baby hamster kidney cells infected with this virus (*11*). The possibility that the filamentous structures which make up the SSLE intranuclear inclusion may represent accumulations of the helical internal component of a virus should be considered.

The similarity in fine structure of SSLE intranuclear inclusions and the inclusions found in measles supports the concept that SSLE is a disease of virus etiology.

Note added in proof: Since submission of this manuscript, a report on one case of subacute encephalitis with intranuclear inclusions of identical ultrastructure has come to our attention (*12*).

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Paraproteinemia and Reticulum Cell Sarcoma in an Inbred Mouse Strain

Abstract. *Mice of the inbred SJL/J strain have a high incidence of a proliferative disease affecting several cell types, including reticulum cells and plasma cells, which is frequently accompanied by γ_1 and γ_2 paraproteinemia. In only some instances can serially transplantable lines of neoplastic cells be obtained; these are reticulum cell sarcomas. Mice with transplanted reticulum cell sarcomas do not have paraproteinemia and may develop profound hypogammaglobulinemia. The disease may be viewed as an abnormal proliferation of reticulum cells which differentiate into plasma cells with consequent paraproteinemia; the subsequent emergence of transplantable reticulum cell sarcoma appears as an end stage in which this capacity to differentiate is lost.*

In 1963 E. D. Murphy described an inbred mouse strain called SJL/J which exhibits a high incidence of reticulum cell sarcoma bearing some resemblance to Hodgkin's disease (*1*). Our SJL/J colony was established in 1964 (*2*) and has shown a high incidence of the characteristic disease—84 percent at an average age of 10 months in a group of 50 virgin female mice observed for at least 14 months. Although neoplasms of similar type have been described in mice (see *3*) they are infrequent. The disease differs markedly from the common form of leukemia seen in mice of strains with a high incidence of spontaneous leukemia (for example, AKR and C58) and in mice inoculated with Gross virus. Leukemia in mice of high-incidence strains is characterized by progressive enlargement of lymphatic tissue, origin from the thymus in most cases, a rapidly fatal course, and a uniform histological picture showing proliferation of lymphoblastic cells. These leukemias have the cellular antigens associated with Gross virus (*4*) and are invariably transplantable to isogenic mice. SJL/J disease also presents with enlargement of lymphatic tissue, but it differs from leukemia of high-incidence strains in all the other respects mentioned. It does not appear to arise from the thymus, the course of the disease is very protracted, the histological picture shows both neoplastic and inflammatory features, the antigens associated with Gross virus are absent, and the disease is not readily transplantable to isogenic hosts.

A further striking difference, which we report here, is that the SJL/J disease is often associated with abnormalities in immunoglobulins. The only neoplasm of the mouse that is accompanied by comparable changes in immunoglobulins is the plasma-cell myeloma (*5*) which rarely occurs spontaneously but can be readily induced in certain strains. However, paraproteins continue to be produced by transplanted myeloma cells, whereas in the case of SJL/J disease we find that paraproteinemia is confined to primary cases and is not seen in mice with transplants.

The SJL/J disease presents with local or generalized enlargement of lymph nodes, and splenomegaly, and may occur as early as 5 months of age. (Moderate enlargement of lymph nodes may be found in even younger mice, but it is not clear how this is related to the disease.) Mice remain in good condition for long periods with progressive and often massive involvement of affected organs; the average survival time of 17 mice that developed the disease while under observation was 14 weeks. Examination post mortem at various stages of the disease shows enlargement of lymph nodes (the cervical, mediastinal, or mesenteric nodes often being grossly involved before other nodes), splenomegaly, diffuse hepatomegaly, and very prominent Peyer's patches. The number of peripheral white cells is not increased, and there may be a decrease in circulating lymphocytes. There is no anemia, even in advanced cases.

