VIRUSES AND THE CONNECTIVE TISSUE DISEASES

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Viruses and the Connective Tissue Diseases

VIRAL ARTHRITIS

The occurrence of arthritis with viral infections such as rubella, mumps, smallpox, vaccinia and serum hepatitis is well known[1]. A number of other virus infections have been reported to produce arthritis but in lower frequency. Isolation of virus from the joint has only rarely been accomplished[2,3]. At the present state of our knowledge, it is not certain whether the arthritis in these cases results from a direct toxic effect of the virus or from the local action of immune complexes made up of virus and anti-viral antibody. Modern immunologic techniques may help to establish at least whether virus is present in the joint. This is illustrated in the following case studied in detail by Dr. Robert Johnson. It appears to be the first case of serum hepatitis with arthritis in which Australia antigen was demonstrated in the synovial fluid of an actively involved joint.

SERUM HEPATITIS WITH POLYARTHRITIS

[redacted], [redacted], [redacted]

In [redacted] of 1970, the patient, a 24-year old [redacted] man, sustained open lacerations over the left fourth MCP joint in a brawl. About three months later, he developed fever, anorexia, vomiting, jaundice, dark urine, and a lightening of the color of the stools. He remained in bed because of extreme fatigue. Four days after resuming activity, he went to a blood bank to sell his blood and was refused because of icterus. He then returned to work as a carpenter. At this time he felt well, but after ten days he noted pain, swelling and stiffness in the left ankle, followed shortly by pain and warmth in the knees.

On [redacted]/70, he presented to the [redacted] emergency room with a two-week history of arthritis. Left knee and right elbow were swollen. On [redacted], his physician injected his joints with Triamcinolone; with this he obtained relief for about 12 hours. On [redacted], he was seen in the [redacted] emergency room again; he had a stiff right elbow and the left knee and both wrists were warm and swollen. WBC was 14,100 with left shift; ESR 48 mm/hr; latex fixation test negative. On [redacted], 20 ml of yellowish-green turbid fluid with reduced viscosity was aspirated from the left knee. Cultures were negative. Serum immunoelectro-osmophoresis was negative for Australia antigen ([redacted]) but the synovial fluid was positive.

On Indocin 400 mg/day, he obtained excellent relief and was again able to walk on the affected knee. When he discontinued the Indocin, his joint symptoms recurred severely. He was admitted to [redacted] on [redacted].

Examination revealed involvement of the right elbow, left knee, left wrist, an MCP joint, and right ankle.

Laboratory studies: WBC 15,800 with 72% neutrophils; LE preparation and latex fixation tests negative. Serum Australia antigen and antibody titers on the serum were negative by immunoelectroosmophoresis. Synovial fluid had 1,650 WBC, and a protein content of 6.7 gm%. There was no growth on bacterial culture. Immunoelectroosmophoresis for Australia antigen on the synovial fluid was again positive (Liver Unit, UTSWMS). Biopsy of synovial tissue showed proliferative synovitis. Alkaline phosphatase was 85 units (normal 30-85); SGOT, 54. Liver biopsy findings were consistent with "resolving or resolved inflammation." ASO titer and renal function tests were normal. The patient was placed on Indocin and his symptoms rapidly improved. He was discharged on [redacted].

The patient was followed in clinic with continuing polyarthritis responding to Indocin and ASA. On [redacted]/71, he had objective involvement of eight joints. Liver was not enlarged. Serum and synovial fluid latex fixation tests remained negative. However, serum and synovial fluid were both positive for Australia antigen by Ouchterlony agar diffusion (Rheumatic Diseases Unit, UTSWMS). Hemolytic complement levels on both the serum and synovial fluid samples were in the normal range. The presence of a lymphotoxin was demonstrated in the synovial fluid.

Comment: That an immune response was involved in this arthritis may be supposed from the fact that the patient developed arthritis about three weeks after onset of his symptoms, at a time when he would have circulating antibody. This is similar to experience with rubella in children[4], in whom the onset of arthritis generally coincides with the appearance of antibody. Arthralgia or arthritis has also been reported in 39% of patients who have received rubella vaccine[5]. This usually occurred during the second and third week after immunization, at the time of the first appearance of circulating antibody. Mumps arthritis, too, usually appears about 10 days after the onset of the parotitis, at a time when serum antibody titers are rising[6].

