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HTLV-III SEROLOGY DISTINGUISHES ATYPICAL AND ENDEMIC KAPOSI'S SARCOMA IN AFRICA

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Summary Serum samples from African patients with Kaposi's sarcoma and acquired-immunodeficiency-syndrome-related (AIDS-related) disorders and from normal subjects in Uganda and Zambia were tested for antibodies to the human T-lymphotropic retroviruses (HTLV) types I, II, and III. Nearly 90% of patients with AIDS-related disorders or with atypical, aggressive Kaposi's sarcoma were seropositive for HTLV-III in both countries, whereas only 17% of patients with classic endemic Kaposi's sarcoma were seropositive. Among the controls 20% were seropositive for HTLV-III in Uganda but only 2% in Zambia. None of the subjects tested had antibodies to HTLV-I or HTLV-II. These results are further evidence of the emergence of a clinically atypical form of Kaposi's sarcoma in Africans, which resembles that seen in American patients with AIDS, and which is associated with HTLV-III infection. The low frequency of antibodies to HTLV-III in the normal Zambian population together with the first appearance of HTLV-III-associated diseases during the past 2 years suggests that this virus is new to Zambia, although it may have been present in Uganda for longer.

Introduction

UNTIL a few years ago Kaposi's sarcoma was rare in North America and Europe but common in central Africa.[1,2] It typically affected older men and ran an indolent course, with many patients surviving for 10 or more years. Occasional cases were seen in children[3] and in renal transplant recipients;[4] in such cases the disease was more aggressive, with widespread involvement of body organs. Since 1981, increasing numbers of cases of Kaposi's sarcoma have been seen[5] in patients with the acquired immunodeficiency syndrome (AIDS); the clinical presentation is similar to childhood Kaposi's sarcoma. A human T-lymphotropic retrovirus, HTLV-III,[6,7] appears to be the causative agent of AIDS, on account of its isolation from many AIDS patients[7] and prevalence of specific antibodies in AIDS risk groups.[8-10]

One of the central questions about this virus is where it came from. Because of the high frequency of Kaposi's sarcoma in AIDS patients it has been suggested that central Africa, where the disorder is endemic, may be a good place to begin a search. AIDS has been reported from Zaire,[11] Rwanda,[12] and Zambia,[13] and cases seen as early as 1976[14] fit the AIDS case definition. Serological evidence indicates that AIDS patients in Zaire have antibodies to HTLV-III.[15,16] There is little evidence for homosexual activity among African AIDS patients and seropositive subjects. In Africa HTLV-III appears to be transmitted through heterosexual contact or exposure to blood through insect bites or scarification.

Biggar et al[16] found that, despite the endemic nature of HTLV-III in Zaire, patients with endemic Kaposi's sarcoma were consistently seronegative. One of us has reported an increase in an atypical form of Kaposi's sarcoma in Zambia[17] which had many features in common with the AIDS-related

HTLV-III ANTIBODIES AMONG AFRICAN SUBJECTS

Group	n	Sex (M/F)	Mean age in yr (range)	No (%) with HTLV-III antibody
Zambia				
AIDS-related disorders	15	7/8	26 (19–47)	13 (*87*)
Endemic KS	17	16/1	44 (16–67)	4 (*24*)
Atypical KS	22	17/5	29 (17–56)	20 (*91*)
Controls	158	26/132	35 (1–58)	3 (*2*)
Uganda				
AIDS-related disorders	4	3/1	21 (11–27)	4
Endemic KS	13	13/0	46 (34–67)	1 (*8*)
Atypical KS	4	3/1	31 (19–44)	4
Controls	51	31/20	27 (2–60)	10 (*20*)

KS = Kaposi's sarcoma.

form. We present here the results of assays for antibodies to HTLV types I, II, and III in these patients and patients from Uganda.

Methods

HTLV-I and HTLV-II antibodies were sought as described previously,[18] by competitive radioimmunoassay for HTLV-I tests and syncytium inhibition assay for both HTLV-I and HTLV-II. HTLV-III antibodies were sought by competitive radioimmunoassay,[10] with antigen inactivated with 0·25% beta-propiolactone before capture by antibody for solid-phase assay, and by membrane immunofluorescence.