The histological findings vary greatly from mouse to mouse and even from tissue to tissue in the same mouse. The most prominent feature in early cases is proliferation of plasma cells in lymphoid organs (Fig. 1a). Later, proliferation of reticulum cells is a feature common to all cases of the disease, ranging from scattered foci to general replacement of organs. The lymphoid follicles of the spleen are progressively replaced by proliferating reticulum cells, and similar foci are found in liver, lung, thymus, and lymph nodes. Other cell types that may be found in large numbers, but with considerable variation from tissue to tissue, are eosin-

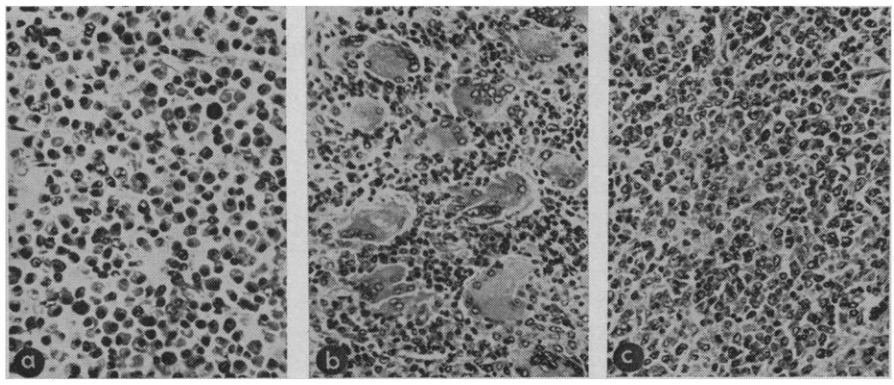

Fig. 1. (a) Accumulation of plasma cells in lymph node of SJL/J mouse with disease. (b) Giant cells in lymph node of SJL/J mouse with advanced disease. (c) Reticulum cell sarcoma in spleen of mouse with fifth passage of an established transplantable line (all × 130).

ophils, histocytes, Langhans' giant cells (Fig. 1b), and atypical giant cells (which appear to be distinguishable from megakaryocytes and occur at sites where megakaryocytes are not normally found in the mouse). Infiltration has not been observed in brain and heart, and occurs only infrequently in kidney. In mice with extensive disease there usually is no obvious involvement of bone marrow, except possibly an increased number of eosinophils; rarely, large numbers of plasma cells are present.

The neoplastic nature of the disease is established by the fact that serially transplantable cell lines can be obtained. However, transplantability is not an invariable characteristic of the primary disease—we have obtained only 6 lines from 16 advanced primary cases in which transplantation was attempted (20 to 85 × 10^6 viable cells from spleen and lymph nodes intraperitoneally). This frequent lack of transplantability distinguishes this neoplastic disease from all other spontaneous malignancies occurring with high incidence in inbred mice. It cannot be ascribed to histoincompatibility due to insufficient inbreeding because skin grafts exchanged among SJL/J mice of our colony are accepted. The diversity of cell types characteristic of the primary disease is greatly reduced in the transplantable lines, which are composed of reticulum cells (Fig. 1c) similar to those seen in the primary disease. Mice inoculated with cells from established lines develop enlargement of lymph nodes and spleen and live for 6 weeks or more with progressively severe disease.

Two leukemia-specific cellular antigens are commonly found in spontaneous leukemias of the mouse: (i) G (Gross) antigen, in all leukemias of strains with a high incidence of lymphatic leukemia (*4*), and (ii) TL (Thymus Leukemia) antigen, an antigen determined by the genome of leukemia cells (*6*). Neither is found in primary SJL/J disease or in transplanted SJL/J reticulum cell sarcomas. (Leukemias induced by Passage A Gross virus in SJL/J mice are of the usual lymphatic type and are freely transplantable.)