VIRUSES AND MULTISYSTEM DISEASE OF ANIMALS

The possible role of viruses in producing multisystem disease received its stimulus from two animal diseases, New Zealand Black (NZB) disease and Aleutian mink disease. Both of these have features in common with the connective tissue diseases, particularly systemic lupus erythematosus.

Aleutian mink disease is characterized by generalized vasculitis, nephritis, mononuclear infiltration of the liver, hypergammaglobulinemia and the occurrence of autoantibodies[7]. It has been transferred by ultrafiltrates of spleen suspensions of infected mink[8] and is accepted as a virus-induced disease.

NZB x NZW F1 hybrid mice have a spontaneous disease, characterized by glomerulonephritis and by positive tests for the LE factor and antinuclear antibody. The glomerulonephritis has the characteritics of an immune complex disease in which discrete deposits of gamma globulin are demonstrable in the glomerular basement membrane[9]. These complexes have been shown[9] to contain DNA and anti-DNA antibody.

Murine Leukemia Viruses in NZB Mice and Autoimmunity - Older NZB mice[10] have a high incidence of malignant lymphomas. Since murine leukemias and lymphomas of several varieties are caused by viruses, Mellors and Huang[11] were impelled to determine whether the immunopathologic disorders of these mice might also be caused by such viruses. They found that when they inoculated weanling NZB mice with a filterable agent obtained from NZB lymphoma tissue, many of the pathologic changes ordinarily present in older animals developed in young animals. In the electron microscope, they identified particles in the tissues of these animals which resembled murine leukemia virus. East et al[12] and Prosser[13], confirming this finding, also identified the viral particles in embryos and in germ-free mice fostered on germ-free mice of another strain, suggesting that they were vertically transmitted. These particles have been observed in other strains, but mainly in the thymus.

In spite of the fact that murine leukemia viruses are apparently transmitted vertically in NZB mice, this strain does not develop tolerance to the Gross leukemia virus in contrast to other strains. Antibody to the Gross antigen is produced in early life, and as antibody is produced, there is a gradual elimination of the Gross soluble antigen from the blood[14]. Corresponding with the disappearance of the antigen, proteinuria and mortality develop. Recently, Mellors and coworkers[15] have, in fact, demonstrated the presence of MuLV antigen in the glomeruli of NZB x NZW mice by immunofluorescence, thus implicating leukemia viruses in the

glomerulonephritis of these animals. In addition, Gross virus soluble antigen and antibody were extracted from the kidney. The association of glomerulonephritis with leukemia virus is not limited to the NZB mouse. It has been observed in other strains infected with Rauscher, Friend or Moloney virus[16].

Non-leukemogenic Viruses and Renal Disease - Parallel with the above experiments indicating a role for leukemogenic viruses in the production of a renal lesion, has been a series of papers from the laboratory of Dr. Frank Dixon, demonstrating that non-leukemogenic virus antigen-antibody complexes are capable of producing glomerular lesions. It was shown[17,18] that mice infected at birth by lymphocytic choriomeningitis (LCM) virus, contrary to previous ideas, were not immunologically tolerant to this virus, but did, in fact, produce anti-LCM antibody throughout most of their lives. Like antibodies to a number of viruses, this was not protective antibody (possibly because of the presence of excess antigen), leading to the false impression that the infected mice were tolerant. The glomerulonephritis of chronic LCM disease had the characteristic of an immune complex nephritis in that circulating LCM virus-anti LCM virus complexes and complement appeared to have been trapped in the glomerulus.

It was further observed[19] that two quite dissimilar viruses, LCM, an RNA virus, and polyoma, a DNA virus, both intensified a number of the abnormalities in the NZB and hybrid mice. The antinuclear antibody (ANA) response was enhanced, the glomerulonephritis was aggravated and the associated mortality increased.

