Arch Virol (1987) 92: 183–186

Archives of Virology

Simultaneous Isolation of Simian Foamy Virus and HTLV-III/LAV from Chimpanzee Lymphocytes Following HTLV-III or LAV Inoculation

Rapid Communication

By

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With 1 Figure

Accepted October 20, 1986

Summary

Re-isolation of virus from HTLV-III B and LAV-infected chimpanzee also yielded a simian foamy virus. This virus, replicated in HTLV-III B and LAV-producing H 9 cells, had identical reverse transcriptase activity and caused similar cytopathic effects in H 9 cells.

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Many lower primates, as well as chimpanzees, have been inoculated with infectious HTLV-III or LAV, recently named human immunodeficiency virus (HIV) (8). Chimpanzees are the only experimental animals thus far demonstrated to be consistently susceptible to infection with the etiological agent of AIDS, HTLV-III/LAV (2, 4, 5) a retrovirus closely related to the subfamily Lentivirinae.Chimpanzees experimentally infected with HTLV-III/LAV become persistently infected, which is analogous to what happens in infected humans. Although infection in man can be asymptomatic or can result in AIDS, infected chimpanzees at most display transient symptoms

but have not yet developed AIDS. Chimpanzees will be of importance for tests of candidate vaccines to prevent AIDS. Investigators should be aware that primates have several indigenous retroviruses, the presence of which may complicate the assessment of isolation of HTLV-III/LAV.

Attempted re-isolations for infectious virus are commonly monitored by the reverse transcriptase assay (RT). However, other confirmatory assays are required to confirm the presence of HTLV-III/LAV and/or other RT-positive retroviruses. During re-isolation of retroviruses from HTLV-III- or LAV-inoculated chimpanzees, we obtained RT activity in three out of three primary cultures of Ficoll gradient-purified chimpanzee lymphocytes cocultivated with H 9 cells. The first rise in RT levels detected at the end of six weeks in culture was associated with the presence of specific HTLV-III/LAV envelope and core proteins. These were identified by radioimmunoprecipitation polyacrylamide gel electrophoresis (RIP-PAGE), competitive p 24 radioimmunoassay, and two-dimensional gp 120 peptide maps of the gp 120's. Examination by electron microscopy revealed that two of the three cultures were producing not only HTLV-III/LAV, but also a chimpanzee foamy virus, which is morphologically distinct (see Fig. 1 A). Furthermore, Fig. 1 B demonstrates, to our knowledge, the first report of these two distinct retroviruses, Lentivirus-like (HTLV-III/LAV) and Spumavirus (primate foamy virus), being produced by the same cell. Further characterization of this foamy virus was accomplished by the formation of large, vacuolated, syncytial cells following infection of a human glial cell line 808 D* with cell-free supernate from the suspect H 9 cultures. The dually infected H 9 cell line is currently stable and produces both RNA viruses effectively at eight months post co-cultivation.

Foamy viruses are members of the family Retroviridae, subfamily Spumavirinae. They possess a ribonucleic acid (RNA)-dependent deoxyribonucleic acid (DNA) polymerase, which like HTLV-III/LAV prefer magnesium over manganese. They are present in a wide variety of animals, including primates and man (1, 9), are considered latent and induce persistent infections in their host (6, 1, 9). Although foamy viruses can be isolated from multiple tissues, white cells from the buffy coat were found to be a consistent source of productive virus cell cultures (7).

Based on the above, it appears reasonable that RT and initial cytopathology (these foamy viruses exhibit comparable cytopathic effects in H 9's as HTLV-III) be used only as a preliminary screening device in the isolation of any simian or human immunodeficiency viruses. Additional confirmatory steps should include inoculation of cell-free tissue culture medium

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* This cell line is a subclone, and derived from a human glioma (H 80) graciously provided to us by Dr. M. Cloyd, Duke University Medical Center, Durham, North Carolina 27710.