An outstanding feature of the disease is the occurrence of paraproteinemia in a high proportion of primary cases. A group of 31 normal virgin female SJL/J mice, aged 5 to 14 months at the beginning of the study, were bled at monthly intervals for a period of 3 months, with the object of detecting serum protein abnormalities preceding or following development of the disease. The serums were examined by cellulose-acetate electrophoresis (*7*) and by immunoelectrophoresis (*8*) with rabbit antiserum to mouse whole serum and goat antiserum to mouse γ-globulin (Hyland Laboratories) (Fig. 2). Of 24 mice that developed overt disease during the 3-month period, three showed no changes in serum proteins (throughout the entire course of the disease) and 21 showed abnormalities of immunoglobulins, which in 12 instances preceded the onset of overt disease. The two most common changes were (i) the occurrence of a fast-migrating γG component which on immunoelectrophoresis produced a distinctive double curvature of the γG line [Fig. 2, SJL/J No. 656 (early)], and (ii) a moderate to massive monoclonal increase in protein with slow γ to β mobility (myeloma-like pattern). Of the 21 mice that showed protein changes, 7 showed the anomalous fast γG, 11 had a myeloma-like pattern, and 3 had a combination of both changes.

The fast γG anomaly is the commonest early change and frequently precedes overt disease. The serums of mice with this anomaly usually show further changes later in the course of the disease. These include progression to a myeloma-like increase in paraprotein in the region of the anomalous fast γG (Fig. 2, SJL/J No. 695), or persistence of the anomalous fast γG with appearance of a myeloma-like increase in a different part of the γG region [Fig. 2, SJL/J No. 656 (late)]. The monoclonal changes tend to remain constant until the death of the animal, although sometimes the paraproteinemia diminishes,

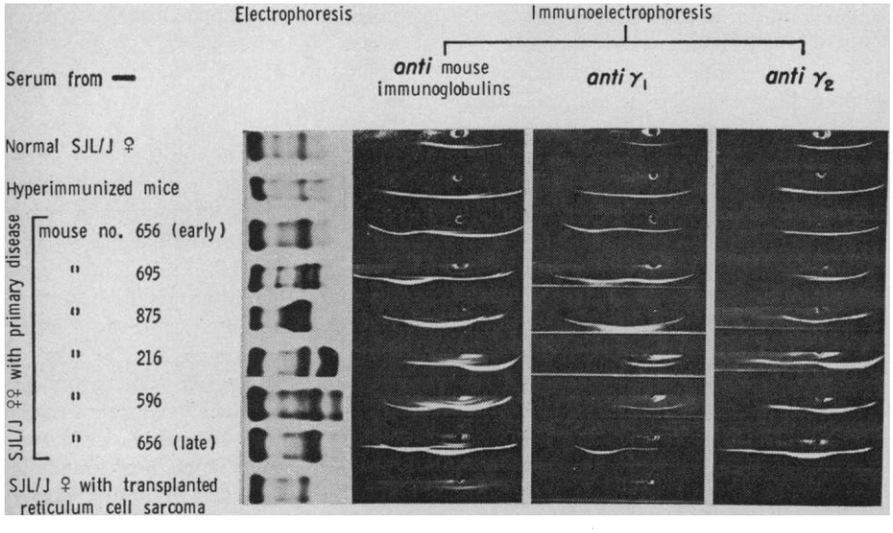

Fig. 2. Electrophoresis and immunoelectrophoresis of serum from normal SJL/J mice and from SJL/J mice with primary or transplanted disease.

Table 1. Typing of paraproteins of SJL/J mice with antiserums specific for γ_1 and γ_2 mouse immunoglobulins.

Serum protein anomaly	Distribution of γ^1 and γ^2	
	Mice without overt disease	Diseased mice
Anomalous fast γG component	4 γ_1; 1 γ_2	7 γ_1
Monoclonal paraprotein + anomalous fast γ_1	1 γ_2	3 γ_2
Monoclonal (myeloma-like) paraprotein	3 γ_1; 2 γ_2	13 γ_1; 6 γ_2
Diclonal paraproteins	1 γ_1 and γ_2	5 γ_1 and γ_2

occasionally to a point where abnormality is no longer apparent or hypogammaglobulinemia supervenes.

