

Indicaciones precisas para genotipificación y/o fenotipificación (Indications for Genotypic and/or Phenotypic Analysis of VIH)

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The dynamics of HIV replication and the high frequency of point mutations have proven to be the Achilles heel of HAART. These random mutations are the basis of therapy failure, regardless of the factors initially leading to failure – non-adherence, poor absorption, inadequate potency of the regimen and so forth. The technology to monitor and identify resistance has become available for clinicians during the past four years and is rapidly becoming an indispensable tool in the management of antiretroviral therapy.

Resistance Tests

There are two basic approaches to study resistance, **genotypic** and **phenotypic** analysis. Genotypic analysis identifies mutations of nucleotides that code for the amino acids that make up the reverse transcriptase and the protease enzymes. Phenotypic analysis measures the susceptibility of the HIV to a given drug. Genotypic analysis is relatively rapid and less expensive while phenotypic analysis requires greater time and is relatively expensive (both methods are expensive relative to T-lymphocyte subset analysis and HIV RNA quantitative assay). The genotypic and phenotypic assays have been supplanted with a “**virtual phenotypic assay**” and a **recombinant phenotypic assay**. A virtual phenotypic assay is actually a genotypic assay with the results linked to a data bank of thousands of specimens tested by both genotypic and phenotypic assays simultaneously. Based on the pattern of mutations detected, the results are linked to the appropriate historical phenotypic results and the report generated. The recombinant phenotypic assay is a true phenotype, but with the portion of the HIV genome encoding for reverse transcriptase and protease implanted in a stock laboratory strain of HIV that is actually used in the assay. This technique yields results more rapidly (~ 14 days) than the traditional phenotypic assay and at lesser expense.

Caveats

These assays theoretically should provide guidance to the selection of drugs and the results of clinical trials are gradually accumulating to define how best to use them. There are difficulties, however, with the interpretation of the resistance assays.

First, in any one patient there may be multiple daughter strains of the original HIV strain, some with mutations, some without. These assays generally detect the predominate family and may

overlook a minority population. Thus, resistant virus may be undetected. Second, the assay may not show whether multiple mutations are present on the same isolate or scattered among several isolates. Third, the first generation of genotypic assays require a viral load of at least 1000 copies and generally 10,000 copies to generate results. The phenotypic assays may yield results with viral loads as low as 500 copies/ml. At low viral loads, however, the laboratory may have difficulty providing results. Forth, strains of HIV with resistance to antiretroviral drugs given in the past are “archived” in a latent stage and will be undetected. For patients who have been treated with multiple regimens in the past, resistance testing may falsely suggest susceptibility because these archived or latent HIV are overlooked. The tendency for the original wild type HIV to overgrow or overgrow the less fit resistant HIV once antiretrovirals are removed means that the patient must be on therapy at the time of testing. Related to the issue of viral “fitness” is the reduced ability of some resistant strains to reproduce as rapidly as the wild type HIV. Thus, even though resistance develops to the antiretroviral regimen, the regimen may still be of clinical benefit. Genotypic analysis cannot detect this phenomena. And it is now clear that the patterns of genotypic mutations associated with loss of susceptibility to protease inhibitors may be overcome by combining ritonavir with other protease inhibitors and achieving higher serum concentrations throughout the day.

Clinical validation

In spite of these shortcomings, resistance testing eventually should prove useful in the management of all patients. And prospective clinical trials demonstrating their utility have been reported and others are ongoing. Two clinical trials, the Viradep and the GART studies, have demonstrated that genotypic resistance typing have clinical utility and lead to better choices of drugs for patients failing therapy. One of two prospective studies of phenotypic assays have confirmed a benefit whereas the other did not. The study which found no benefit had enrolled heavily treated patients and underscores the difficulty of finding drugs with activity against the infecting HIV for this population.

Current Recommendations for Resistance Testing

Because of the expense and the limitations of our knowledge regarding interpretation of results, resistance testing is currently recommended for patients who are failing therapy, with failure defined as incomplete suppression of HIV-1 RNA copies or a rebound after suppression to non-detectable levels. Resistance testing is not recommended for treatment naïve persons with chronic HIV infection because of the low level of resistance in this population. The use in persons with acute HIV infection is less well defined. If there is reason to suspect transmission of a resistant strain of virus, resistance testing. As the HIV epidemic matures and treatment is more prevalent, the transmission of resistant HIV will become an increasingly common problem.

Which assay to use will depend upon many factors. The genotypic assays require interpretation and some of the laboratories currently performing the assays are not very helpful with the interpretation. This should change. The phenotypic assays are arguably the better test but so expensive and take so long for results to be returned that they are unavailable for many clinicians. Hopefully, the cost and time for results will decrease. The laboratory and clinical experience with resistance testing is rapidly growing and physicians can expect the field to change considerably over the next several years.

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