INTRODUCTION

HIV continues to be a major global public health issue. In 2018, an estimated 37.9 million people were living with HIV, with a global HIV prevalence of 0.8% among adults. Around 21% of these same people do not know that they have the virus. Many of the problems related to human immunodeficiency virus (HIV) infection and AIDS, particularly those affecting public health policies, have not yet been fully realized, because the total number of infected individuals in many parts of the world is still rising. According to National Strategic Plan (NSP) for HIV/AIDS and STI 2017 – 2024, one important focus of the national programme will be on achieving the 90-90-90 goal (i.e.) 90% of those who are HIV positive in the country know their status, 90% of those who know their status are on treatment and 90% of those who are on treatment experience effective viral load suppression.

The early detection assumes paramount importance at the present scenario. The Centers for Disease Control and Prevention (CDC) in 2006 recommends that diagnostic HIV testing and opt out HIV screening be apart of routine clinical care in all health care settings, while maintaining the individual concerned for not opting the testing for optimal clinical and preventive care. Among the unresolved issues surrounding HIV infection are those related to diagnosis: costs for the majority of countries most heavily affected, logistic problems associated with traditional methods, and use of specimens obtained by invasive procedures.

Saliva have been used as a diagnostic tool to investigate cystic fibrosis, in protein analysis, and it can also be used successfully to detect antibodies to a variety of viral agents including hepatitis A virus, hepatitis B virus and rubella virus. Currently, there is considerable interest in the use of oral fluids for HIV antibody testing. The use of oral fluids for HIV testing offers several advantages over that of blood. Most importantly, sample collection is safer since occupational risk from needles, disposal of needles, and cuts from broken glass tubes are eliminated. Disposal of wastes is an important consideration in

Background: HIV continues to be a major global public health issue. In 2018, out of an estimated 37.9 million people living with HIV, 21% of the same people do not know that they have the virus. The early detection of HIV infection assumes paramount importance at the present scenario. Preference for less invasive specimen collection techniques is always high (e.g., by use of saliva). It has now been shown that antibodies to HIV from the oral cavity can be detected with a sensitivity and specificity that are essentially identical to those of tests with serum (Sensitivity- 99.21%, Specificity-100%). The aim of our study was to correlate the presence of salivary anti-HIV-I and II antibodies with serum anti-HIV I and II antibodies in adolescents diagnosed with HIV infection and under antiretroviral therapy.

Methods: A cross sectional study was conducted with a convenient sample of 20 HIV-positive adolescents from an institutionalized setup. Salivary antibody presence assessed using HIV Oral Mucosal Transudate HIV antibody detection kit was correlated with serum antibody presence assessed using the gold standard serum antibody test result.

Results: The mean age of the 20 participants was 15.75±2.14. Salivary antibody presence had a strong positive correlation of 1.0 with serum antibody presence but was not statistically significant.

Conclusions: Emergency care providers are most commonly prone for infections from contact with patient’s blood. Thus, incorporating this simple to perform test kit, using noninvasive means, can greatly reduce the risk of transmitting the infection. This test kit is an effective screening device of HIV antibody.

Key Words:
HIV, ART, SCREENING.
countries where incineration or autoclaving facilities are not available and where waste materials are buried or placed in open refuse areas. With oral fluids the disposal risk is minimized since a single collection device, usually made from an absorbent material, can be discarded with greater safety than a blood collection tube and needle.4

Preference for less invasive specimen collection techniques is always high (e.g., by use of saliva). It has now been shown that antibodies to HIV from the oral cavity can be detected with a sensitivity and specificity that are essentially identical to those of tests with serum (Sensitivity- 99.21%, Specificity-100%). The use of saliva in reference methods has now become equally feasible, when such protocols are appropriately modified.5

The majority of potent and broadly neutralizing antibodies against HIV have been isolated from untreated patients with acute or chronic infection.6 However, a recent study reported high antibody levels with modest neutralization when ART was initiated several years after established chronic infection;7 thus, raising the possibility that HIV specific immune responses evolve over time on ART. There could be continuous antigenic stimulation or persistent immune dysfunction and antibody production even when virus is controlled with ART.8

Thus, the aim of our study was to correlate the presence of salivary anti-HIV I and II antibodies with serum anti-HIV I and II antibodies in adolescents diagnosed with HIV infection and under antiretroviral therapy.

