A new model to monitor the virological efficacy of antiretroviral treatment in resource-poor countries

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Monitoring the efficacy of antiretroviral treatment in developing countries is difficult because these countries have few laboratory facilities to test viral load and drug resistance. Those that exist are faced with a shortage of trained staff, unreliable electricity supply, and costly reagents. Not only that, but most HIV patients in resource-poor countries do not have access to such testing. We propose a new model for monitoring antiretroviral treatment in resource-limited settings that uses patients' clinical and treatment history, adherence to treatment, and laboratory indices such as haemoglobin level and total lymphocyte count to identify virological treatment failure, and offers patients future treatment options. We believe that this model can make an accurate diagnosis of treatment failure in most patients. However, operational research is needed to assess whether this strategy works in practice.

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Introduction

Despite the fact that the WHO's "3 by 5" target—ie, getting 3 million people in low-income and middle-income countries on antiretroviral therapy by the end of 2005—was not reached, a lot of progress has been made.[1] Today it is evident that to scale up the roll out of antiretrovirals even more, we will need to further decentralise antiretroviral services, and this will require new and less expensive ways to monitor the virological efficacy of treatment.[2,3]

In developed countries, the decision to change antiretroviral therapy is based on a rise in viral load, a fall in CD4+ lymphocyte counts, and the presence of drug resistance.[4] Monitoring the efficacy of treatment in countries with limited resources is more difficult because of inadequate laboratory facilities, a shortage of trained staff, unreliable electricity supply, and costly reagents. Even where such tests can be done, most HIV patients in developing countries do not have access to them.[5]

Here, we review existing methods to monitor the effectiveness of antiretroviral treatment, and propose a new model that could be used in resource-limited settings. The purpose of the model is to diagnose virological treatment failure early enough to allow a switch to second-line regimens. Changing treatment early will prevent resistance from developing further, both in the individual and in the population.

The model is largely a result of the experience of physicians working at the Infectious Diseases Institute (IDI) of the Makerere Medical School in Kampala, Uganda. The institute's clinics in Mulago hospital offer medical treatment free of charge to more then 12 000 adults and 3000 children with HIV, of whom about 3000 are being treated with antiretrovirals. The IDI laboratory can measure CD4+ lymphocyte cell counts and test viral load, but does not have enough funding to offer these tests routinely to all patients. Weekly meetings are held to decide which patients are failing antiretroviral treatment, and whether a switch to second-line antiretroviral therapy is justified.

Existing methods for monitoring the efficacy of antiretroviral therapy

The effectiveness of antiretroviral treatment can be monitored virologically, immunologically, or clinically.

Virological monitoring

Virological monitoring has been considered the ideal method for assessing the efficacy of an antiretroviral regimen. However, HIV RNA viral load testing is costly ($15–150 per test), and needs both adequate laboratory infrastructure and well-trained personnel. Alternative technologies are being developed for viral load testing—eg, heat-denatured quantitative p24 antigen measurement, the Cavidi real-time reverse transcriptase assay, and real time PCR assays.[6-12] But these tests are expensive too (about $3–15 per test). The p24 antigen test might be useful in diagnosing HIV infection in infants,[13] but this test and others need further field validation in resource-poor settings before they are recommended for monitoring antiretroviral treatment.

Ideally, drug resistance should be tested for in any case of virological treatment failure. Complete genotypic resistance testing, however, costs about $300 per test. A way to reduce this cost could be to develop tests that identify only key genetic mutations such as the Met184Val mutation, indicating resistance to lamivudine, and the Lys103Asn and Tyr181Cys/Ile mutation, indicating resistance to nevirapine or efavirenz.[14] In developing countries, the generic fixed drug combination of lamivudine, stavudine (or zidovudine), and nevirapine is most widely used.[15–17] Therefore, knowledge about the presence or absence of these key mutations could help decide whether an antiretroviral regimen containing these drugs should be stopped.

Choosing the best second-line therapy needs additional information about the presence or absence of thymidine analogue mutations, since thymidine analogues—eg, zidovudine or stavudine—are often used in resource-poor countries.

Immunological monitoring

CD4+ lymphocyte counts are useful in detecting asymptomatic patients or patients with minor symptoms (WHO stage II disease) who need antiretroviral therapy, or determining when to start or stop prophylaxis for opportunistic infections. But it is of less value as an indirect measure of antiretroviral efficacy. Indeed, in most instances once a patient has failed immunologically, viral resistance has already evolved.

