Current Concepts of Autoimmunization:  
An Interpretive Review

By WILLIAM DAMESHEK, ROBERT SCHWARTZ AND HERMAN OLINER*

WHEN an animal of one species is injected with the serum, tissues or blood cells of another, it forms antibodies against the injected material; this is known as heteroimmunization. Likewise, animals of one species, when injected with cells or tissues of animals of the same species, usually react immunologically; this is isoimmunization. Thus, Ehrlich and Morgenroth,1 when they injected red cells of goats into other goats, were able to produce antibodies (i.e., isoheemolysins, isoagglutinins). However, Ehrlich was never able to induce the formation of antibodies by injecting the cells or tissues of one animal back into the same animal; i.e., he was unable to induce autoimmunization. From these negative experiments, Ehrlich formulated his famous concept of horror autotoxicus, which indicated that the body could not—perhaps would not—produce antibodies against itself, since this would lead to self-destruction. Nevertheless, within a few years of this formulation, Donath and Landsteiner2 demonstrated, in a patient with paroxysmal cold hemoglobinuria, an autohemolysin—a substance which had the property of attacking and destroying the patient's own red blood cells under appropriate conditions of body temperature. The validity of these observations, reported at the turn of the century, has been maintained over the ensuing years.

In the early years of the 20th century, the French physicians Chauffard and Troisier3 and Widal, Abrami and Brulé4 demonstrated autoheemolysins and autoagglutinins in various forms of acquired hemolytic anemia. This work was forgotten or dismissed as of no significance for a generation and then rediscovered.5 Since then, with the development of newer technics for the demonstration of antibodies either within the blood or adherent to cells or tissues, the label of autoimmunization has gradually been applied to a number of clinical conditions. Thus, an ever-increasing number of workers are convinced that many cases of acquired hemolytic anemia, idiopathic thrombocytopenic purpura (ITP), systemic lupus and rheumatoid arthritis develop as the result of a process of autoimmunization. On the other hand, this view has been attacked on two grounds: (1) The antigen for the presumed antibody has ordinarily not been found, and (2) experimental autoimmunization has not—until recently—been demonstrated.

Witebsky, for many years one of the foremost opponents of the concept of autoimmunization, consistently stressed these two features: (1) Know the antigen, and (2) reproduce the autoimmune process experimentally! By a
strange quirk of fate and experimentation, Witebsky was able to fulfill both of these conditions in his work on experimental thyroiditis and thus to place the field of autoimmunization on a much firmer basis. From his clinical and experimental observations, he adduced the following postulates as essential for proving auto-immunization to be present in a given case:

1. Direct demonstration of free, circulating antibodies that are active at body temperature, or the demonstration of cell bound antibodies by indirect means.
2. Recognition of the specific antigen against which this antibody is directed.
3. Production of antibodies against the same antigen in experimental animals.
4. Appearance of pathologic changes in the corresponding tissue of an actively sensitized animal that are similar or identical to those seen in human disease.

In the hematologic disorders which are usually considered as autoimmune, it has ordinarily been impossible either to define the antigen or to reproduce the disease in animals. The occasional finding of antibodies directed against specific red cell antigens in certain cases of autoimmune hemolytic anemia has not helped the cause greatly. Thus, autoimmunization has remained for some an unproved and highly speculative concept and therefore unacceptable pending further evidence. It should be pointed out, however, that certain known facts lend strong support to the concept that autoimmunization may indeed be important in the pathogenesis of certain diseases of man. Thus, in autoimmune hemolytic anemia, a protein substance on the surface of the red blood cell has been demonstrated by the antiglobulin technic; this protein can furthermore be isolated and characterized. It is a gamma globulin with specific immunologic affinity for normal red cells, and its gradual reduction with therapy is associated with clinical and hematologic improvement. Furthermore, this globulin, which without doubt has arisen within the individual's own body, has the capacity to shorten the red cell survival time not only of the patient but of other individuals as well. The only difference between the reactions of this antibody and those found, for example, in the isoemolytic anemia of erythroblastosis fetalis is that in the latter condition, the antigen is known, whereas in autoimmune hemolysis knowledge of the antigen has been conspicuously lacking. Many attempts to produce autoimmune hemolytic anemia experimentally have ended in failure. Perhaps the constant emphasis on "What is the antigen?" has dulled thinking to such an extent that the important possibility of an abnormal type of antibody productive mechanism has been ignored. Recently, our experience both in the clinic and the laboratory has led us more into the latter channel, and this will be explored below.

