

Analysis of Resistance Testing in South Trinidad

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ABSTRACT

The introduction of antiretroviral therapy in Trinidad and Tobago in the 1980s has resulted in a decrease in mortality of HIV-infected persons. Poor adherence to antiretroviral therapy (ART) has resulted in the development of multidrug resistant HIV. Resistance testing done on 40 samples showed that 64.8% of patients had K103 mutation, 75.6% of patients had M184 mutations and 62% of patients showed resistance to tenofovir suggesting that the K65R mutation was highly likely to be present. There was reduced activity to the protease inhibitors; no resistance was found to the protease inhibitor, darunavir. Thus, there is a need for salvage therapy to be introduced which will result in virologic suppression and potentially stop the spread of multidrug resistant HIV. Darunavir, a new generation protease inhibitor, is an essential part of salvage therapy and needs to be introduced into the national formulary.

Key words: HIV, resistance testing

Análisis de Prueba de Resistencia en Trinidad Sur

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RESUMEN

La introducción de la terapia antiretroviral en Trinidad y Tobago en la década de 1980, ha producido una disminución en la mortalidad de personas infectadas por el VIH. La adhesión pobre a la terapia antiretroviral (TAR) ha conducido al desarrollo de una variedad de VIH resistente a las multidroga. Las pruebas de resistencia realizadas a 40 muestras mostró que el 64.8% de los pacientes tenían mutación K103, 75.6% de los pacientes tenían mutaciones M184, y 62% de pacientes mostraron resistencia al tenofovir, lo que indica una alta probabilidad de mutación K65R. Había actividad reducida respecto a los inhibidores de la proteasa; mientras que no se halló ninguna resistencia en el inhibidor de la proteasa, darunavir. Así, hay necesidad de introducir una terapia de salvamento que produzca una supresión virológica y potencialmente detenga la diseminación del VIH resistente a las multidroga. El darunavir – inhibidor de nueva generación frente a la proteasa – es una parte fundamental de la terapia de salvamento y necesita ser introducido en el formulario nacional.

Palabras claves: VIH, prueba de resistencia

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INTRODUCTION

Trinidad and Tobago, with a population of 1.3 million, has a HIV prevalence of 1.3%. The treatment of patients infected with the Human Immunodeficiency Virus (HIV) has advanced from monotherapy to triple or quadruple antiretroviral therapy (ART) in an attempt to achieve virologic suppression. This has traditionally been done without the use

of resistance testing data as a guide to antiretroviral management. At the San Fernando General Hospital, the major tertiary care health facility in South Trinidad, 937 patients who are infected with the HIV virus are actively followed. The HIV outpatient clinic has an adherence rate of approximately 90%, but there exists a subset of patients who have been persistently failing their antiretroviral therapy (ART) as per national guidelines to treatment. In 2009, approximately 37 patients were selected for resistance testing based on decreasing CD4 count and persistent viral load above 1000 copies/ml, the resistance data obtained from these patients was then used as a guide to the initiation of potent ART in this subset of patients.

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SUBJECTS AND METHOD:

Patients who were deemed to be failing second line ART as per national guidelines were identified and blood samples were taken for resistance testing. Blood samples were taken from 40 patients and sent to Puerto Rico for molecular analysis. Samples were transported as *per* strict protocol for the transportation of biohazard material with all shipping protocols observed under IATTA certification. The molecular microbiology laboratory is one of two WHO laboratories in the region certified to do HIV resistance testing. There are two main types of genotypic resistance testing, sequencing assays and point mutation assays. Sequencing assays scan the complete sequence of a gene whereas point mutation assays look for mutations at specific locations or codons in the gene sequence. Though these assays differ in how the genetic structure is analysed, they both start by using polymerase chain reaction (PCR) technology to amplify (copy) the viral RNA or DNA present in a patient’s blood sample. The method used at this laboratory is the point mutation assay.

Results:

Twenty-four (64.8%) patients had K103 mutation shown in Figures 1 and 2 while 28 (75.6%) patients had M184 mutations shown in Figures 1 and 2. Twenty-three (62%) patients

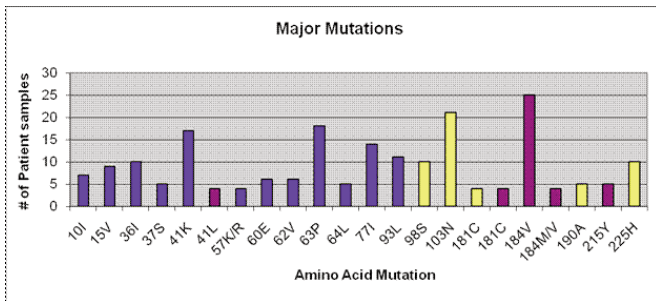


Fig. 1: Major Mutations: 64.8% show K103 mutations and 75.6%, M184 mutations.