Moreover, the change in CD4+ count might vary from one patient to another regardless of the virological efficacy of a treatment regimen. A patient with only a slight increase in CD4+ levels (<50 cells/μL after one year of highly active antiretroviral therapy [HAART]) has not necessarily failed treatment.[18,19] Conversely, a patient on an ineffective regimen, and in whom the HIV strain is resistant to only one or two drugs in the treatment, may have a continued rise in CD4+ count.[20,21] WHO criteria for immunological treatment failure are a CD4+ count that falls below the baseline level or one that falls by more than 50% after an initial increase.[1]

Ideally, CD4+ levels should be measured when the patient does not have an active opportunistic infection since intercurrent infections can cause these levels to fall.[22] But in resource-poor settings, many opportunistic infections are difficult to diagnose. Therefore, in a patient who is not doing well clinically, with a falling CD4+ count, it is often unclear whether this is because of an intercurrent illness, HIV disease progression, or both.

In Africa, measuring CD4+ levels with flow cytometry can cost $4–25 per test. Because of this high cost, alternative tests are under investigation, including microscopic, bead-based manual methods (Dynabeads, cytospheres) and more affordable modified flow cytometry (Guava, Cyflow, PointCARE) and other methods.[23,24] But these tests are expensive and can be labour-intensive.[23–29]

Measuring the extent of immune activation could be another way to provide useful information about the virological response and immune reconstitution of a patient taking antiretroviral therapy. In European cohorts, the proportion of CD8+ T cells expressing the activation marker CD38+ correlated well with the virological response.[30] Although data from the Cote d'Ivoire suggest that monitoring immune activation could have a role in assessing treatment failure,[31] its use in the presence of endemic parasitic or other co-infections remains unclear, since such infections may render these markers non-specific.

In poor countries, few people have access to CD4+ count testing. Therefore, it is worth investigating whether simple laboratory tests such as total lymphocyte count and haemoglobin levels might be used to predict immunological failure during HAART. Several investigators have shown that once a patient's total lymphocyte count falls below 1200 cells/μL, the likelihood that they have a CD4+ count below 200 cells/μL is more than 90%.[32–38] There is, however, no total lymphocyte count cut-off value that has both a high enough specificity and sensitivity for detecting patients with a CD4+ count lower than 200 cells/μL.[32–38] In the Multi-AIDS Cohort Study, a rapid fall in total lymphocyte count or haemoglobin concentration indicated an increased likelihood that HIV infection would progress to AIDS.[39]

In another study, within the first 2 years of HAART, whether or not total lymphocyte count rose or fell emerged as a strong marker for the direction of concomitant change in CD4+ (sensitivity 86–94% and specificity 80–85%, depending on length of interval).[40] Despite the fact that studies demonstrate increases in total lymphocyte count in patients receiving HAART,[39,40] whether this assay could be a useful method for monitoring the efficacy of treatment is unclear.[41–43]

Haemoglobin response to antiretroviral therapy may also be monitored inexpensively. Although it might not be the case in all patients treated with zidovudine,[44] studies have shown that haemoglobin levels generally increase in patients on HAART.[45,46] In a Belgian study in patients on HAART,[42] an increase of haemoglobin level and total lymphocyte count above baseline values at week 24 diagnosed virological treatment failure with a sensitivity of 46·9%, a specificity of 59·8%, a positive predictive value of 76%, and a negative predictive value of 29·2%. A fall in haemoglobin level and total lymphocyte count below baseline was as accurate in predicting virological treatment failure as was a fall in CD4+ count below baseline.[42]

Clinical monitoring

As resource-poor settings get better access to antiretrovirals, they will need monitoring systems that are different from those in developed countries. Without laboratory support, especially in rural settings, there will be a need to rely more on clinical monitoring, using symptoms and signs predictive of virological failure. HAART decreases the incidence of opportunistic infections, including *Mycobacterium tuberculosis* infection.[47,48] Clinical signs suggesting treatment failure, proposed by WHO are: the appearance of new or recurrent WHO stage III and IV conditions.[1]

However, during the first 3–6 months after starting HAART, clinicians might find it difficult to diagnose treatment success or failure on the basis of clinical findings alone. In the months after starting on HAART, patients can develop symptoms that are not caused by treatment failure but instead represent the side-effects of antiretroviral therapy, an immune reactivation inflammatory syndrome (IRIS),[49] opportunistic infections that continue to appear because the patient is still immunocompromised, or an infection or re-infection by a common endemic pathogen such as tuberculosis or malaria.