The observation of Lambert and Dixon[20] that NZB x NZW mice produce more anti-DNA upon immunization with DNA-methylated albumin than other strains has led these authors to the conclusion that there is a native hyperreactivity to DNA in this strain. However, this alone was not thought sufficient to cause disease in these mice because the parental NZW strain also manifested hyperreactivity without developing disease. Accordingly, these authors have suggested that chronic virus infection, either by leukemia virus or by others such as LCM or polyoma, might in some non-specific manner lead to the development of disease. This susceptibility to chronic viral infection could be derived from the NZB parent, which has been claimed by Mellors to be especially susceptible to murine leukemia virus infection.

VIRUS-INDUCED MULTISYSTEM DISEASE IN MAN

With the stimulus provided by the experience with virus-induced multisystem disease in the mouse and the availability of a sensitive technique for the immunological identification of at least one commonly occurring virus of man, it is not surprising that viral-induced multisystem disease was soon demonstrated in man. Gocke et al[21] have recently described four patients among eleven with biopsy proven polyarteritis nodosa who had Australia (Au) antigenemia. The patients were characterized initially by fever, polyarthralgias, myalgias, rash and urticaria. Over a period of 2 to 4 months, hypertension and peripheral neuropathy developed and eosinophilia, hematuria, and azotemia appeared. Vasculitis was demonstrated by muscle and liver biopsy. In one patient studied in detail, deposits of Au antigen, IgM and the β_{1c} component of complement were demonstrated by immunofluorescence in the blood vessels of muscle.

The incidence of four in eleven patients was regarded as significant, since in a prospective study of post-transfusion hepatitis, the frequency of Au antigenemia was only about one in 100.

Following the presentation of this paper, sera from ten patients with polyarteritis nodosa, which were available in our serum bank, were tested; positive reactions were found in three. This is a highly significant incidence in view of the fact that the incidence of positive Au antigen tests in the Dallas hospital population is between 0.2% and 0.6%. The case histories of two of the patients are given below.

POLYARTERITIS NODOSA, AUSTRALIA ANTIGEN POSITIVE

[redacted]., [redacted]

A 44-yo [redacted] woman was admitted to [redacted] on [redacted]/69 with a 3-month history of nausea, vomiting and epigastric pain. CBC was within normal limits, icteric index 6, amylase less than 320. One month before she had a transient maculopapular rash on the hands, feet, and trunk which disappeared without special treatment. The patient had been previously treated in the Outpatient Clinic with a 10-year history of hypertension.

A diagnosis of acute cholecystitis was made and an emergency operation was performed on the evening of admission. An inflamed gall bladder was resected at surgery. There was no other palpable pathology.

Workup following surgery showed a WBC of 16,000 which rose to 20,000 during the convalescent period; urine negative, bilirubin 1.2 mg%; SGOT 81 units, falling to 64 units; alkaline phosphatase 16 KA units, rising to 30 units; LE preparation negative.

Histological examination of the gall bladder revealed a necrotizing vasculitis in many arteries. Diagnosis: polyarteritis nodosa.

The operative site healed badly and required reclosure. The patient was discharged on [redacted]/69, appearing weak and depressed. She returned to the hospital a few weeks later because of increasing weakness and was readmitted. In the hospital, the wound opened once again and was again closed on [redacted]. About this time, there was progressive weakness of the upper and lower extremities with numbness and diminished reflexes. She also complained of severe abdominal cramps. The symptoms of peripheral neuropathy progressed to bilateral wrist drop with loss of sensation. These changes were accompanied by the onset of diffuse muscle tenderness. Electromyogram confirmed the presence of a neuropathy. At this time, also, the operative site had developed a draining fecal fistula and the patient had become psychotic. She appeared gravely ill.

In the face of a non-healing operative wound and fistula, the patient was placed on prednisone 40 mg and Cytoxan 100 mg daily for the treatment of polyarteritis nodosa. Within approximately two weeks, she became more oriented. After three weeks, there was some recovery of sensation and disappearance of abdominal cramps. Cytoxan dosage was raised to 200 mg daily and improvement continued. At a month, there was much improvement in sensation and the wound appeared to be healing. She was discharged on prednisone 30 mg and Cytoxan 100 mg daily.

At home, the patient continued to improve on the prednisone-cyclophosphamide regimen until [redacted]/69 when she developed upper abdominal pain and vomiting and was admitted to the hospital. Cytoxan was halted and prednisone dosage tapered. She was treated for a pneumonia associated with mixed flora. Hyperglycemia, due to steroid-induced diabetes, was treated with insulin. She was discharged on [redacted]/70.

