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Detection of human immunodeficiency virus (HIV) infection in formalin-fixed, paraffin-embedded tissues by DNA amplification

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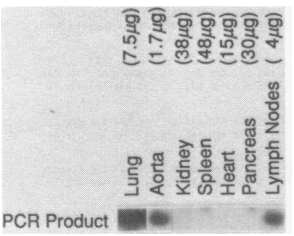
Detection of HIV provirus in blood samples from AIDS patients (1) and in infected cells (2) has been performed by amplification of target DNA using the polymerase chain reaction (PCR) technique (3). We report here that DNA extracted from formalin-fixed, paraffin-embedded tissue is suitable as a template for the detection of HIV provirus using PCR. These procedures offer several advantages: I) formalin fixation and paraffin embedding render the tissue non-infectious; II) routinely obtained biopsy and postmortem material can be studied; III) the epidemiology of the virus can be further elucidated via study of archival tissue obtained prior to the identification and isolation of the virus.

Ten tissue sections (5 μ each) were placed in anylon tissue biopsy bag (Shandon, Inc.) and deparaffinized in xylene for one hour. The tissue samples were then rehydrated and suspended in a minimum volume of TE buffer. Protease K and Sarkosyl were added to a final concentration of 1 mg/ml and 0.2%, respectively. The digestion was carried out at 50 °C overnight. The samples were extracted with phenol, followed by phenol/chloroform, and the DNA was precipitated with ethanol. The DNA was resuspended in 50 μl of TE buffer and the concentration was measured by fluorometer. Twenty-five cycles of PCR were performed using SK 38/39 and SK 68/69 primers (1) in a Perkin Elmer-Cetus DNA thermal cycler according to the manufacturer's recommended conditions. Twenty 1 of each amplified sample were run on a 2% agarose gel and HIV-specific sequences were detected by Southern blot hybridization using ^{32}P labelled oligomers SK19 and SK70.



Tissue sections from an AIDS-Related Complex (ARC) patient were examined. The amount of DNA extracted varied 25-fold (Figure). HIV-specific products were detected in lung, aorta and lymph node and were not related to the amount of template DNA used in PCR. This methodology should be adaptable as a routine non-infectious screening for HIV in tissue biopsies or blood samples (as formalin-fixed cell buttons).

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References

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