In South Africa, Grant and co-workers have shown that individuals on HAART had a major reduction in

Panel: HIV-related symptoms or signs predicting treatment failure

- Prurigo
- Unexplained persistent diarrhoea
- Unexplained persistent fever
- Unexplained weight loss
- Unexplained polyneuritis*
- Unexplained cognitive impairment
- Loss of developmental milestones in children
- Growth retardation in children

*Drug-induced polyneuritis should be excluded

mortality, although this benefit was only seen after several months of treatment.[50] If a patient adheres to a good HAART regimen and has not previously had antiretrovirals, very few treatment failures will occur during the first 6 months.[51] After 6 months of HAART, clinical manifestations will be more useful for predicting treatment success or failure. This will be particularly so in patients who were symptomatic (WHO stage III and IV) at the start of antiretroviral therapy, a group that currently represents most of those who start antiretroviral therapy in countries with limited resources. These patients' symptoms will have regressed or disappeared, and they will have gained weight. In addition, although they might still have occasional episodes of oral candidiasis, they should not develop new severe opportunistic infections.

We therefore propose that in symptomatic patients the effect of HAART can be seen in HIV-related symptoms and signs—eg, prurigo, chronic diarrhoea, HIV-related polyneuritis, and HIV-related cognitive disorders in adults. In children, growth, neurological, and sexual development are additional features to follow (panel).

In our experience in Uganda, the disappearance or reappearance of prurigo seems to be a good indicator of treatment success or failure. Prurigo occurs in at least 10% of African patients with advanced HIV disease.[52] Once prurigo appears, in the absence of antiretroviral treatment, itching and papular eruptions generally persist and symptomatic treatment is ineffective. Occasionally, the number of prurigo lesions might rise shortly after starting HAART (probably due to IRIS) but in most patients the prurigo disappears within a few months after starting antiretroviral therapy and reappears within weeks after HAART is stopped (RC, H Byakwaga, personal observation).

Asking what symptoms were present before antiretroviral therapy was started is important. Often, these symptoms will have disappeared during therapy. If some of these symptoms reappear, this, in our experience, suggests treatment failure. HIV-related polyneuritis is a much less useful clinical index because these patients have been exposed to antiretroviral and other drugs capable of producing peripheral neuropathy. An unexplained weight loss after an initial weight gain during antiretroviral treatment is another clinical indicator that might suggest treatment failure.

Kaposi's sarcoma lesions, particularly if lesions are very extensive and associated with oedema, may increase after starting HAART because of an IRIS.[53] However, mucocutaneous lesions that reduce in size are a good indicator that the patient is on an effective antiretroviral treatment.[54]

A new model for monitoring antiretroviral treatment

The traditional western model of monitoring patients on antiretroviral treatment with regular viral load and CD4+ lymphocyte counts is not feasible in poor countries. On the other hand, monitoring only for clinical and immunological failure is a risky strategy, both for patients and for the community as a whole. Waiting for clinical failure might leave patients susceptible to serious opportunistic infections and allowing patients to fail virologically for prolonged periods of time will lead

Risk factors for virological failure	Predictive value of the criteria
Treatment history	
Previous monotherapy or bi-therapy with NRTIs for more than 6 months	Minor
Previous exposure to nevirapine for the prevention of mother-to-child transmission of HIV	Minor
Infected with HIV by partner with a history of antiretroviral exposure	Minor
Current "weak" antiretroviral regimen (eg, 3 NsRTI, or 2 NsRTI and 1 NtRTI)	Minor
Long-term use of drugs that could reduce antiretroviral drug levels in the system	Minor
Adherence history	
Day-to-day adherence score (<95% but ⩾80%)	Minor
Day-to-day adherence score (<80% but >60%)	Major
History of stopping an NNRTI-containing regimen without continuing NRTIs for at least 5 days	Minor
Clinical history	
Appearance or worsening of unexplained prurigo	Minor
Reappearance of unexplained prurigo and at least one other HIV-related symptom or sign (not in first 6 months, or not thought to be IRIS)	Major
Reappearance of at least two other HIV-related symptoms or signs (not in first 6 months or not thought to be IRIS)	Minor
Body weight equal or lower than the patient's weight before starting HAART or more than 10% weight loss from peak values in the absence of signs of lipoatrophy	Minor
Development of a new WHO stage IV opportunistic infection (excluding extrapulmonary tuberculosis and IRIS) or malignancy (not in first 6 months or not thought to be IRIS)	Major
A recurrent WHO stage III opportunistic infection	Minor
Tuberculosis and no evidence of tuberculosis IRIS (abscess /cavity formation)	Minor
Worsening Kaposi's sarcoma	Minor
Worsening after initial improvement of Kaposi's sarcoma	Major
Laboratory history	
Unexplained fall of haemoglobin of 10% on two occasions and a reduction in TLC of 50% from peak values on consecutive testing or haemoglobin and TLC falling below baseline on two consecutive tests*	Minor
A reduction in CD4+ count to 50% from peak values on two consecutive tests or A CD4+ count below baseline values on two consecutive tests*	Minor

*These tests should ideally be done in the absence of an acute intercurrent illness. NRTI=nucleotide reverse trasncriptase inhibitors, singly phosphorylated (NsRTI) or triply phosphorylated (NtRTI); NNRTI=non nucleoside reverse transcriptase inhibitors; TLC=total lymphocyte count.