By [redacted], 1971, one year after the diagnosis of polyarteritis nodosa, the patient had shown a remarkable remission and was taking only 5 mg of prednisone daily. When last seen on [redacted]/71, two years after the diagnosis, she continued to be remarkably well on 5 mg of prednisone daily. There was only slight weakness of flexion of the right index finger; there was no sensory defect;

the feet were normal. General health was good; abdomen was asymptomatic. Liver was palpable 8 cm below the right costal margin. The following were her laboratory findings: bilirubin 0.3 mg%, cephalin flocculation 1+/2+, thymol turbidity 16U, alkaline phosphatase 21 KA units, SGOT 127U, aldolase 3.5U, creatine phosphokinase 50U, A/G 5.0/3.8, serum complement level normal, latex fixation 2+, sensitized sheep cell agglutination titer 1:56 (positive), immunoglobulin levels by radial diffusion, normal.

Five measurements of Australia antigen in the serum, made between [redacted]/69 at the time of the first admission and [redacted]/71 when the patient was last seen, were all positive.

Comment: Treatment of a patient with polyarteritis nodosa and Au antigenemia with prednisone and cyclophosphamide has been followed by a dramatic remission which has lasted for over 18 months. Au antigenemia persists. Whether viral immune complexes are still present in the serum is to be investigated.

POLYARTERITIS NODOSA, AUSTRALIA ANTIGEN POSITIVE

[redacted]

A 36-yo [redacted] woman developed pain and swelling of the finger joints and feet in [redacted], 1965. She was admitted to another hospital in New Mexico and treated with 40 mg of prednisone daily with improvement. On reducing the dosage, her symptoms exacerbated and she developed a fever. This was followed by weakness and paresthesias in the upper and lower extremities and numbness of the fingers and toes. Liver function tests showed elevated BSP retention and alkaline phosphatase levels. Liver biopsy was read as "post-hepatitic cirrhosis with chronic active hepatitis."

The patient was referred to Dr. Combes and admitted to [redacted]. Physical examination disclosed gallop rhythm but no cardiac enlargement, a markedly enlarged liver and palpable spleen tip, major loss of muscle mass both proximally and distally, tenderness in the deltoid muscles, interosseus atrophy, left foot drop and decreased deep tendon reflexes.

Laboratory tests: ESR 38 mm, WBC 11,900 with left shift and elevated muscle enzymes in the serum. BSP retention 30%, icteric index 4.1, alkaline phosphatase 17 KA, SGOT 55, total bilirubin 1.3 mg% and cephalin flocculation 3+/4+. Serum immunoosmoelectrophoresis for Australia antigen was positive.

The liver biopsy previously taken was reread at PMH. It showed marked small and medium artery inflammation. A muscle biopsy showed focal arteritis. *Impression:* Polyarteritis nodosa. Electromyogram showed neuropathic changes. The patient was discharged on [redacted]/69 on prednisone therapy. At home, she had a rapid downhill course and died on [redacted]/69. Autopsy disclosed multiple organ involvement.

VIRAL GLOMERULONEPHRITIS IN MAN

A variety of renal abnormalities have been described with systemic viral infection[21a]. The following is a case of nephrotic syndrome with Au antigenemia, described recently by Shorey et al of this department. It represents the first case in which a viral antigen has been demonstrated in the human kidney.

AUSTRALIA ANTIGEN-INDUCED MEMBRANOUS GLOMERULONEPHRITIS

[redacted] [redacted]

A 53-year old [redacted] man was well until he suffered severe injuries in an automobile accident in [redacted], 1968. He was transfused with four units of whole blood. Four months later, in [redacted], 1969, he developed acute hepatitis as an apparent complication of the earlier transfusions. He was started on prednisolone by his physician and the serum bilirubin returned to normal. However, the SGOT level remained persistently abnormal suggesting continued hepatic activity. In [redacted], 1970, pedal edema appeared and in [redacted], abdominal swelling with ascites. Urinalysis revealed 4+ proteinuria.