**The Usual or Normal Immune Response**

The earliest discernible reaction following the introduction of antigen into an animal is the proliferation of certain cells located in or in close relationship to the reticuloendothelial system. Immunologic responses, long thought of as
agglutinins, hemolysins, and the like, are fundamentally cellular. The usual forms of immunization may be considered as temporary proliferations of immunologically competent cells. In autoimmunization, one is dealing with a continuous, self-perpetuating type of proliferation, which, unlike leukemia, is not invasive and does not metastasize. On the other hand, many relationships may be discovered between the leukemic proliferations and the autoimmune state. It seems appropriate to return to the cell as the basis of the immunologic reactions—whether hetero-, iso- or auto- in type.

As has been demonstrated by the elegant immunofluorescent technics of Coons and his collaborators, the cells which proliferate in response to an antigenic stimulus produce a protein of the globulin variety, and after a relatively brief time a new globulin appears in the blood. This protein, which has the capacity of specific chemical union with the substance which provoked its appearance is called antibody. It is evident that this response may serve as a protective measure against the invasion of foreign material into the host environment. How the body cells can discriminate between what is “self” and what is foreign or “not self” is quite obscure, as are the actual mechanisms by which antibody is produced. In recent years, two main concepts have evolved from the central question of the mechanism of antibody formation: The first proposed that antigen serves to instruct a homogeneous population of potential antibody forming cells to produce protein of a specific type, while in the latter, the role of antigen is to select from a heterogeneous population a clone of cells genetically destined to produce a highly specific antibody molecule. These questions, which are agitating the minds of immunologists everywhere, are beyond the scope of the present discussion. It is apparent that the usual immunologic response, both in the experimental animal (heteroimmunization) and in the clinic (isoimmunization), is provoked by substances foreign to the body. The autoimmune reaction is evidently different; its genesis is obscure; the antigen is usually elusive; once begun, autoimmunization usually continues indefinitely; and, unlike the usual or normal antigen-antibody response, it is by no means beneficial to the body. In fact, the autoimmune process is actually harmful and results in an attack on cells and tissues, thus leading to such clinical conditions as autoimmune hemolytic anemia, thrombocytopenic purpura and systemic lupus. How then does the autoimmune process originate?

Mechanisms by which Autoimmunization May Develop

Two mechanisms by which autoimmunization develops are at least reasonably clear and well documented. These are (1) through the “hapten” mechanism and (2) via the breakdown of anatomic or physiologic isolation.

In the hapten mechanism, so clearly demonstrated by Landsteiner, a chemical apparently becomes attached to a normal body component (red cells, platelets, leukocytes), with the result that a complex is formed which has antigenic potentiality for the individual’s own antibody productive cells. This represents an alteration of “self” to “not self,” to use Burnet’s older nomenclature. In any event, there can be no question that such a combination, however loose, between body-own material and hapten may be antigenic with
the result that antibody (autoantibody) is produced. This is clearly seen in Sedormid\textsuperscript{18} or quinidine-induced\textsuperscript{19} ITP. Here, the development of sensitization always requires the previous administration of the drug. Successive re-administrations result in the development of an antiplatelet antibody which acts only in the presence of the drug, and, when this occurs, further re-administration of the drug results in the sudden development of thrombocytopenia. The resultant antibody is unusual in that it can act against the specific cell or tissue component only in the presence of the specific hapten. Thus, the antibody alone does not attack platelets; antibody plus quinidine will, however, result in agglutination of platelets or in their lysis when complement is present. Complement fixation has been shown to occur during the immunologic reaction and when passively transferred to another individual the antibody induces thrombocytopenia only when quinidine or Sedormid have been administered to the recipient. There seems little question, therefore, that this is a true example of autoimmunization.

(2) The development of autoimmunization, apparently due to the liberation of previously isolated tissue substances, has been demonstrated rather convincingly with the lens of the eye,\textsuperscript{20} with thyroglobulin\textsuperscript{21} and with spermatozoa.\textsuperscript{22} Certain tissues or substances which develop and mature in isolation from the reticuloendothelial system could conceivably act as antigens if they were liberated into the circulation at some future time. Although no direct proof exists, it is conceivable that such isolation from immunologically reactive cells would prevent the development of immunologic tolerance to these isolated structures. Thus, these materials, when liberated at some later time into the circulation of the mature individual, might be considered as foreign or "not self," and thus of possible antigenic potentiality. This would result in the formation of antilenticular or antithyroglobulin antibody and thus in injury either to lens (sympathetic ophthalmia) or to thyroid (thyroiditis). The latter phenomenon has been studied extensively by Witebsky.

