Low-cost monitoring of HIV infected individuals on highly active antiretroviral therapy (HAART) in developing countries

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The standard methods to monitor HIV infection are flow cytometry-based for CD4+ T lymphocyte count and molecular assays to quantify plasma viral load of HIV. Few laboratories in resource-limited countries can run these tests as a majority of the HIV infected individuals are poor. A number of currently available low-cost assays which require less expensive equipment and reagents, may be well-suited to such countries. These include manual and ELISA based CD4 cell assays, and ultrasensitive reverse transcriptase quantitation (Cavidi) and p24 (ELAST) assays to monitor virus load. But better internal quality assurance and quality control (QA/QC) programmes are essential. This review discusses the low-cost assays and their role in clinical monitoring of HIV infected individuals in resource-limited countries like as India.

Key words CD4 T-lymphocyte count - HIV monitoring - low-cost assays - plasma viral load - resource-limited countries

The cost of ART (antiretroviral therapy) has reduced dramatically due to the production by generic manufactures and this has led to the increased usage of highly active antiretroviral therapy (HAART) in developing countries1-5. But the cost of monitoring is prohibitively high. As antiretroviral therapy is becoming more affordable and accessible, inexpensive laboratory tests are also needed to monitor the progression of disease in HIV infected individuals, living in resource-limited environments most heavily impacted by the epidemic6.

Though clinical assessment remains the most essential basis for monitoring HIV infection, it lacks sensitivity in determining both disease stage and progression and, is thus used in conjunction with laboratory measures7. These include total lymphocyte counts(TLC), CD4+ T lymphocyte counts (CD4+ T cell), HIV plasma viral load (PVL), and other surrogate markers, such as β2-microglobulin levels8. Ideally, a surrogate marker should be biologically plausible, predictive of disease progression and measurable by standardized assays. Historically, a number of candidate markers have been explored for monitoring the course of HIV infection and response to treatment. While the level of absolute numbers of peripheral CD4+ T cells and the PVL have eventually become the reference markers in clinical practice for monitoring HIV infected subjects, several additional parameters are still being evaluated. These indicators are used to determine disease stage and progression, assist in decisions regarding when to start or change antiretroviral therapy, and assess treatment responses9. In addition, access to HIV genotypic or phenotypic
analysis is increasingly available and used to guide the choice of antiretroviral treatment. The recent reduction in cost and thus availability of generic antiretroviral drugs within many resource-limited countries has focused attention on the lack of infrastructure for effective monitoring of HIV infection in these countries. It is unlikely that the cost of laboratory assays to monitor HIV infection will be swayed to the same extent by similar pressure brought to bear on manufacturers of antiretroviral drugs.

The reference standard for CD4+ T cell testing is flow cytometric enumeration of T cell subsets. For PVL, a number of molecularly based assays that use reverse transcriptase-polymerase chain reaction (RT-PCR), branched DNA (b-DNA), and nucleic acid sequence based amplification (NASBA) technology are approved in many countries. Such reference assays are expensive, require sophisticated equipment as well as trained personnel, and are not widely available in resource-limited countries. Geographic difficulties, such as lack of ready access to technical support and quality control and quality assurance (QA/QC) programmes, also limit their potential use in such settings.

Over the past decade, low-cost, less technically demanding assays, that quantify CD4+ T cell and PVL have been developed and assessed but have not become generally established in resource-poor settings due to several reasons (Tables I, II). There is now a resurgence of interest in implementing these tests in regions in which flow cytometric and molecular facilities are not affordable or practical. The possibility of extending potent antiretroviral therapies to developing countries has raised the need of simple, reliable and cost-effective tests to measure prognostic markers for disease evolution and assessment of therapy efficacy. This review summarizes the benefits and limits of reference and candidate surrogate markers and their integration for optimal antiretroviral therapy.