MATERIALS AND METHODS

Ethical clearance

This cross-sectional study was conducted following approval by the Ethical Committee, Ragas Dental College and Hospital, Chennai. The general guidelines to ensure the rights of participants were followed. Before the investigation, parental/guardian consent was obtained, and the study information was reaffirmed orally.

Participants

The sample size was calculated according to previously reported data, suggesting an overall HIV prevalence rate of about 0.2% in the Indian general population, using a power of 80%. G power (version 3.1) software was used for sample size calculation.

It was estimated that at least 20 patients would be required. A convenient sampling was used to include 20 HIV-positive adolescents from an institutionalized setup, at Chennai.

The sample was diverse in nature pertaining to gender, ethnicity, sexual orientation, socioeconomic status, and education.

Inclusion criteria

1. Adolescents diagnosed with HIV infection with vertical transmission as the route of infection, and under HAART, at least for the past three years with CD4 count >200 cell/mm³, who never had a clinical AIDS-defining symptom.

2. Adolescents who agreed to participate, and whose parents / guardian give consent. Consent was also obtained from the Head of the Institution.

Exclusion criteria

1. Adolescents with concurrent diseases or medical conditions, oral pathologies, non-HIV viral infections.
2. Brushing of teeth, use of mouthwash or smoking tobacco immediately prior to testing.
3. Concomitant medications, and food or drink immediately prior to testing.
4. Chronic debilitating conditions or mental health disorders that would preclude informed consent.

MATERIALS

HIV (1/2) Oral Mucosal Transudate HIV antibody detection kit (Voyage medical co., Ltd, China)

The HIV-1/2 Oral Mucosal Transudate (OMT) test is a single-use, qualitative, noticeable results, in vitro- immunoassay for the detection of Human Immunodeficiency Virus (HIV) Type 1 and Type 2 antibodies in human OMT specimens. It is intended to use as a point-of-care aid in the clinical diagnosis of HIV infection.

Principle

It’s based on the principle of colloidal gold lateral chromatography. The qualitative test results (negative/positive) can be interpreted 20 minutes after the test strip placed in the diluted specimen.

The test strip contains test zone and control zone. Recombinant proteins representing the immunodominant regions of the HIV-1 gp41 and HIV-2 gp36 transmembrane proteins are immobilized onto the nitrocellulose membrane in the Test Zone. Goat anti-human IgGF (ab) 2 fragment polyclonal antibody is immobilized in the Control Zone. Colloidal gold-labelled proteins are immobilized onto the Colloidal Gold Cushion.

Data collection was done in two phases for HIV-infected children. In the first phase, participants’ ELISA test results for HIV using serum samples (gold standard HIV-screening test) was verified and recorded. The demographic data and CD4 level of each participant for the past 6 months were collected retrospectively from their respective medical records which served as the secondary data. In the second phase, ELISA test using HIV (1/2) Oral Mucosal Transudate HIV antibody detection kit (Voyage medical co., Ltd, China) was done for the same participants whose serum results were recorded, which served as the primary data. The patient’s preference for both blood and saliva tests were also recorded.

Method

The subjects was first instructed about the test and collection of samples.

To perform the assay, an oral fluid specimen collection swab was wiped across the upper and lower gums of the tested individuals, and this swab was then placed in the tube of oral fluid sample buffer and mixed. The liquid in the swab was
squeezed out and the used swab was discarded. The test strip was then placed vertically into the test tube containing the specimen/buffer mixture. The diluted specimen migrated up the assay test strip. The specimen/conjugate mixture continued to migrate up the strip, and first reached the Test Zone which contained the HIV antigens. If the specimen contained antibodies to HIV, the IgG/conjugate complex will bind to the antigen and become immobilized at the antigen line in the Test Zone and a reddish colored line appears (Positive reaction); The lack of a reddish colored line in the Test Zone indicates that the specimen does not contain HIV antibodies (Negative reaction). A reddish control line will appear in the Control Zone regardless of the sample is reactive or negative for antibodies to HIV-1 or HIV-2. The presence of the control line was a quality and performance control ensuring the test was performed properly.