Results

All the patients were negative for antibodies specific to HTLV-I and HTLV-II.

Endemic Kaposi's Sarcoma

We studied 30 patients with endemic Kaposi's sarcoma, 13 in Uganda and 17 in Zambia, including 3 originating from Kenya and 1 from Mozambique. 1 Ugandan patient and 4 Zambian patients were seropositive for HTLV-III (see table). Of the positive Zambian patients, 2 responded well to chemotherapy but the disease in another has became aggressive. The fourth patient was borderline for HTLV-III antibodies in 1984 and remains in complete remission: he presented in 1981 with endemic disease which regressed completely during treatment with indomethacin.

Atypical Kaposi's Sarcoma

22 of 26 patients with atypical disease were Zambian; the clinical features of 13 have been described previously.[17] Only 2 had no antibodies to HTLV-III when tested in 1984—a 19-year-old woman with lymphadenopathic disease who responded well to chemotherapy in 1983, and a man with many skin lesions in unusual sites who died shortly after admission to hospital. 11 patients in this group have died.

Among the 4 seropositive patients with atypical disease from Uganda 2 had disease of recent onset and 2 have died. In 1 who died, a 27-year-old woman who presented with gross lymphadenopathy and hepatosplenomegaly, Kaposi's sarcoma was confirmed in lymph nodes and 6 months later in her gastrointestinal tract. The other patient who died was a 44-year-old man with lymphadenopathy, severe weight loss, and persistent diarrhoea. Lymph-node biopsy samples showed Kaposi's sarcoma and within 3 months the lungs were involved and the liver and spleen were enlarged. Clinical and necropsy findings will be reported elsewhere.

AIDS-related Disorders

All 15 Zambian patients in this group had unexplained persistent general lymphadenopathy; some had other symptoms, such as diarrhoea, night sweats, weight loss, or fatigue. Lymph-node enlargement was strikingly symmetrical; biopsy samples showed non-specific reactive hyperplasia. Serial sections from some nodes were examined but none showed microscopic Kaposi's sarcoma. 3 of these patients presented in 1982 and 1983 and the rest were first seen in 1984. Among the 4 Ugandan patients, 2 had persistent general lymphadenopathy, 1 persistent diarrhoea, and in 1 *Escherichia coli* bacteraemia developed after blood transfusion. The latter 2 patients should not be definitively classified within the AIDS-related complex.

Controls

Only 3 of 158 serum samples from Zambian controls had antibodies to HTLV-III. All the positive controls were women. 1, a prostitute, had shared lodgings with another prostitute who died with aggressive HTLV-III-related Kaposi's sarcoma in 1983. The other seropositive control is a nurse working in a tumour ward (1 patient with persistent general lymphadenopathy is also a nurse). 123 samples were from recently delivered mothers (approximately equal numbers of livebirths and stillbirths); none had antibodies to HTLV-III. Male controls were healthy medical students, doctors, and laboratory staff.

20% of Ugandan controls were seropositive for HTLV-III, although most had low titres. These control subjects included patients with various disorders as well as healthy students and doctors; positive sera were found among patients and healthy subjects.

Discussion

Although the endemic form of Kaposi's sarcoma has long been known in Zambia, the atypical aggressive form first appeared in 1983, and persistent general lymphadenopathy was first seen at about the same time.[17] Our serological data show that persistent general lymphadenopathy and the atypical form of Kaposi's sarcoma are strongly associated with HTLV-III infection. The serological tests[10] are highly specific for HTLV-III, and only antibodies to closely related retroviruses would be expected to give a crossreacting signal. None of the samples tested in this survey had specific antibodies against HTLV-I or HTLV-II.

