EFFICACY OF DRIED BLOOD SPOT IN EARLY INFANT DIAGNOSIS OF HIV INFECTION

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ABSTRACT

Introduction: *Nucleic acid analysis has been the most reliable diagnostic technique for detecting HIV infection in the neonates because of the presence of maternal antibodies in infants less than 6 months. As this is an expensive test, Direct Blood spot was used to detect HIV infection in early infanthood to prove the technical and economical advantage over the whole blood assays.*
Methodology: *115 infants of HIV seropositive mothers were subjected to DBS test and subsequently to Antibody and whole blood PCR analysis. The results of the DBS sample titres were compared with the standard whole blood analysis reports.*
Results: *Of the 115 infants who underwent the diagnostic tests, 10 children were found to be HIV positive of which 50%were found positive with the HIV antibody tests and the rest were DBS positive. The sensitivity and specificity of DBS was found to be 100% compared to the whole blood PCR analysis.*
Conclusion: *DBS is an ideal choice as a screening tool for detecting HIV infection in early infant hood.*

INTRODUCTION

HIV infection in infants occurs due to the transmission from infected mothers either during pregnancy, delivery or during lactation. Early detection of HIV in such infants will go a long way in managing and preventing them from serious life threatening outcomes. Since the serological diagnostic methods have drawbacks like; they cannot differentiate between maternal and infant antibodies, early detection is not possible and they have a very low sensitivity and specificity. Virological diagnostic methods like PCR are more reliable and help in early detection of HIV infection. While plasma viral load monitoring is widely available in high-income settings, it is rarely used in resource-limited regions because of high cost and need for sophisticated sample transport. Dried blood spot (DBS) as source specimens for viral load measurement has shown promise and has also been recommended by WHO, as an alternative to plasma specimens and is likely to be a useful tool in resource poor settings[1]. So the present study was conducted to compare DBS sampling to the whole blood viral load assay for early detection of HIV in infants with HIV infected mothers.

AIMS AND OBJECTIVES

To identify the HIV status in infants born to HIV seropositive mothers and to compare DBS with the standard viral load assay.

METHODOLOGY

All the infants of HIV seropositive mothers to the ICTC centre were included in the study. Informed consent was taken and DBS test was done at 6 weeks of all the infants and the test was repeated using whole blood for PCR analysis if DBS was positive. A total of 115 infants underwent sampling for DBS. Samples collected from pin prick was coated onto filter paper (Whatman No. 903). The spotted filter papers were allowed to dry for at least 4 hours at room temperature and placed in individual zip locked bags containing a silica desiccant. All these samples were transported to State referral centre for viral assay. The Positive infant samples were repeated with another whole blood collected in EDTA vaccutainer tubes for PCR.

If DBS was negative, another whole blood sample was taken to confirm with PCR. In infants above 6 months, rapid Ab test was done and if found positive they were subjected to DBS as well as PCR analysis. At 18 months all patients

underwent HIV Ab tests, using three rapid HIV antibody assays at ICTC

RESULTS

115 children of HIV positive mothers were tested for HIV between February 2009 and January 2014, of these 87(75.6%) children were of less than 6 months age, and underwent DBS test. The remaining 28(24.4%) children under went HIV Ab test because they were more than 6 months old. Totally 10(8.6%) children were found to be HIV positive of which 50%were antibody positive and the rest were DBS positive. 17 children came at 18 months for confirmatory test. 15 (88.25%) were negative. 2 (11.76%) were positive. Of the 2 positive at 18 months, one (5.8%) had undergone DBS test (<6months) and one (5.8%) (>6 months) had undergone Ab test. Of the 15 who were negative at 18 months, 6 (40%) had undergone DBS and 9 (60%) had undergone Ab test. **(Figure 1)**

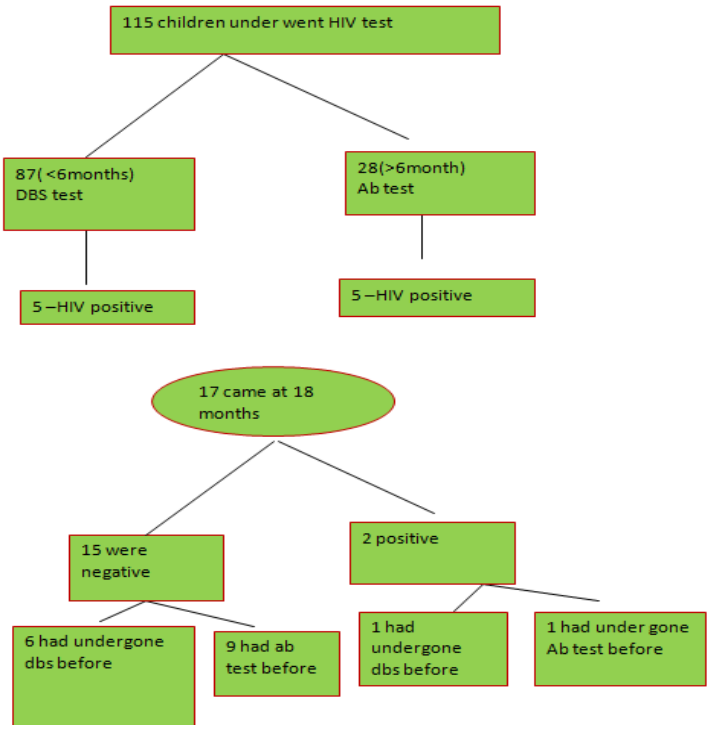

Figure 1: Summary of the Results of the study

The sensitivity and specificity of DBS was found to be 100% compared to the whole blood PCR analysis.

DISCUSSION

The use of dried blood spot specimens as a source for diagnostic tests has become increasingly popular in recent years. DBS has been used to identify genetic and metabolic disorders in neonates[2], detection of HIV-1 antibody[3] and HIV-1 DNA for infant diagnosis of HIV infection[4]. The WHO has recommended the use of DBS for HIV drug resistance (HIV DR) surveillance for monitoring transmitted drug resistance in resource-limited settings[2]. The reasons for popularity of DBS as a method of sample collection storage and analysis can be understood when one considers the numerous advantages with this method.

This format greatly facilitates the logistics of sample collection, processing, shipping and is cost effective. Whole blood saved as DBS can be transported or mailed to reference laboratories without refrigeration and has low biohazard risk[5]. Blood collection is easy for the phlebotomist and other healthcare workers; and no formal training is required .Whole-blood

can easily be coated on the filter paper from heel stick or finger punctures in infants; thus avoiding the use of syringes and vaccutainer tubes which adds to the biomedical waste. A small quantity of blood, i.e., 50 µl, is enough to make a dried blood spot (DBS) whereas for the whole blood 500 µl is required to perform the test. Blood is coated on filter paper which in turn lyses the cells and binds the DNA. Therefore, the sample centrifugation and extraction procedures are reduced. It appears biologically stable and can be stored at room temperature. It can be transported easily and therefore it is convenient to use the DBS in resource-limited settings[5].

Conclusion

DBS as a sample source is the best method to perform the diagnosis of HIV infection in infants born to HIV seropositive mothers. .

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