The reference standard and low-cost monitoring of CD4+ T-lymphocyte

CD4+ T cell monitoring in conjunction with clinical status will be of benefit in terms of assessing prognosis, guiding the treatment, and subsequently monitoring the response. Also, CD4+ T cell count allows timely institution of primary prophylaxis against specific opportunistic pathogens and an associated reduction in hospital admissions, with use of affordable drugs such as trimethoprim-sulphamethoxazole. The precise CD4+ T cell count at which the risk of specific opportunistic infections justifies prophylaxis has been well established in the United States and Europe. At the population level, CD4+ T cell monitoring is necessary for regional studies of the natural history of HIV infection and assessing interactions of HIV with locally prevalent infectious diseases.

The reference standard for CD4+ T lymphocyte count: Multiparameter flow cytometric quantification of CD4+ T cells with microprocessor-assisted analysis is the reference standard. A wide range of instruments is available commercially from different companies, including Becton Dickinson, Coulter Corporation and Partec as well as newer instruments such as FlowCare (PointCare Technologies). The high initial cost of equipment and problems associated with equipment routine preventive maintenance, lack of technical support, and lack of access to QA/QC programmes have prevented widespread use of flow cytometric monitoring of CD4+ T cell counts in resource-limited countries. The cost of reagents to perform full lymphocyte subset multiplatform analysis, in which CD45+ T cells are identified by staining with 8 monoclonal antibodies (MAbs) by means of 3-colour fluorescence to identify CD3+/CD4+ and CD3+/CD8+ T cells, is also prohibitive in many countries. However, new approaches include primary gating, in which CD4+ T cells are defined by an autogate in a single histogram of CD4+ T cell fluorescence intensity versus light scatter by use of a single-platform volumetric flow cytometer. This is considered relatively economical compared with use of the full test panel of MAbs. Among the Flow cytometers available, the FACSCount (Becton Dickinson) instrument is a simpler, relatively less expensive and automated for clinical laboratories that quantifies CD4+/CD8+ T cell counts by simultaneous fluorescent MAb staining of CD3, CD4, and CD8 surface molecules by use of different fluorochromes attached to each MAb.
### Table I. Standard and low-cost assays to enumerate CD4+ T cells

<table>
<thead>
<tr>
<th>Assay</th>
<th>Key features</th>
<th>Advantages</th>
<th>Disadvantages</th>
<th>Cost/test in INR (Approximate)</th>
</tr>
</thead>
<tbody>
<tr>
<td>FACSCCount (Becton Dickinson): 2 color Flow cytometer *</td>
<td>Dedicated (single bead platform) flow cytometry instrument for CD4, CD8 and CD3 absolute counts and CD4/CD8 ratio</td>
<td>Fully automated, no red cell lysis, no flow cytometric experience required</td>
<td>No CD4 %, closed cell lysis system, specific reagents, long processing time, require accurate reverse pipetting skill, high cost per test</td>
<td>1200.00</td>
</tr>
<tr>
<td>Manual CD4 kit, Cytosphere assay (Beckman Coulter)</td>
<td>Latex bead-based kit for light microscope, Manual count of rosetted cells</td>
<td>Absolute CD4 result, simple, low cost microscope, pipette and haemocytometer, can be performed at remote sites</td>
<td>No CD4 % value, low throughput, high cost per test, labour intensive. The samples should be processed within 6 h</td>
<td>600.00</td>
</tr>
<tr>
<td>Dynabeads CD4/CD8 kit (Dynal, Biotech)</td>
<td>Kit based Magnetic bead cell isolation, requires fluorescent microscopy or ordinary microscope</td>
<td>Absolute CD4 result, simple, low-cost microscope, pipette, can be performed at remote sites</td>
<td>No CD4 % value, low throughput, high cost per test, labour intensive, long process time, specific magnet and mixer required from the company. The samples should be processed within 24 h</td>
<td>700.00</td>
</tr>
<tr>
<td>Capcellia (BioRad)</td>
<td>ELISA based using CD2 magnetic bead isolation, color reaction detection for CD4/CD8</td>
<td>CD4/CD8 absolute from standard curve calculation, 40 tests/ELISA plate, 15 samples/h</td>
<td>Labour intensive, requires microplate reader, magnet, multichannel pipette, calculation based on standard curve with controls, Batch analysis required. The samples should be processed within 6 h</td>
<td>400.00</td>
</tr>
<tr>
<td>Easy CD4/CD8 (Guava Technologies)</td>
<td>Micro-capillary flow cytometry (single platform volumetric), absolute CD4/CD3 and/or CD8/CD3,</td>
<td>Additional applications, require small volume of blood, minimal waste disposal, require minimal training for the operator. Allows 48 h samples</td>
<td>No CD4%, daily bead calibration, computer literacy, requires red cell lysis and specific antibody reagents, ~20 samples/day</td>
<td>300.00</td>
</tr>
<tr>
<td>Partec CyFlow</td>
<td>Dedicated (single volumetric platform) flow cytometer. Absolute CD4/CD8 depends on choice of reagent</td>
<td>No lyse system, no beads, 15 min turnaround, can give percentage CD4</td>
<td>Volumetric counting stability is questionable, no independent validation of performance, computer software literacy</td>
<td>200.00</td>
</tr>
</tbody>
</table>