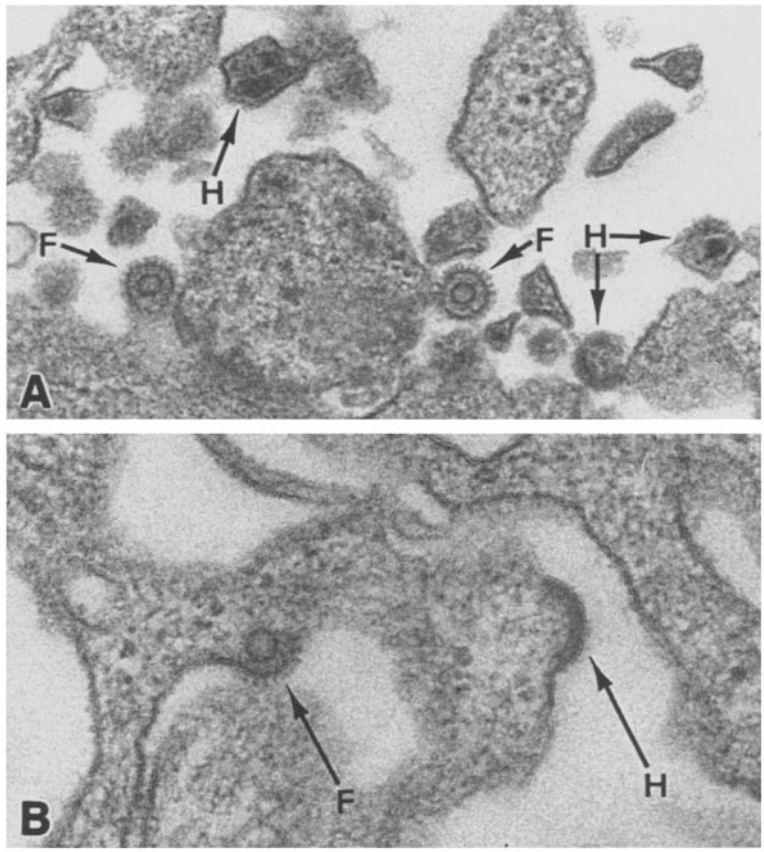

Fig. 1. Transmission electron micrograph of co-cultivated H 9 cells persistently infected with HTLV-III *(H)* and chimpanzee foamy virus *(F)*. *A* Thin section of H 9 cells showing mature virions of both HTLV-III and chimpanzee foamy virus (66,000 ×). *B* Thin section of a single cell depicting typical budding of HTLV-III and chimpanzee foamy virus (90,000 ×)

with non-lymphoid cell lines to observe the cytopathic effects of the foamy virus, usually seen in 7–10 days. Second, electron microscopy for characteristic morphological features such as complete, intra-cytoplasmically located central electron-lucent core with large (5–15 nm) radiating brush-like spikes, knobs, or projections, would identify a Spumavirus (3). Third, RT positive co-cultivations require immunological confirmations of the presence of specific viral structural antigens by RIP-PAGE, radioimmunoassay p 24 analysis, or other assays. Fourth, the above parameters should be repeated in these cultures prior to experimental use because these cell lines may cycle in their production of the foamy virus. Purification schemes for HTLV-III from chimp cells dually infected may include limiting end-point dilutions in the presence of homologous neutralizing foamy virus serum. This can be obtained from the lymphocyte donor animal because the humoral

response of the host to these viruses appears to be quite type-specific, and no family-specific, (6, 7, 10) broadly-reactive sera are available. This has been the case with foamy viruses of chimpanzees and other primates in which a persistently high-titered, specific, neutralizing antiserum exists (6, 9). In anticipation of the chimpanzee being utilized in pathobiologic studies and vaccine development, we suggest that current observations should increase the awareness for potential infection with several retroviruses.

Acknowledgements

We appreciate the expert scientific and technical assistance of Kunio Nagashima and Beth Vessey of the Laboratory of Cell and Molecular Structure, Program Resources, Inc., and Nancy Dunlop, Virus Control Section, NCI–Frederick Cancer Research Facility, P.O. Box B, Frederick, Maryland 21701. Research sponsored, at least in part by the National Cancer Institute, DHHS, under contract number NO1-CO-23910 with Program Resources, Inc. The contents of this publication do not necessarily reflect the views or policies of the DHHS, nor does mention of trade names, commercial products, or organizations imply endorsement by the U.S. Government.

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Received October 9, 1986