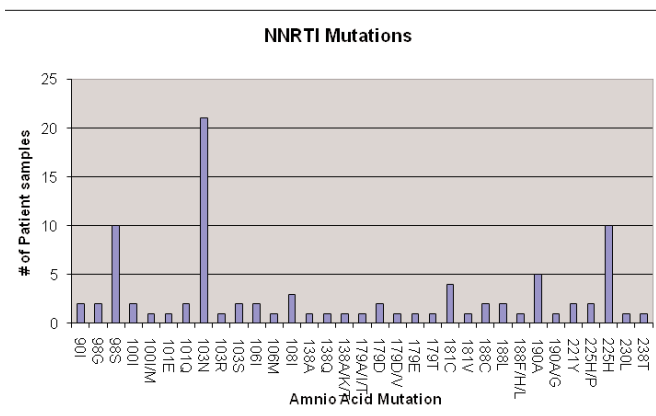


Fig. 2: Non-nucleoside reverse transcriptase inhibitor mutations: K103 mutation identified in 64.8% of patients.

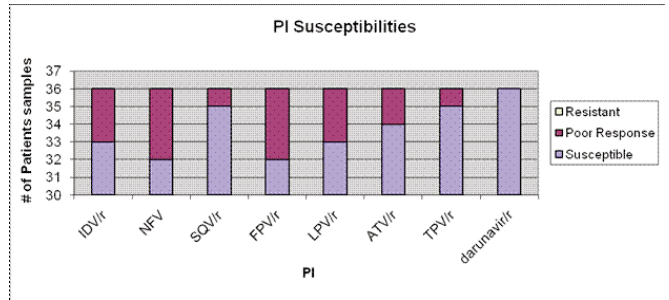


Fig. 3: Protease Inhibitors’ Susceptibilities: No resistance but reduced activity.

showed resistance to tenofovir shown in Figures 1 and 2 which means that K65R mutation was highly likely to be present. No protease inhibitor mutations were noted in this study but there exists reduced activity to the protease inhibitors as seen in Figure 3. No resistance was found to the protease inhibitor, Darunavir.

DISCUSSION

It is accepted that patients failing first and second line ART will benefit from salvage and deep salvage therapy (1, 2). This has been demonstrated in numerous international multi-centre trials of treatment-experienced-patients with triple class resistance (1, 2). Raltegravir, a new drug belonging to the class of integrase inhibitors has been approved to be used in treatment experienced patients who are failing ART (3). The BENCHMARK trial showed a definite benefit in using Raltegravir with an optimal backbone; 62% of patients did achieve virologic suppression (3).

Darunavir, a new generation protease inhibitor, has a high genetic barrier to resistance and is well tolerated by patients (4). This high barrier to resistance has lead to its use as a major drug in salvage therapy (4). The POWER trial demonstrated that darunavir boosted with ritonavir was effective in achieving virologic suppression in treatment-experienced-patients who were previously failing therapy (5). In the DUET trial, Etravirine (a new drug in the NNRTI class of drugs also has a high genetic barrier to resistance) in combination with ritonavir, darunavir was shown to be very effective in achieving virologic suppression in patients who are treatment experienced and are failing therapy and this makes it a potent drug in salvage therapy (5). In the DUET trial, 60% of patients who were previously failing second line therapy achieved virologic suppression (5).

From the analysis of the resistance testing in the present study, it is clear that there is a role for the introduction

of medications for use as salvage therapy into the national formulary. There exists universal sensitivity to the protease Inhibitors; thus this will be part of the Optimized Backbone therapy used in salvage therapy. From figure 2, one can appreciate an accumulation of Thymidine Analogue Mutations (TAM) which only increases the likelihood of decreased activity to drugs in the NNRTI class. Thus, it is clear that these patients will benefit from a NNRTI with a high genetic barrier to resistance. Etravirine, darunavir boosted with ritonavir and raltegravir should be key components to salvage therapy in these patients and with this evidence, one can create a case for the introduction of these drugs into the national formulary. Both darunavir and etravirine have a high genetic barrier to resistance (6, 7) and will be key components to salvage therapy. Although resistance testing should be done on all newly diagnosed patients prior to initiating ART, it is not practical due to cost. However for patients failing ART, resistance testing should be routinely

done and the data obtained should be used to provide clinical guidance to ART.

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