In a series of elegant experiments Witebsky and his colleagues\textsuperscript{23} demonstrated that (a) crude rabbit thyroid extracts are antigenic in rabbits; (b) thyroidectomized rabbits reinjected with extracts of their own thyroid tissue produce thyroid specific antibodies; (c) hemithyroidectomized rabbits
injected with extracts of their own thyroid gland not only form thyroid-specific antibodies, but the remaining portion of the thyroid gland undergoes extensive morphologic changes characterized by atrophy and infiltration by lymphocytes, eosinophiles and plasma cells, changes resembling those seen in patients suffering from chronic thyroiditis; (d) many patients with chronic thyroiditis have thyroid specific antibodies in their serum. These findings may be said to have established the role of autoimmunization in the production of chronic thyroiditis in man and experimental animals on a firm basis.

It should be mentioned at this point that some authors have considered the formation of autoantibodies a normal process, and autoimmune disease the end result of a pathologic exaggeration of this physiologic activity.24 Little attention has been paid to this possibility, except by Tyler, who demonstrated a globulin with anti-venom activity in the serum of the Gila monster, notable for its very potent venom.25 The teleologic significance of this finding is obvious. Tyler has also constructed an intriguing theory of embryologic development, based on autoimmune concepts.26 Perhaps the cold hemagglutinin found in low concentration in so many normal people is another example of the physiologic production of autoantibody. The accumulation of these cold agglutinins at sites of local trauma in normal individuals27 may represent, in miniature, the pathologic exaggeration of normal autoantibody formation.

A Third Form of Autoimmunization

The present argument or thesis regarding autoimmunization derives in part from the common finding of autoimmune hemolytic anemia and other immunologic disturbances (ITP, white cell antibodies) in the lymphoproliferative disorders (chronic lymphocytic leukemia, lymphosarcoma)28,29 and in part from studies of runt disease. From both these clinical and experimental observations, it has become apparent that neoplastic (i.e., leukemic, lymphosarcomatous) or transplanted immunologically competent tissues may produce antibody which can then attack the host’s own cells or tissues. This thesis will be developed below.

Green of Sheffield has put forward the ingenious hypothesis that since the lymphocytes of chronic lymphocytic leukemia are presumably abnormal, normal red cell antigen coming in contact with these presumed antibody producing cells might not be recognized as “self,” with the result that red cell antibody would be produced, thus inducing autoimmune hemolytic anemia.30 We have also stated a somewhat similar position in discussing the mechanisms of hemolysis in the various lymphoproliferative disorders.28

The second body of evidence, which has a far firmer base than the clinical viewpoint, has come from the experimental work in runt disease. It is apparent that this disorder represents a reaction of grafted tissue against the recipient (graft vs. host) in which certain cells of the graft produce antibodies which attack various cells and tissues of the host animal. Runt disease may be said to develop under the following circumstances: (a) acceptance of the grafted tissue by the host animal, (b) the presence of immunologically competent cells in the transplanted tissue and (c) an antigenic difference between host and graft.
In the experiments done in our laboratory, runt disease was induced by the injection of parental strain spleen cells into normal adult F1 hybrid mice. In these animals not only did a positive antiglobulin test develop but there was hemolytic anemia, thrombocytopenia and leukopenia. Thus, the injection of host-tolerated, immunologically competent cells resulted in various disturbances similar to those seen in autoimmune diseases of man. This graft vs. host reaction could be conceived of as one involving the reaction of abnormal antibody-producing cells vs. body-own antigen. This can be contrasted with the normal or usual immunization procedure, which is that of foreign antigen vs. normal antibody producing cells. Fundamentally, the antigen-antibody reaction may be the same, but in the graft-host reaction, the central feature is the presence of foreign antibody producing cells, whereas in the usual immunization procedures it is the antigen which is foreign, with the antibody-productive cells being normal. The constant stimulation of abnormal antibody production by the normal host antigens could result in the "self-perpetuating" feature so characteristic of autoimmune disease.

Runt disease, an experimentally-induced reaction, may have its counterparts in the clinic. For example, chronic lymphocytic leukemia, which might be interpreted as a condition in which a large group of foreign, but host-tolerated immunologically competent cells has developed, is often associated with autoimmune hemolytic anemia. The latter disturbance may be present at the beginning of the illness, or may occur rather quickly following spray x-ray therapy or after a large dose of one of the alkylating agents. As already suggested by Kaplan and Smithers, the sudden development of autoimmune hemolytic anemia after radiation may be due to the breakdown of previously existing immunologic tolerance between host (patient) and "foreign" (leukemic) cells, thus leading to a "graft-host" disturbance. We have recently shown by the fluorescent antibody technic that the lymphocytes of chronic lymphocytic leukemia appear to manufacture the red cell antibody responsible for the hemolytic anemia.