The patient was referred to the Liver Unit late in September for appraisal of his chronic liver disease. At this time, his serum was found to contain Australia antigen and this has persisted in all subsequent determinations. Serum albumin was less than 3 gm% when edema was first observed and reached a nadir of 1.7 gm% with the onset of ascites. In November, 1970, the patient was admitted to this hospital. In the hospital, protein excretion ranged between 1.2 and 3.9 gm per 24 hours. C3 was 195 mg% (normal). Renal biopsy showed diffuse thickening of the capillary walls and slight mesangial thickening. There was no cellular proliferation and the glomerular capillaries were patent. The findings were characteristic of a diffuse membraneous glomerulonephritis.

In the electron microscope, dense deposits were seen widely distributed along the subepithelial margin of the glomerular basement membrane. There was generalized fusion of the epithelial foot

processes. Upon immunofluorescent staining, IgG and the C3 component of complement were observed in a granular distribution. In addition, Australia antigen was demonstrated in the glomeruli using the indirect immunofluorescent staining technique.

Comment: Of the four patients with Au antigenemia and extrahepatic involvement presented, this is the only one with renal involvement. Presumably, the amount and type of immune complex formation determines this happenstance.

ENDOTHELIAL CYTOPLASMIC INCLUSIONS

Another reason for interest in the role of viruses in the connective tissue diseases has been the observation of myxovirus-like endothelial cell inclusions in the tissues of patients with SLE. Such interwoven tubular structures were first seen by Shaw[22] in the brain of patients with subacute sclerosing panencephalitis, a complication of measles. In this condition, measles virus has been isolated from the brain. Subsequently, Chou[23] saw similar structures (though smaller than usually seen in SLE) in muscle from a patient with polymyositis; and Fresco[24] observed somewhat larger structures than usually seen in SLE in the dense deposits of the glomerular basement membrane of an SLE patient.

In 1969, Gyorkey et al[25] and Norton[26], almost simultaneously described the presence of characteristic endothelial cell cytoplasmic inclusions in two series of patients with SLE. Both laboratories observed the structures in kidney tissue; Norton also observed them in muscle and skin. In the kidney, they were present in the cytoplasm of glomerular endothelial cells and consisted of interwoven tubules, 230 Å in outer diameter, 80 Å in inner diameter and up to 1000 Å in length. Shortly after, their occurrence was desdribed by Hurd, Eigenbrodt, Ziff and Strunk[27,28], not only in systemic lupus erythematosus, but also in other kidney diseases.

In appearance, these structures resembled the internal ribonuclear component of unenveloped nucleocapsids of myxo or paramyxo viruses. They were also similar in size and morphology to the virus of Herpes simplex encephalitis in man. Moreover, they appeared to be in close contact with the endoplasmic reticulum. Such structures have since been reported in a number of sites in the skin of discoid lupus erythematosus[29], in the capillary endothelial cells of skin and muscle in patients with dermatomyositis[30], in the kidney of patients with Sjogren's syndrome[31], scleroderma[30], and Goodpasture's syndrome[30], and skin of a patient with congenital rubella syndrome[29].

Hurd et al[28] have observed the structures in renal biopsies of all of 35 SLE patients, 24 of 113 non-SLE patients and in none of 8 volunteer prisoners. The non-SLE patients had acute glomerulonephritis, chronic glomerulonephritis, gold salt nephropathy, sickle cell disease with nephritis, secondary syphilis and eclampsia. Inclusions were seen in five biopsies of SLE patients whose kidneys were normal by light microscopy. When observed in non-SLE patients, the inclusions were far less numerous than in the SLE group.

Identity of tubular structures - Three possibilities exist for the identity of the endothelial cell inclusions. They may be:

(1) Ribonucleoprotein strands of a widespread passenger virus.

(2) Cellular material released into the circulation from damaged cells and phagocytosed by endothelial cells.

(3) Altered endoplasmic reticulum.

Sinkovics[32] has suggested that they represent unenveloped viral nucleocapsids found in the replication cycle of a defective paramyxo, corona or oncoRNA virus. Attempts to stain the structures by immunofluorescence using antisera to a number of viruses have proved unsuccessful[26]. However, if they are unenveloped nucleocapsids, they may lack the required antigenic sites for antibody binding.