Serums from 46 SJL/J ♀♀ with discrete serum protein anomalies were typed with antiserums specific for γ_1, γ_2, γA and γM (provided by J. L. Fahey) (*9*). The results are summarized in Table 1, and examples are illustrated in Fig. 2. With one exception, the anomalous, fast-migrating γG typed as γ_1. There were 34 mice with myeloma-like paraproteins; 16 of these were γ_1 and 12 were γ_2; the other six mice each had two paraproteins, γ_1 and γ_2. No case showed two myeloma-like paraproteins of the *same* antigenic type. The fast-migrating γG component was associated with a separate γ_2 monoclonal increase in four cases but was not found associated with a separate γ_1 monoclonal increase; this supports the view that the fast γ component is an earlier stage of a γ_1 monoclonal increase. There was no instance of a myeloma-like increase in γA or γM (in contrast to the frequency of γA paraproteins produced by myelomas of the mouse). As a rule, mice with the isolated fast γG change showed an elevation in γA, whereas serums with a monoclonal increase of γ_1 or γ_2 showed a decrease in γA and γM. Urines from four mice with the anomalous fast γG and four mice with monoclonal increases were concentrated and tested for paraproteins. Only one showed an anomalous urinary protein; this was a fast-migrating γ_1 similar to that in the serum. Paraproteins are not found in mice bearing transplanted lines. However, these animals may develop marked hypogammaglobulinemia (Fig. 2). (No serum protein abnormalities were observed in 123 normal SJL/J mice aged 6 to 12 weeks.)

The lesions of this disease, with admixture of neoplastic and apparently reactive cells, prompts comparison with Hodgkin's disease, although the striking changes in serum proteins are not seen in Hodgkin's disease. With regard to etiology of the disease (*10*) we may suggest three general possibilities (which do not exclude initial causation by a virus), as follows.

First, it might be a polyclonal malignancy affecting various types of cells; the invariable emergence of only the reticulum cell in transplanted lines is against this interpretation.

Second, it might be a monoclonal malignancy of reticulum cells with an exuberant host reaction to the malignant cells or their products, which would explain the frequent lack of transplantability. However, transplanted lines do not elicit a proliferative response of plasma cells such as is seen in primary cases, and the serum protein abnormalities that are seen in primary cases take the form of discrete paraproteins suggestive of abnormal cellular proliferation rather than diffuse increases indicative of reactive hyperplasia.

Third, the disease might be basically a proliferative disorder of certain clones of reticulum cells that at an early stage have the capacity to differentiate into plasma cells; as the disease progresses their capacity to differentiate is lost, mature plasma cells and the accompanying paraproteinemia disappear, and finally, in many cases, an autonomous transplantable reticulum cell sarcoma is established. This is the possibility that we believe best fits our present knowledge of the disease.

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Substructure of Certain Cytoplasmic Microtubules: An Electron Microscopic Study

Abstract. *Negatively stained cytoplasmic microtubules of lung-fluke sperm show a helical structure that is not found in peripheral doublet tubules of axial units. In transverse sections, the wall of such microtubules appears to comprise about eight subunits.*

The structure, relationships to other cell organelles, and possible functions of cytoplasmic microtubules have received much attention since Slautterback's (*1*) discussion of their occurrence and relationships in hydra. My work deals with the substructure of microtubular elements in a spermatozoon as seen in both sectioned and negatively stained material.

Trematode worms (*Haematoloechus medioplexus*) were obtained from the lungs of *Rana pipiens*. Pieces of the body, containing the seminal receptacle or testes, were fixed in cold 6 percent glutaraldehyde (*2*) buffered with *s*-collidine (*3*) at *p*H 7.4 to 7.7. Material was embedded in Epon 812 (*4*); sections were cut with glass or diamond knives and stained with lead citrate (*5*), uranyl acetate, or both. Negative-staining was achieved with 1 percent sodium phosphotungstate (*6*). Sperm were obtained from seminal receptacles or testes teased in a drop of sodium phosphotungstate. Specimen screens, coated with Parlodion and a thin film of amorphous carbon, were then flooded with this fluid by means of a micropipette. Seminal receptacles or testes to be ultrasonically disrupted were collected in about 1 ml of sodium phosphotungstate and treated for 1 to 4 minutes with a Bronwill Biosonik generator at 75 percent maximum probe intensity (125 watts). Micrographs were made