*Standard assay
Total lymphocyte count as a surrogate marker for absolute CD4+ T cell count: In April 2002, World Health Organization (WHO) released provisional guidelines for implementation and scaling up of antiretroviral treatment in resource-limited settings. Recognizing the urgent need for ART in developing countries, where sophisticated and expensive laboratory monitoring is not widely available, WHO is advocating the use of simple and inexpensive laboratory measures in determining response to treatment. The total leucocyte count (TLC) is available to most clinics worldwide at very little cost and it requires just the leucocyte count plus the differential i.e., the total leucocyte count is multiplied by the percentage of lymphocytes to obtain the TLC. Where CD4 count is unavailable, TLC use has already been recommended for the timing of opportunistic infection prophylaxis in Latin America, Africa, and Asia. For example, in a cohort of HIV-positive south Indians, a TLC<1400 cells/µl is a good predictor of CD4+ T count <200 cells/µl and thus, an appropriate surrogate marker for initiating co-trimoxazole prophylaxis in resource-limited settings.

Table II. Standard and low-cost assays to quantify HIV

<table>
<thead>
<tr>
<th>Assay</th>
<th>Key features</th>
<th>Parameters measured</th>
<th>Advantages</th>
<th>Disadvantages</th>
<th>Cost/test in INR (Approximate)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Roche Amplicor (version 1.5)*</td>
<td>Cut-off 50 copies/ml with standard assay and 400 copies/ml with ultrasensitive assay. Semi-automated (manual) or automated (COBAS)</td>
<td>HIV RNA by RT-PCR</td>
<td>• Can be used for other diseases such as HBV, HCV and CMV. • Can be used for all clades • High throughput</td>
<td>• Requires PCR set-up. • COBAS for automated and PCR thermocycler for Manual. • Contamination risk • Skill. • Higher cost. • Infrastructure • Need good technical support</td>
<td>4000.00</td>
</tr>
<tr>
<td>Perkin Elmer Ultrasensitive p24</td>
<td>Clinical cut-off around 30,000 copies/ml</td>
<td>HIV P24 antigen</td>
<td>• Equipment can be shared (ELISA:reader/washer) • Uses same consumables as ELISA testing • Easy training • 1 day turnaround time • Easy to perform assay • High throughput</td>
<td>• May not be effective for all clades • Batch analysis alone reduce substantial cost</td>
<td>700.00</td>
</tr>
<tr>
<td>Cavidi ExaVir</td>
<td>Cut-off approximately 5,000 copies/ml.</td>
<td>HIV reverse transcriptase activity</td>
<td>• Easy training • Can be used for all clades? • Easy to perform assay</td>
<td>• 32°C dedicated incubator needed • performance time about 3 days • Batch analysis alone reduce substantial cost • Needs more evaluation • Positive and negative control not supplied</td>
<td>900.00</td>
</tr>
</tbody>
</table>