Pincus et al[33] have disagreed with the suggestion that the inclusions are nucleocapsids of the myxoviruses for the following reasons:

(1) They are consistently located within dilated cisternae of the endoplasmic reticulum, while myxoviruses are found outside the endoplasmic reticulum.

(2) They are larger in size than the myxoviruses.

(3) Cocultivation of SLE kidney with embryonic kidney cells, hela cells, etc. yielded no cytopathic effects; and hemadsorption, immunofluorescence and complement fixation tests were negative.

Pincus et al have pointed out that tubules of similar size and location may be seen under various *in vivo* and *in vitro* experimental conditions induced by viruses and suggested that the tubules were a unitary modification of the endoplasmic reticulum which may develop in transitional forms, particularly under the influence of viruses.

The weight of evidence would appear to be that the inclusions either represent defective inclusions of viruses themselves or represent altered endoplasmic reticulum which has undergone degeneration as consequence of the presence of virus in the cells.

VIRAL ANTIBODY TITERS IN SLE

On the assumption that the anti-DNA antibody present in SLE may be a result of viral infection, much interest has developed in the antibody titers of these patients to the more common viruses of man, particularly the myxo and Herpes-like viruses which resemble the endothelial inclusions in appearance.

In initial experiments, Phillips and Christian[34] found that antibody titers to measles and parainfluenza type I viruses were significantly increased in systemic lupus erythematosus. Titers were also increased in Reiter's syndrome.

Hurd, Dowdle, Casey and Ziff[35] have made an intensive study of antibody titers in SLE to a group of myxo and other viruses. Control groups had tuberculosis, rheumatoid arthritis, bronchial asthma and miscellaneous disease. By the complement fixation method, the only antibody significantly elevated in SLE above all control groups was measles antibody. The increase in titer, though significant, was, however, small. By the hemagglutination inhibition method, measles antibody titer was elevated significantly above all but the tuberculosis group. When the $\log^2$ antibody titers for the individual viruses were averaged, the SLE group had a greater overall titer than all control groups except the tuberculosis group. The antibody titers in SLE were not correlated with gamma globulin levels.

These data indicate that there is a moderate hyperreactivity to viral antigens in SLE patients which, nevertheless, is not significantly greater than in patients with active tuberculosis. Active tuberculosis may derive some augmentation of viral antibody titers from the adjuvant effect of the tuberculosis infection. A similar adjuvant effect of an as yet unknown nature may exist in SLE.

It is of interest that Phillips and Christian have recently[36] tested a larger group of SLE patients for measles and parainfluenza 1 antibody titers and found no significantly greater titers than in control subjects with inflammatory disease. They did observe a correlation of antibody titer with gamma globulin

level and suggested that whatever tendency to elevated viral antibody titers is noted in SLE is a result of a generalized immunologic hyperreactivity of the SLE patient.

Interest has recently turned also to the Epstein-Barr virus (EBV) in SLE. This is a Herpes-like DNA virus which is the putative cause of Burkitt's lymphoma and infectious mononucleosis. The titer of EBV antibody is known to be increased in infectious mononucleosis, Burkitt's lymphoma, nasopharyngeal carcinoma and sarcoidosis. To this list has recently been added SLE[37]. Sixty-two per cent of patients had elevated titers compared to 11 per cent in the general population. The titers were not related to the activity of the disease. It is of interest that the elevated EBV titers in sarcoidosis have also not been related to the activity of the disease[38]. The increasing number of diseases in which EBV antibody is elevated suggests that EBV may be merely a passenger virus in at least some of them. Its presence may be related, possibly to the occurrence of immunologic anergy, or to the presence of an immunoproliferative state, which may foster EBV multiplication.

STATE OF THE IMMUNE RESPONSE IN SLE

The Humoral Antibody Response - Zingale and coworkers[39] noted an increased antibody response to blood group antigens in SLE patients. Baum and Ziff[40] observed a decreased 19S antibody response to brucella antigen, and Sarkany[41] a normal antibody response to tetanus toxoid. Abe and Homma[42] have found somewhat decreased response to tetanus toxoid.
It would appear from these observations that there is no evidence of increased humoral antibody responsiveness to exogenous antigens in SLE patients.