The test was carried out

Test result and interpretation

1. Positive: If both test and control lines appear, whether the lines are clear or not, this test result is interpreted as a preliminary positive for HIV antibodies. One of these lines may be darker than the other. (The intensity of the test line in a positive result does not necessarily correlate with the amount of anti-HIV antibody in the specimen.)
2. Negative: Only the red control line appears. The test line does not appear. This test result is negative.
3. Invalid: Only test line appears but no control line is present. Neither test line nor control line appears. Absence of control line indicates either the assay procedure is not correct or the test kit expired.

Statistical analysis

Data collection and management were conducted using the Microsoft Office Excel package in association with the SPSS 20.0 software package (SPSS Inc.) for the statistical analysis. Qualitative data were presented as frequency and percentages. Correlation between variables for salivary and serum antibody presence were characterized by Spearman correlation analysis.

RESULT

A total of 20 children, aged 12–18 years of age diagnosed with HIV and under HAART participated in this study. The meanage of the study population with both males and females was 15.75 ± 2.14 years of age. The characteristics of the sample are presented in table I.

<table>
<thead>
<tr>
<th>CHARACTERISTIC</th>
<th>n (20)</th>
<th>Participants (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of participants</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>8</td>
<td>40</td>
</tr>
<tr>
<td>Female</td>
<td>12</td>
<td>60</td>
</tr>
<tr>
<td>Antibody presence</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Serum</td>
<td>20</td>
<td>100</td>
</tr>
<tr>
<td>Saliva</td>
<td>19</td>
<td>95</td>
</tr>
</tbody>
</table>

As presented in Table II, salivary antibody presence had a strong positive correlation of 1.0 with serum antibody presence but was not statistically significant.

All of 20 (100%) patients reported to be convenient when tested using oral fluid when compared to 4 (20%) patients who were apprehensive with the blood test involving the needle prick. Thus, there is 100% acceptability of HIV-test using oralfluid. The saliva test results were obtained within 15to20minutes, while the serum test results were available within few hours.None of the test results showed ‘invalid’ results. Only one of the saliva sample had shown false-negative result when compared with the serum test.

DISCUSSION

Technological advances in the diagnosis of HIV infection provide the clinicians with greater opportunities to reduce HIV transmission rates. It is estimated that increased awareness of serological status will decrease the number of new infections mainly by behavior modification of HIV-positive people and treatment that decreases viral loading ininfected individuals. Earlier, only traditional ELISA wa savable for detection of anti-HIV antibodies,serum. Overtimethere have been improvements of performance of ELISAs from usage of viral lysates to usage of recombinant antigens, synthetic peptidesor combination of these both. The ELISA more recently has been employed to detect HIV antibodies in whole blood, urine and salivary samples too. The Western blot remains the gold standard in HIV antibody confirmation.

Rapid point of care HIV tests introduced by FDA since 1992, greatly aids in knowing sero-status by providing faster and accurate results in minutes. This can be used in are as with limited laboratory resources, requires little training andthey provide convenience in testing on site results by allowing the delivery of definite negative or provisional reactive results greatly enhancing interventional programs. These rapid HIV tests can be done using both oral fluid and whole blood. The use of oral fluids such as saliva for detecting antibodies to HIV has been suggested as an alternative to the use of blood.

Oral rapid point of care HIV tests score over blood tests in their quality, rapidity, convenience, ease of sample collection and
feasibility. The efficacy of these tests has been evaluated individually and also by comparing both the tests. A study was conducted by Cheingsong-Popov R et al. (1993) on individuals from London and the United Republic of Tanzania, in which the use of oral samples obtained using a collection device was evaluated for the detection of antibodies to various synthetic peptides of the HIV. The reactive antibodies were of the IgG class and were detected in whole saliva suggesting that the source of antibodies in saliva is plasma transudation. In all cases, antibodies were detected in oral fluid; the titres paralleled those in serum. These results suggest that oral fluid is a potential specimen source for the measurement of the humoral response to the infection, and therefore may be applicable for the monitoring of antibody responses in those infected with HIV.

