screening dried blood samples on filter paper for human immunodeficiency virus type 1 DNA.

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Abstract

PCR is a highly sensitive method for the detection of human immunodeficiency virus type 1 (HIV-1) nucleic acids in blood mononuclear cells and plasma. However, blood separation techniques require extensive laboratory support systems and are difficult when a limited volume of blood is available, which is often the case for infants. The use of blood samples stored on filter paper has many advantages for the detection of perinatal HIV-1 infection, but current methods require extraction and purification of target DNA prior to PCR amplification. We report a highly sensitive and rapid method for the extraction and detection of HIV-1 DNA in infant blood samples stored on filter papers. Because this rapid protocol does not involve steps for the removal of potential inhibitors of the PCR, the highest sensitivity is achieved by testing the filter paper lysate in quadruplicate. Assays for HIV-1 DNA were done by using nested PCR techniques that amplify HIV-1 gag DNA from blood spot samples on filter paper and from corresponding viably frozen mononuclear cells separated from venous blood samples obtained from 111 infants born to HIV-1-seropositive mothers. PCR results with blood from filter papers showed 100% specificity (95% confidence internal [CI] 93.1 to 100%) and 96% (95% CI, 88.65 to 98.9%) and 88% (95% CI, 79.2 to 94.5%) sensitivity (for quadruplicate and duplicate tests, respectively) compared to PCR results with blood mononuclear cells. Moreover, this method could detect HIV-1 sequences of multiple subtypes.