More interesting have been recent studies of the antibody response to nucleic acids in SLE. The occurrence of anti-DNA antibodies in SLE sera is well known. This has usually been interpreted to mean that SLE patients produce these antibodies in response to either a foreign nucleic acid antigen or to their own nucleic acids. Recently, Schur and Monroe[43] have demonstrated serum antibodies to RNA in 16 of 89 patients with SLE. These were directed primarily toward double-stranded RNA (poly I-C, poly A-U and Statolon viral RNA). Since almost all of the RNA of the body is of the single-stranded type, these findings have suggested that the anti-RNA antibodies in SLE are directed primarily against a foreign antigen such as a double-stranded RNA virus. Similar findings have been obtained in the NZB-NZW mouse[44].

THE DELAYED HYPERSENSITIVITY RESPONSE

Although there is evidence for a decrease in delayed hypersensitivity in the NZB and F_1 hybrid mice, the status of delayed hypersensitivity in SLE is not clear. Hyporesponsiveness was noted only to PPD among five antigens tested by Block et al[45]. Bitter et al[46] reported a decrease in skin reactivity to all of seven antigens tested. They also observed decreased blastic transformation in response to phytohemagglutinin on the part of SLE lymphocytes. The mixed lymphocyte response was also reduced. Abe and Homma[42] have found decreased responsiveness to dinitrochlorobenzene and PPD in a group of 20 SLE patients of whom only 3 were receiving more than 10 mg of prednisolone daily. However, Goldman et al[47] observed normal skin test responses to PPD and to Candida, and almost all of these patients reacted adequately to phytohemagglutinin.

Whether there is definitely a diminution of delayed hypersensitivity in SLE is not clear on the basis of current information since available data are somewhat contradictory. The evidence is more cogent in the case of the NZB mice. The spleen cells of these animals were deficient in the induction of graft-versus-host disease[48,49], and in their response to mitogenic agents[50].

One further line of evidence argues for a deficiency of the delayed hypersensitivity mechanism in SLE. It has been suggested that the frequent development of lymphomas in old NZB mice is a consequence of a failure of the immunologic surveillance mechanism usually associated with the cellular immune response. In the case of the NZB mouse, this could be a failure either to reject the tumor or to control a murine lymphoma virus or both. There is some evidence that the SLE patient may also be prone to the development of lymphoma and leukemia, since at least seven instances of the occurrence of either of these conditions in SLE patients have been reported[52-55]. This may well signify an increased incidence since SLE has until recently been associated with a relatively short life span.

CONCLUSION

In attempting to integrate the data summarized above, the following features of SLE must be taken into account:

(1) Anti-DNA.

(2) Anti-double stranded RNA.

(3) Other autoantibodies.

(4) Endothelial cytoplasmic inclusions.

(5) Probable depression cellular immunity.

(6) Overall moderate elevation viral antibody titers.

It is also necessary to take into account the cogent evidence from studies of natural models indicating:

(1) Likelihood of a viral etiology for the diseases of NZB mice and Aleutian mink.

(2) Hyperreactivity to nucleic acid antigens in NZB-NZW mice.

(3) Depressed cellular immunity in NZB mice.

It appears reasonable to consider seriously the possibility that SLE patients do have a persistent viral infection. This, either through thymic inury (which has been suspected to occur naturally in SLE[56]) or through the appropriation of thymus-derived lymphocytes (which are believed to be necessary for antigen recognition) could lead to depression of cellular immunity. Depression of cellular immunity has been noted with LCM[57] and Gross[58] virus infection in mice (Fig. 1).

It is also reasonable to expect that persistent viral infection could have an adjuvant effect on the humoral antibody response, particularly to nucleic acid antigens[19]. LDH virus offers an example of a virus which may stimulate serum antibody formation[59,60] while it depresses cellular immunity.

Viral inflammation could produce the adjuvant effect proposed either by stimulating lymphoid proliferation or by making nucleic acid available through tissue breakdown. The adjuvant effects of

nucleic acids are well known[62]. X-irradiation[63] and endotoxin[64] are, in fact, believed to exert their stimulatory effects on the humoral response through the release of nucleic acids from injured tissues. The availability of DNA in such a milieu could then lead to the formation of anti-DNA (Fig. 2).