The Zambian results are supported by those for Ugandan Kaposi's sarcoma patients; atypical Kaposi's sarcoma was also rare in Uganda until recently. For example, of 96 adult cases seen in Kampala between 1956 and 1962, only 5 would be regarded as atypical[19] and a study of the disorder in the West Nile District between 1951 and 1976 found only typical, endemic cases.[20] Atypical disease may always have been associated with HTLV-III infection. A retrospective study indicates that HTLV-III has been present in Uganda for at least 10 years (G. de Thé, personal communication). The low prevalence of antibodies to HTLV-III in the normal controls in Zambia indicates that the virus is not yet widespread in that country, but it, or a related virus, appears to be endemic in Uganda, occurring in 20% of our small control sample.

There is some overlap in clinical presentation of atypical and endemic Kaposi's sarcoma, with histological identity,

which might explain why 4 of 17 Zambian patients classed as endemic cases were HTLV-III positive. Despite the differences in clinical presentation, the two forms of Kaposi's sarcoma seen in Africa may have the same underlying aetiology. HTLV-III infection may act primarily by inducing a degree of immunosuppression that allows Kaposi's sarcoma, possibly caused by a separate agent, to develop with the atypical, aggressive course, as in transplant recipients who have been chemically immunosuppressed.[4] By analogy, Epstein-Barr-virus-positive lymphomas occur most frequently in regions of holoendemic malaria infestation and in immunosuppressed transplant patients.

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INCREASED EXPRESSION OF HLA ABC CLASS I ANTIGENS BY MUSCLE FIBRES IN DUCHENNE MUSCULAR DYSTROPHY, INFLAMMATORY MYOPATHY, AND OTHER NEUROMUSCULAR DISORDERS

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Summary The distribution of HLA ABC class I antigens in human skeletal muscle obtained by needle biopsy was investigated by means of a monoclonal antibody (W6/32) and an immunoperoxidase technique. Five samples from normal individuals and twenty-nine from patients with various neuromuscular disorders were examined. Normal muscle fibres and those from patients with congenital muscular dystrophy expressed little or no class I antigens, whereas muscle fibres of patients with myositis and various X-linked muscular dystrophies showed consistently strong expression. In other neuromuscular diseases expression was more variable. The presence of class I antigens on diseased muscle fibres may render them susceptible to cytotoxic T cells; these antigens may thus have an important role in the destruction of muscle fibres.

Introduction

HUMAN class I (HLA A,B, and C) antigens are integral membrane glycoproteins composed of a highly polymorphic chain (molecular weight 45 000) non-covalently associated through its extracellular portion with β_2-microglobulin, a non-polymorphic, non-glycosylated polypeptide of molecular weight 12 000.[1] The large chain is encoded by the genes of the major histocompatibility complex (MHC) on the short arm of chromosome 6;[2] the β_2-microglobulin gene is on chromosome 15.[3]

Results of cytotoxic assays of single-cell suspensions or absorption assays of alloantisera or heteroantisera suggested that all nucleated cells express class I antigens.[4] However, such assays are inadequate for the characterisation of antigens within organs, and it has become possible to define the distribution of tissue antigens in situ only with the introduction of frozen tissue sections, monoclonal antibodies, and enzyme-linked secondary antibodies. With this combination of techniques, the class I antigen distribution in various human tissues taken from one donor at necropsy has been surveyed. Although the antigens were expressed by most nucleated cell types, they were absent or expressed only at low levels in several tissues,[5] including both skeletal and cardiac muscle. Smooth muscle showed strong expression of class I antigens.

We have investigated the distribution of the non-polymorphic class I determinant detected by the monoclonal antibody W6/32 in human skeletal muscle in health and in various neuromuscular disorders.

Patients and Methods

Specimens of muscle tissue were obtained by needle biopsy of the vastus lateralis from patients attending the paediatric muscle clinic at Hammersmith Hospital. Diagnosis was established by clinical criteria, serum enzymes, histochemistry, and electronmicroscopy.[6] As controls we used muscle biopsy samples that showed no evidence of any neuromuscular disease.

Muscle samples were rapidly frozen in 'Arcton 12' (ICI, Cheshire) cooled with liquid nitrogen. Unfixed, transverse cryostat sections (10 μm) were mounted on glass coverslips and incubated for