Similarly, in our study, presence of salivary anti-HIV I and II antibodies positively correlated with serum anti-HIV I and II antibodies in adolescents diagnosed with HIV infection.

It has been reported that HIV antibodies can be detected, with a high degree of sensitivity and specificity, in saliva and in oral mucosal transudate (OMT) from HIV-1-infected patients. A large number of studies, indicate that most ELISAs can be used to test oral fluids with a sensitivity of 95-100% and a specificity of 99.5-100%. Of particular note is the excellent specificity, which indicates that the use of oral fluids may be appropriate as an epidemiological tool, as suggested by Frierichs et al., or as a possible confirmatory strategy. Several studies have suggested that the use of oral fluids for HIV testing as an alternative to serum would be advantageous for surveillance owing to the high specificity obtained.

Also the collection of oral fluids may be simpler than that of venous blood, particularly from children, obese individuals, and from persons whose veins are not easily accessible. In some instances, adequate amounts of blood are difficult to obtain because of cultural/religious reasons and/or collapsed veins, and several reports indicate that collection compliance is greater for oral samples than for blood. In addition, the use of oral fluids could help to reduce infection through the re-use of unsterilized needles. There may also be a cost saving when oral fluids are used as samples because collection requires minimal training of personnel. Furthermore, samples can be collected simultaneously from groups, while self-collection offers additional cost and time savings if large numbers of samples are to be collected. The cost of materials is less when whole saliva is used. Although blood collected by finger-, heel-, or ear-lobe-prick may be the least costly of all methods, it may also be the most painful and involves disposal hazards.

There have been several comparisons of the collection compliance rates of subjects for blood versus oral samples. One of the advantages of using oral fluids for HIV testing is the potential for a higher degree of collection compliance among subjects being tested for surveillance purposes, thereby reducing sampling bias. According to Major CJ et al. (1991), compliance rates were 83.3% for whole saliva compared with 69% for blood obtained by finger-prick. In Rwanda, compliance rates were over 99% for direct collection of whole saliva compared with a similar study that used blood, where the compliance was only 80-85%.

Like wise in our study, 100% patients reported to be convenient when tested using oral fluid when compared to 20 % patients who were apprehensive with the blood test involving the needle prick, as the oral fluid test was a painless, simple and non invasive procedure. It is also safe with least occupational exposures, easier sample collection and simple to per form. The test offers an immense benefit in pregnant patients without prenatal care and also in case of occupational exposures.

**Limitation**

Limitations are similar to that of rapid serum HIV testing. Firstly, there is need for confirmation of the serological status of the patient using Westernblot, if found seropositive with this oral fluid-based HIV antibody detection test. Secondly, it is unable to specify whether the sample contained HIV-1 or HIV-2 antibodies specifically, HIV-2 being less prevalent than HIV-1.

**CONCLUSION**

Dentists and emergency care providers are most commonly prone for infections from contact with patient’s bloodandoral fluids. Due to lack of adequate sterilization measures and detection facilities, there is a greater chance of not only acquiring infection but also spread in git among other patients unwittingly. Thus, incorporating this simple to perform test kit, using non invasive means, can greatly reduce the risk of transmitting the infection.

Our results provide justification for use of the rapid saliva test oral fluid samples successfully as an alternative to serum samples for HIV testing in community based settings. It highlighted the impact that this new technology may have on HIV screening programs. Rapid saliva testing may identify increasing numbers of HIV antibody–positive individuals, drawing attention to the disease by making more people aware of their status. This test kit is efficacious as an effective screening device for HIV antibody detection; further studies are warranted in larger group of population involving the dental set-up, emergency care units, pregnant patients and high-risk groups directed at diagnosis accuracy and to introduce as a routine office screening procedure.

**References**


---

How to cite this article:
Dr. M. Monica Gurupriya, Dr. P. Iyapparaja and Dr. P.D. Madankumar.2020, Correlation Between Serum and Salivary HIV Antibodies in A Cohort of Well-Controlled HIV Infected Adolescents. *Int J Recent Sci Res.* 11(08), pp. 39440-39444. DOI: http://dx.doi.org/10.24327/ijrsr.2020.1108.5504

*******