A viral adjuvant effect could also bring about the abrogation of tolerance to autoantigens which is a striking feature of SLE. Such abrogation of tolerance to autoantigens has been frequently observed in a number of experimental autoimmune diseases in which Freund's adjuvant has been administered with the autoantigen[65].

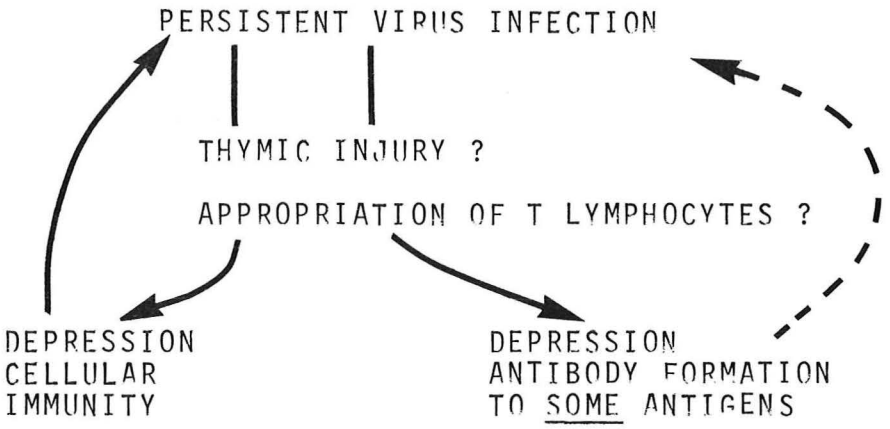
PERSISTENT VIRUS INFECTION
THYMIC INJURY ?
APPROPRIATION OF T LYMPHOCYTES ?
DEPRESSION
CELLULAR
IMMUNITY
DEPRESSION
ANTIBODY FORMATION
TO SOME ANTIGENS

Fig. 1

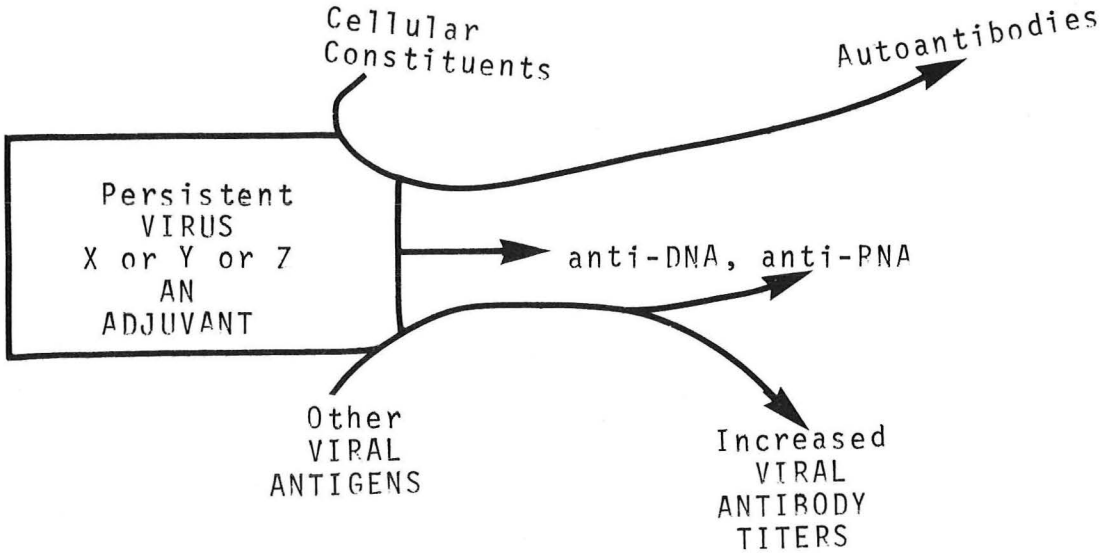
Cellular
Constituents
Autoantibodies
Persistent
VIRUS
X or Y or Z
AN
ADJUVANT
anti-DNA, anti-PNA
Other
VIRAL
ANTIGENS
Increased
VIRAL
ANTIBODY
TITERS

Fig. 2

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