*Standard assay
and TLC; the latter is not sufficiently useful to predict CD4 cell counts in many settings. In a study of 2777 HIV-seropositive persons in South Africa\(^ {20}\), the overall correlation between CD4 cell and total lymphocyte count was only modest \((R=0.70)\). This is supported by a prospective study in the United Kingdom\(^ {21}\), in which the correlation between 1535 paired absolute lymphocyte and CD4 cell counts was 0.64 for persons with asymptomatic HIV infection, and a study of 32 Nigerian adults\(^ {22}\) with HIV infection who commenced treatment with antiretroviral drugs, in which there was only a very weak correlation between total lymphocyte and CD4 cell count \((R=0.25)\). These studies indicate that TLC is an imperfect predictor of CD4 cell count. Therefore, the larger longitudinal studies involving long term cohort follow up may answer the question.

**Low-cost alternative assays for CD4+ T cell count**: Non-cytofluorographic methods are largely of the ELISA format (Capcellia CD4/CD8 Whole Blood Assay; BioRad and TRAx CD4 Test Kit assays; T Cell Diagnostics, which is currently unavailable), or manual methods, using magnetic beads (Dynabeads T4-T8 System; Dynal Biotech ASA) or cytospheres (Coulter Manual CD4 Count kit). The assays with modified flow cytometry technologies are EasyCD4/CD8 (Guava Technologies) and CyFlow (Partec). Extensive evaluation studies have been carried out in different countries on the efficiency of the each assay in providing accurate and reproducible results.

The Capcellia CD4/CD8 whole blood assay is an ELISA that uses MAb-coated paramagnetic microparticles to identify T cells (by use of anti CD2 MAb) followed by anti-CD4 (or anti-CD8) MAb-peroxidase conjugate. The sample should be collected in EDTA anticoagulant and processed within 6 h. The assay is in a 96-well format, and up to 43 samples can be assayed in a single run per kit (plus standards). A magnetic frame, plate reader, manual washing manifold, and pipettes (including a multichannel pipette) are used for the assay. The Capcellia assay provides an absolute CD4+ T cell count calculated from a 4-point standard curve. This measurement of CD4+ T cells has been found to correlate well with flow cytometry in European laboratories \((R=0.81)\)\(^ {23}\), although its performance was lower in West Africa in an evaluation of HIV infected and uninfected persons \((R=0.61)\)\(^ {24}\). The studies from India also reveal the significant correlation with the standard methods\(^ {25,26}\).

The Dynabeads T4-T8 system uses “dynabeads”, which are magnetic particles coated with antibodies to CD4 or CD8 molecules. The sample should be collected in EDTA or acid-citrate-dextrose anticoagulant and processed within 24 h. Only 250 µl of blood is required for the assay. Monocytes are removed by using additional beads coated with anti-CD14, thus avoiding potential confusion when counting cells under the microscope. CD4+ T cells are isolated by use of the magnet and lysed, and nuclei are counted after staining with Sternheimer-Malbin staining solution (containing crystal violet, safranin O, ammonium oxalate, and ethanol). Alternatively, nuclei are stained with acridine orange and counted by fluorescence microscopy. The absolute CD4 cell count is obtained by counting all the cells in one side of a 0.1 mm haemocytometer and dividing the number by 0.9. Because counts must be done within 1 h after addition of lysis reagent, the batch size needs to be limited to <6 samples. In addition to a microscope, a haemocytometer, a manual counter, a rotating wheel to mix samples, and a magnet are required. Several investigators have reported good correlation with flow cytometry \((R=0.9)\)\(^ {27,28}\). Recently the study from West Africa has shown that the implementation of this low-cost method was easy and successful in the West African context\(^ {29}\).