Table: **System for assessing the risk of virological failure for a first-line regimen. To be used after at least 6 months of treatment; treatment failure is estimated as probable if at least one major or at least three minor criteria from different categories are met.**

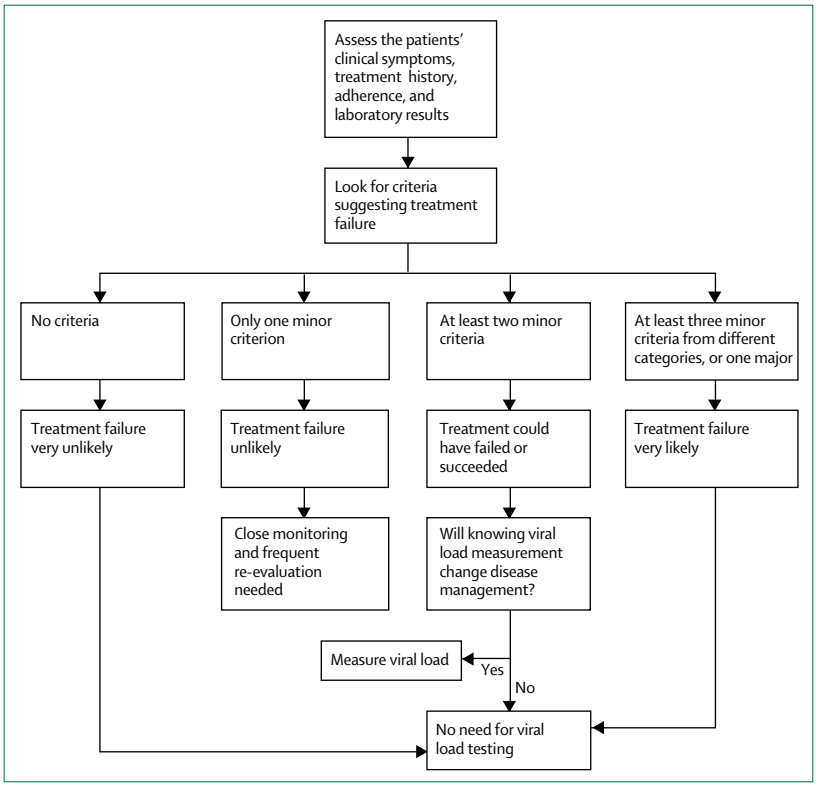

Figure: **Algorithm to monitor the virological efficacy of an antiretroviral regimen when there is no or little access to viral load testing**

to drug-resistance mutations in the HIV virus and facilitate the spread of resistant viral strains.

If this happens on a large scale, the benefits of the antiretroviral scale-up programmes will be short-lived. To limit the development of resistance, and to keep monitoring feasible and affordable, we propose a new model for monitoring the virological success of antiretroviral treatment.

Steps to assess virological treatment failure

The table and figure show our algorithmic approach for determining the need to change therapy or seek further testing. The assessment system (table) uses minor and major WHO criteria (panel) to predict virological failure. Criteria not detailed by WHO were scored as major or minor by doctors at the IDI, and by all co-authors and reviewers of the paper (see Acknowledgments). The steps involved in the assessment system are as follows.

Obtain an antiretroviral treatment history

Previous use of antiretrovirals either with monotherapy or bi-therapy, particularly if these drugs are also included in the new drug regimen, will increase the risk of treatment failure. One dose of nevirapine during deliver had initially been shown to cause resistance in about 20% of women,[55,56] but more recently researchers have shown using real-time PCR analysis that an additional 40% of women with previously undetectable resistance also had the Lys103Asn mutation.[57] Therefore, women should always be asked about the use of nevirapine during pregnancy.