How can one explain the situation in idiopathic autoimmune hemolytic anemia, in which there is no indication of undue lymphocytic proliferation, leukemia, or ordinarily of any other disease? Here the possibility is present that as the result of various inciting factors (viral, chemical, radiation, etc.) groups of abnormal antibody-producing cells develop which then result in the production of autoantibody by the same formula, i.e., normal red cell antigens stimulating abnormal clones of immunologically competent cells to produce antibody. Burnet has alluded to this possibility and has suggested that in some diseases, e.g., systemic lupus, "forbidden" clones may become activated via various "trigger" mechanisms. The concept of "forbidden clones" suggests that some clones previously dormant, perhaps genetically abnormal, become activated, thus resulting in abnormal globulins. The reaction of these with the antigens of normal cells and tissues results in what is called autoimmunization. Although this is an interesting speculation, it does not seem essential to the burden of the present discussion, since it is conceivable that groups of cells can be made abnormal by somatic mutation induced by viruses, radiation, etc., without having been previously selected or isolated.
We have considered that systemic lupus is a complex autoimmune disease in which, for some obscure mechanism, a spectrum of abnormal antibodies arises within the body which then attack the patient's cells and tissues resulting in the protean manifestations of the disease. In our present thinking, the possibility is considered that SLE may be a clinical analogue of runt disease. Thus, it is conceivable that clones of abnormal antibody-producing cells (it should be pointed out, however, that the actual demonstration of clones of lymphoid cells has not yet been made) develop in some individuals—some of them active at one time, some at another. This would result in, for example, ITP and arthritis today, and at a later time in endocarditis, nephritis, or other clinical abnormalities. Finally, a whole complex of disturbances is present simultaneously. If this idea is acceptable, one has to explain how these clones of cells arise within the individual. Recourse can be had to two possibilities mentioned above: (1) immunologically competent cells which may have been transferred from the mother in fetal life become abnormally active; (2) groups of abnormal cells develop by some means (mutation?); these may then attack the patient by the production of one variety of antibody after another. The well-known sensitivity of patients with systemic lupus to sunlight suggests that this nonionizing but potent source of radiation may accelerate this process. These concepts are substantially the same, the important point being the presence of groups of abnormal antibody producing cells which are tolerated by the host. In fact this is the crux of the autoimmunization hypothesis as we see it: "foreign" or abnormal antibody-producing cells living within the milieu of a host of normal antigens with the result that abnormal autoantibodies may be produced having harmful effects on the host's cells or tissues.

There can be little doubt that the mechanism of autoimmunization, at one time received with considerable skepticism, must now receive serious consideration as a prime factor in many diseases, ranging from hemolytic anemia to glomerular nephritis and from ITP to rheumatoid arthritis. At first considered a possibility in certain forms of hemolytic anemia, it was later postulated in ITP, thrombohemolytic thrombocytopenia, systemic lupus and more recently in many other disorders including rheumatoid arthritis, nephritis, certain forms of hepatitis, and even in other conditions such as those involving the central nervous system. That a group of diseases—autoimmune disorders—which make up a distinct segment of the disorders affecting man, are just as important in their way as other groupings of disease (infectious, hereditary, neoplastic) can no longer be denied. In this speculative discussion, three mechanisms by which auto-immunization may develop have been discussed: these are the hapten, the "isolated" antigen and the abnormal clone. Chief emphasis has been placed on the third form, whose basis may be sought for in such conditions as the lymphoproliferative disorders and the graft vs. host reaction.

REFERENCES


William Dameshek, M.D., Director, Blood Research Laboratory, New England Center Hospital; Professor of Medicine, Tufts University School of Medicine, Boston, Mass.

Robert S. Schwartz, M.D., Research Associate, Blood Research Laboratory, New England Center Hospital, Boston, Mass.

Herman L. Oliner, M.D., Senior Research Fellow in Hematology, Blood Research Laboratory, New England Center Hospital, Boston, Mass.
Current Concepts of Autoimmunization: An Interpretive Review

WILLIAM DAMESHEK, ROBERT SCHWARTZ and HERMAN OLINER