The Coulter Manual CD4 Count kit uses “cytospheres” (latex beads coated with anti-CD4) that bind to CD4+ T cells to form a cell sphere rosette, detected by light microscopy. Monocytes (which also express CD4 on their surface) are identified by binding to smaller spheres coated with anti-CD14. Blood should be processed within 6 h, according to the manufacturer’s instructions. Only 100 µl of blood, collected in EDTA anticoagulant, is required. Very little equipment is needed for this assay: a microscope with a 40X objective, a haemocytometer, a manual counter, test tubes, and calibrated pipettes. Erythrocytes are lysed, and crystal violet stained CD4+ T cells with beads attached are identified by counting in a haemocytometer. Cells in all squares on both sides of a 0.1mm haemocytometer are counted.
and the number multiplied by a dilution factor of 7.3 to obtain the absolute CD4+ T cell count. The sample is stable for only approximately 15 min after addition of lysis/stain reagent, which limits the batch size to approximately 2-4 samples. Several investigators have found a good correlation between CD4+ T cell counts measured by the cytosphere method and flow cytometry ($R > 0.91$)\textsuperscript{30-32}. The recent study from our laboratory has shown excellent correlation with the standard assay\textsuperscript{33}. The correlation coefficient of the cytosphere assay compared with that of flow cytometry for CD4+ T cells was 0.97 ($P < 0.0001$), with a confidence interval (CI) of 0.95 to 0.98. The sensitivity, specificity, positive predictive value, and negative predictive value of the cytosphere assay in enumerating absolute CD4+ T cell counts of less than 200/µl were 94.9, 96.4, 92.5 and 97.6 per cent, respectively. Overall this manual assay yielded CD4+ T cell counts greater than flow cytometry by a mean of 10 cells/ml (95% CI, -152 to 132 cells/ml); as assessed by Bland-Altman analysis. However, another study\textsuperscript{27}, which analyzed only 10 samples, has found poor correlation with flow cytometry ($R = 0.45$) and the poor correlation could be due to sample bias. In general, this is a simple inexpensive method and does not require special equipment.

**CyFlow** is a portable flow cytometry system. The machine is less expensive and more robust than conventional FACS systems, uses less expensive reagents, and is able to produce an absolute CD4+ T cell count without additional instrumentation. A mobile CD4+ T cell counting laboratory can be established for the field level use. CyFlow is able to produce an absolute CD4+ T-cell count using two monoclonal reagents. Investigators from Cambodia, Thailand, Malawi, Negeria and Rwanda have evaluated the precision of the CyFlow system and observed good correlation with the standard assay\textsuperscript{34}.

**Guava EasyCD4/CD8 assay** is a micro-capillary flow cytometry technology based system. The reagent kit contains three direct immunofluorescence reagents for enumeration of mature CD4+ and CD8+ T cells. The kit consists of a murine monoclonal anti-human CD3 antibody conjugated to the tandem dye phycoerythrin (PE)-Cy5 (PECy5), a murine monoclonal anti-human CD4 antibody conjugated to PE and a murine monoclonal anti-human CD8 antibody conjugated to PE. The CD3 antibody uniquely identifies T cells and recognizes an epitope expressed on the epsilon chain of the CD3/T cell antigen receptor (TcR) complex. This assay provides a method for enumeration of CD4+ and CD8+ T cells. Whole blood is stained with the antibodies from the EasyCD4/CD8 Reagent Kit, which bind to antigens on surface of lymphocytes; then erythrocytes are removed by lysis by treating with Lysing Reagent to lyse and finally analyzed on the Guava PCA instrument to obtain the absolute CD4+ and CD8+ T cell counts. This assay requires EDTA blood and the sample should be processed within 48 h of collection for optimal results. Our study (unpublished) has shown that it is easy to perform, generate reliable results (correlation coefficient 0.99) and the technicians needs substantially less training than the standard flow cytometry. This assay requires micro-volume of specimen and generates very minimal biowaste. Also the studies from America and South Africa have shown the good correlation\textsuperscript{35,36}.

