idiotypic determinants of immunoglobulins produced by other clones that have been simultaneously triggered.

Immune complexes formed after injection of bacterial LPS in mice are responsible for the development of an immune complex glomerulonephritis. Therefore, BALB/c mice were studied for a possible renal deposition of T15 idiotype-anti-T15 idiotype immune complexes after injection of bacterial LPS. Using immunofluorescence, we first investigated whether rabbit anti-T15 idiotype antibodies could detect T15 idiotype-bearing immunoglobulins within the immune complexes deposited in the renal glomeruli. From day 6 to day 28 after LPS injection, rabbit anti-T15 idiotype antibodies were found to react with molecules deposited in a granular fashion in the glomeruli, suggesting a glomerular deposition of immunoglobulins bearing the T15 idiotype. Further evidence for a glomerular deposition of T15 idiotype-anti-T15 idiotype immune complexes was obtained by the analysis of kidney eluates from mice injected 18 days earlier with LPS. An idiotype specific reaction of the eluates with the TEPC-15 myeloma protein was seen, indicating the presence of anti-T15 idiotype antibodies in the kidney eluates. Therefore, the potential role of idiotypic interactions in the pathogenesis of lupus associated renal lesions should certainly be considered.

2. SLE-like disease resulting from allogeneic interactions after induction of neonatal tolerance to transplantation antigens

Neonatal injection of allogeneic lymphoid cells in mice induces a state of specific
tolerance to the corresponding alloantigens characterized by survival of skin allograft, cytolytic T cells (CTL) unresponsiveness, and lack of mixed lymphocyte reaction\(^\text{12}\). Semi-allogeneic F1 hybrid cells are usually selected for the induction of neonatal tolerance, because their inoculation does not lead to the fatal runt disease observed with fully allogeneic cells. However, some reports indicate that induction of neonatal tolerance by F1 hybrid cells can lead to non-lifethreatening pathologic changes such as splenomegaly and enlargement of lymph nodes\(^\text{13}\). In different situations where allogeneic cells persist in an individual, allogeneic interactions have been found to be associated with autoimmunity and with the formation of pathogenic immune complexes. For instance, features of systemic lupus erythematosus have been observed in experimental graft-versus-host disease (GVHD) induced in F1 hybrid mice by injection of parental cells\(^\text{14}\).

Recently, we have found that the induction of transplantation tolerance by semi-allogeneic F1 hybrid cells in mice was associated with the development of autoimmunity and immunopathology\(^\text{15}\). BALB/c mice neonatally injected with C57BL/6 × BALB/c F1 hybrid spleen cells develop hypergammaglobulinemia, autoantibodies and features of IC disease in close association with transplantation tolerance as assessed by graft survival and by the inability of their spleen cells to generate CTL response to C57BL/6 alloantigens. A variety of antibodies are triggered in this situation and some of them are autoantibodies with a spectrum similar to that observed in SLE. Thus, antinuclear antibodies were detected with a high incidence and were associated with anti–ssDNA and anti–dsDNA IgG1 antibodies. In addition, rheumatoid factor–like antibodies reacting with rabbit IgG and thymocytotoxic antibodies were found with various incidence in these animals. The thymocytotoxic antibodies were reacting with thymocytes of different strains, regardless of the alloantigens expressed and were active at 37°C. Neonatal induction of transplantation tolerance is known to result in long-lasting chimerism\(^\text{16}\). Since the hypergammaglobulinemia and the production of autoantibodies were already marked at 5 weeks and still observed at 24 weeks of age, one could wonder whether F1 donor B lymphoid cells were involved in their production. Indeed, anti-DNA bearing the allotype specific of the F1 donor were detected in all the tested sera.

Several mechanisms could be involved in the development of autoimmunity after neonatal induction of transplantation tolerance. It has been shown that allogeneic interactions can result in the triggering of autoreactive B cells\(^\text{17}\). During the course of the graft-versus-host reaction, the production of autoantibodies has been attributed to the reaction of alloreactive T helper cells with allogeneic autoreactive B cells\(^\text{18}\). It is possible that allogeneic interactions between host and donor lymphocytes also occur in mice made neonatally tolerant to alloantigens, in spite of the lack of demonstrable \textit{in vitro} cytotoxic reaction. The hypothesis of a T cell–dependent triggering of autoreactive B cells is consistent with the generation of autoantibodies of the IgG1 class. This hypothesis was further supported by the observation that the injection of F1 spleen cells into newborn BALB/c nu/nu mice does not lead to any autoimmune manifestation. In contrast, when spleen cells from F1 mice primed against DNP–KLH were used to induce tolerance in normal newborn BALB/c mice, there was an allogeneic effect allowing for a stimulation of the anti-DNP response in presence of DNP–conjugated to another carrier.

In addition to hypergammaglobulinemia and autoimmunity, mice made neonatally tolerant to alloantigens developed \textit{features of IC disease}. Indeed, as early as 5 weeks
after the neonatal injection of F1 hybrid spleen cells, IC were detected in the circulation and deposits of immune reactants were found in various organs. In kidneys, typical features of membranous glomerulonephritis were observed with slight impairment of the glomerular function as indicated by the increased albuminuria. Immunoglobulin deposits were also detected in the choroid plexus and along skin basement membrane. A marked thrombocytopenia was also seen and the direct Coomb's test was often positive. The occurrence of pathogenic IC in association with autoimmunity suggests that some of the IC could result from the reaction of autoantibodies with corresponding antigens. The possible formation of DNA–anti–DNA IC and their involvement in the pathogenesis of the renal lesions should be particularly considered, as suggested by the detection of anti-ssDNA antibodies in kidney eluates. Antigenic determinants of immunoglobulins are other potential candidates for the formation of IC. In mice neonatally injected with F1 hybrid lymphoid cells, one should particularly consider the possibility of interactions between certain immunoglobulins of the host and corresponding anti-idiotypic antibodies produced by F1 hybrid donor cells.

In conclusion, this new model of lupus–like diseases indicate that a chimeric state associated with an incomplete tolerance to corresponding alloantigens can lead to “abnormal” T help, similar to that occurring during graft–versus–host disease, and generate autoimmunity. Similar potentially pathogenic situations do exist in man in congenital chimerism and after bone marrow grafts. Features of SLE have been described in such circumstances17, 18. In addition, triggering or exacerbation of SLE might result from foeto-maternal lymphocyte transfer during pregnancy or from blood transfusion–induced chimeric states.

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References


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