A related question is whether a non-nucleoside reverse transcriptase inhibitor regimen has been interrupted outside efforts to prevent vertical transmission (eg, because of side-effects or inability to pay for the drugs). Stopping the combination of stavudine, lamivudine, and nevirapine without continuing the nucleoside reverse transcriptase inhibitor for at least 7 days may lead to resistance because of the long half-life of nevirapine.[58]

Assess the quality of the HAART regimen and concomitant medication

A regimen containing three nucleoside analogues may not be sufficiently effective[59–62]—eg, one containing efavirenz, tenofovir, and didanosine is associated with treatment failure.[63,64] In addition, the use of concomitant medication—eg, rifampicin, carbamazepine, or phenytoin—might reduce antiretroviral drug levels.[65]

Assess adherence to treatment

A clear association between extent of adherence and treatment outcome has been documented in several studies.[66–73] Assessing a patient's treatment adherence is believed to be difficult.[69] Our experience in Uganda is that obtaining reliable information about adherence is possible for most patients, provided that experienced, non-judgmental counsellors establish a good relationship with the patient. In Uganda, the visual analogue scale has proven to be a simple and useful method to evaluate adherence.[74] Uncomplicated treatment cards, similar to those used for tuberculosis patients, can be used to improve and assess a patient's adherence to treatment, and new devices such as computerised pill boxes, allowing remote monitoring by wireless technology of adherence by transmitting information about the time the pill box is opened, could be useful tools for the future.

Adequately monitoring treatment adherence could help prevent treatment failure and the development of drug resistance. A patient who meets one of the adherence criteria for treatment failure will most probably have a detectable level of viral load. However, this does not automatically mean that the patient is infected with a drug-resistant virus. If there are no other criteria suggesting treatment failure, improving adherence with the current treatment regimen should be tried before considering a change in therapy.

Assess clinical symptom development and laboratory tests results

Viral load testing should be considered when patients have mixed indicators that suggest both treatment

failure and success. However, treatment providers must always take into account their patients' treatment options. For many patients in resource-poor settings, these options are limited. If a patient without access to second-line antiretroviral therapy has clinical symptoms or signs suggesting treatment failure, they have only two options—continue first-line therapy or stop it. If the patient's decision is made irrespective of the result of a viral load test, then such a test is a waste of resources.

If treatment failure is suspected, there is little sense in maintaining the antiretroviral regimen in view of the high cost of treatment, the public health risk of transmitting the resistant virus to others, and the risk to the patient of accumulating genetic mutations that might make future treatment less likely to work. On the other hand, even a failing antiretroviral regimen, particularly one containing nucleoside analogues and a protease inhibitor, can still have a beneficial effect on a patient's immune system.[75] This might be one reason to continue a failing antiretroviral regimen if no other treatment option is available. If this option is chosen, at each visit health-care workers must reinforce the use of condoms to prevent the transmission of resistant virus.

The model's potential effect

With an optimal non-nucleoside reverse transcriptase inhibitor first-line regimen and a good strategy to obtain the best adherence to treatment, we expect that at least 80% of people will have an undetectable viral load at 12 months, similar to the rates in industrialised countries[51] and in reports from resource-poor countries.[76]

If we assume that our model could identify 90% of the 80% of patients on a successful treatment and 15% of the 20% patients on a failing regimen, viral load testing will be needed to guide therapeutic decisions in only 15% of patients. Thus, only a few samples would need to be sent to a laboratory. If the medical infrastructure in poor countries could provide reference laboratories and new, simple, and inexpensive but reliable ways to send samples to these laboratories, the application of our model could greatly reduce the number of expensive laboratory tests needed, without compromising antiretroviral treatment.

Whether or not our model will achieve its potential will need to be proven in prospective studies in a variety of settings and by different categories of health-care workers.

Conclusion

Our model aims to improve the sensitivity, specificity, and positive predictive value of the WHO's criteria for treatment failure.

Although our model is based mainly on experience and few published data, we felt it important to publish our proposal since a growing number of people in poor countries are on ineffective antiretroviral regimens, and doctors in these countries find it difficult to diagnose treatment failure.

The model's effectiveness will rely on health-care workers having good clinical skills. These workers might need training to improve clinical skills and the ability to interpret supporting laboratory results.

Evaluation of adherence has a key role in the model. antiretroviral programmes must include strategies to obtain optimal treatment adherence to prevent the emergence of drug-resistance. It is probably more cost-effective to fund training programmes for treatment counsellors and to support a multidisciplinary medical staff dedicated to following up each patient than to spend money on expensive laboratory testing.

This model could become a valuable tool during the scaling-up of antiretroviral treatment in developing countries, but it needs further study to determine whether this strategy will be useful in resource-limited settings.

Conflicts of interest

We declare that we have no conflicts of interest.

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