**Flow cytometry versus low-cost assays to enumerate CD4+T cell count**: There are advantages and disadvantages of each system (Table I). Flow cytometry is considered the reference standard. However, disadvantages include the fact that, it is expensive in terms of initial cost of equipment and requires a haematologic analyzer to provide the absolute CD4+ T cell count. The use of the equipment is technically complicated, and highly trained personnel are generally needed to run these instruments. There are reports that lysis of RBCs during flow cytometric analysis may be problematic\textsuperscript{37}; resistance to lysis (possibly associated with elevated levels of nucleated erythrocytes in blood) has been reported in African patients\textsuperscript{28}. Further, lysis may result in cell loss (up to 20%), with significant donor and lysing solution variability\textsuperscript{38}. The advantages of flow cytometry include provision of percentage and absolute CD4+ T cell count. Whereas most clinicians generally use the absolute CD4+ T cell count in the clinic, the CD4+ T cell percentage is very useful as a built-in control to determine whether the CD4+ T cell count is changing or stable, needed because of often large variation in absolute CD4+ T cell counts despite clinical stability\textsuperscript{39}. Also biological variables, including diurnal variation,
exercise or rest, and smoking, can significantly alter CD4+ T cell count.

For major cities in regions, where there is a high incidence of HIV infection and a large number of samples need to be analyzed per day, manual methods are not practical. Provided there is access to company technical support and QA/QC programmes, automated evaluation of CD4+ T cells (perhaps by use of an instrument such as the FACSCount) may be the most economical method on a long-term basis. The FACSCount is simple; the equipment, although still costing significantly (approximately Rs.22 lakhs), it still is less expensive than other standard flow cytometry instruments. It does not require a highly trained personnel and lysis of blood samples is also not required. A disadvantage of the method is that only the absolute CD4+ T cell count is provided and haematology analyzer has been used for percentage of CD4+ calculation.

The manual methods such as Dynal and Coulter assays are suitable for smaller regional centres where <10 samples are requested per day and where there is no ready access, including convenient and reliable transport, to laboratories with flow cytometry facilities. Both of these methods are simple and inexpensive and require very little investment for the equipment (ordinary light microscope). Both provide only absolute CD4+ T cell counts. Counting under the microscope is both time consuming and laborious (approximately 10 min/sample). The studies show that higher cell counts are more prone to inaccuracy compared with values obtained by flow cytometry. The Capcellia assay is also appealing because, it is an ELISA-based assay and as such it is readily established in most small laboratories and Voluntary Counselling and Testing (VCT) centres.

Though the manufacturers of these manual assays state that blood should be assayed within the storage period (6 h for Coulter) after the collection, this is not always possible in regions, where transport may be delayed, and thus the robustness of these assays for use in the field requires scrutiny. Analysis of specimen transport condition and performance of these assays with the aged samples require further studies and currently the work is in progress in our laboratory.

Low-cost monitoring of HIV load

Molecular assays, including the Amplicor HIV Monitor RT-PCR assay (Roche Diagnostic Systems) are now available in many countries (this can be done manually or with fully automated using COBAS equipment). Early version (1.0) of the Amplicor RT-PCR assay was insensitive in detecting certain HIV subtypes. In an evaluation of the Amplicor assay, detection of subtypes A and E was considerably improved with version 1.5 compared with version 1.0. The Quantiplex HIV RNA assay, which uses b-DNA technology (Bayer Diagnostics), and NASBA (Organon Teknika), are the other reference standards for monitoring plasma HIV load. The NASBA assay has also been reported to fail detection of or inadequately quantify, HIV RNA in plasma of persons infected with non-B subtypes, whereas the b-DNA assay has generally been reported to perform well with non-B subtypes.

There are very few low-cost assays (with no need to purchase any special equipments) that may provide an alternative for virus load measurement. Although several low-cost assays for plasma HIV load are at the research level, there are two assays (Table II), which have been evaluated in different countries. These include a p24 Ultrasensitive Assay; ELAST (Perkin Elmer Life Sciences) and an ultrasensitive reverse-transcriptase (RT) assay (RT Viral Load Kit; Cavidi Tech).

The HIV p24 Ultrasensitive Assay provided a 3-log increase in sensitivity above the standard p24 assay by means of a signal amplification-boosted ELISA method (enzyme-linked amplified sorbent test; ELAST) following heat denaturation of the plasma sample to release immune-complexed HIV p24 antigen. The assay provides results comparable to those of RT-PCR, is in a microplate ELISA format and is significantly inexpensive than the standard. The plasma p24 levels appeared to be correlated significantly with the plasma RNA viral loads. The heat-denatured p24 antigen assay was capable of measuring the plasma level of p24 derived from all the HIV-1 subtypes and recombinants selected for this study, in contrast to the RNA viral load test which lacked sensitivity towards HIV-1 group O.
Cavidi RT assay (ExaVir Load): The procedure has only 2 major steps (purification of virions and recovery of RT, including removal of potential inhibitors, followed by quantification of RT activity in a 96-well microtiter plate format). Very little equipment is necessary (provided by the company as a start-up kit at free of cost): mainly a vacuum pump, waste bottle, some rubber tubing and an ELISA reader or a fluorometer and some buckets for the wash procedure. The company reports excellent correlations between standard RT-PCR assay and the Cavidi RT assay ($R=0.9$, $P<0.0001$). The fluorometer appears to be more sensitive than the ELISA reader. The assay is not affected by the presence of RT inhibitors (drugs) in plasma of HIV infected persons. However, this assay needs more evaluation with different subtypes before using it for the ARV monitoring. Another advantage of this low-cost viral load assay is that samples can be stored and batch testing can be done. Currently our centre is evaluating this assay for the suitability of the kit in monitoring HIV infected individuals, who are on generic HAART. These low-cost viral load assays have their greatest value in smaller laboratories in resource-limited countries, where the cost is a major issue and, where setting up of RT-PCR or similar molecular assays is difficult and these assays are unavailable.

Other surrogate markers for HIV monitoring

Extensive studies have been carried out to evaluate whether B2-microglobulin and neopterin could be used as markers of disease progression and also as prognostic factors. In general, these have not been found to be reliable. These factors in combination with CD4+ T cell counts, have been shown to have good predictive value. One potential reason for lack of correlation may be the fact that a number of other infectious agents may also increase B2-microglobulin and neopterin levels. Thus, B2-microglobulin and neopterin may be an unreliable indicator of HIV infection or disease stage in those co-infected with other pathogens.

HIV monitoring in resource-limited settings

The HIV/AIDS statistics are well known, with resource-limited countries bearing more than 90 per cent of the global disease burden. Added to the cost of ARV therapy are substantial costs associated with laboratory monitoring, which also include liver function tests (LFT) and CBC (complete blood count). The inclusion of some important parameters of the ART toxicity-related assays such as platelet, haemoglobin, erythrocyte sedimentation rate (ESR), bilirubin (direct and indirect) and serum glutamate pyruvic transaminase (SGPT) could also be a cost-saving strategy in the developing nations.

Although the introduction of antiretroviral therapy to resource-limited countries should not be prevented because of a lack of monitoring tests, every effort should be made to introduce appropriate laboratory monitoring assays. Sustainability of assays (recognizing issues such as ongoing costs, delivery of reagents and kits, kit shelf-life, access to instrument service, and QA/QC programmes) must also be considered at the time of technology transfer. As a minimum, a low-cost CD4+ T cell count should be monitored as an adjunct to clinical examination. Establishing CD4+ T cell monitoring facilities must include implementation of QA/QC program. Since there are some CD4+ count difference between standard assay and alternative technologies, the normal reference ranges should be established with the local population for any particular low-cost assay. Implementation of low-cost viral load assays awaits rigorous evaluation of their performance against standard assays.

The incidence of HIV infection and clinical disease continue to increase rapidly in the developing nations. As antiretroviral therapy is becoming more affordable and accessible in India, inexpensive laboratory tests are very much needed to monitor the disease progression and ART. Hence the implementation of HIV monitoring by low-cost assays would reduce the economic burden of the patients in this country.

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