Brief report

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Resistance to HIV integrase strand transfer inhibitors in Argentina: first interim survey

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ABSTRACT

Objectives. No data on resistance to HIV integrase strand transfer inhibitors (InSTIs) in Argentina are available as access to these drugs and to integrase genotypic resistance test is limited. We aimed to evaluate the clinical profile of patients who underwent an integrase genotypic resistance test, prevalence of InSTI resistance mutations and predicted efficacy of raltegravir, elvitegravir and dolutegravir in our country.

Patients and methods. Retrospective multicentric pilot survey from January 2011 to November 2017 of InSTI-failing patients assisted at two private and one public healthcare institutions located in Buenos Aires city, Argentina.

Results. Sixty seven patients were included. Patients had a median of 5 (4-7) prior treatments. All patients had InSTI-containing regimens (median exposure of 22.5 months); 94% were under raltegravir therapy and 71.9% had InSTI-resistance mutations. Predominant major mutations were N155H (35.1%), Q148H/R (15.8%) and G140A/S (14%). Considering Stanford HIVdb program, extremely low and identical activity of raltegravir and elvitegravir was described while dolutegravir remained either partially or fully active in 97.7% of patients.

Conclusions. Integrase resistance test was prescribed almost exclusively in heavily pretrated raltegravir-exposed patients. The three main mutational pathways were described, with a predominance of N155H. Despite almost null susceptibility and extensive cross resistance was shown among raltegravir and elvitegravir, dolutegravir remains active in the majority of patients.

Keywords: HIV infection, drug resistance, integrase inhibitors

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Resistencia a inhibidores de la integrasa en Argentina: primera encuesta interina

RESUMEN

Objetivos. No hay datos disponibles sobre resistencia a inhibidores de la integrasa (INIs) en Argentina, ya que el acceso a estas drogas y al estudio de resistencia genotípica es limitado. Nuestro objetivo fue evaluar el perfil clínico de los pacientes a los que se les indicó un estudio de resistencia genotípico de integrasa, la prevalencia de mutaciones de resistencia INIs y la predicción de eficacia para raltegravir, elvitegravir y dolutegravir en nuestro país.

Pacientes y métodos. Encuesta piloto retrospectiva multicéntrica, enero de 2011 a noviembre de 2017, de pacientes con fallo virológico a INIs asistidos en dos instituciones de salud privadas y una pública en Buenos Aires, Argentina.

Resultados. Se incluyeron 67 pacientes, con una mediana de 5 (4-7) tratamientos previos. Todos tenían regímenes con INIs (exposición media de 22,5 meses); el 94% estaba recibiendo raltegravir y el 71,9% tenía mutaciones de resistencia a INIs. Las mutaciones primarias predominantes fueron N155H (35,1%), Q148H/R (15,8%) y G140A/S (14%). Considerando el programa de HIVdb de la Universidad de Stanford, se describió una actividad extremadamente baja e idéntica para raltegravir y elvitegravir, mientras que dolutegravir se mantuvo parcial o totalmente activo en el 97,7% de los pacientes.

Conclusiones. La prueba de resistencia a la integrasa se indicó casi exclusivamente en pacientes experimentados en tratamiento antirretroviral y expuestos a raltegravir. Se describieron las vías mutacionales principales, con predominio de N155H. Pese a la susceptibilidad casi nula y extensa resistencias cruzada entre raltegravir y elvitegravir, dolutegravir permaneció activo en la mayoría de los pacientes.

Palabras clave: infección por VIH, resistencia a drogas, inhibidores de integrasa

INTRODUCTION

Integrase strand transfer inhibitors (InSTI)-based antiretroviral regimens are highly efficacious at suppression of HIV replication and are recommended for initiation of HIV therapy, as for subsequent regimens in different guidelines. InSTIs were found to be highly potent in the clinic; their use resulted in faster declines in viral load upon treatment initiation than had previously been observed [1]. Despite this, Latin America had a slow process of inclusion of the InSTI in its guidelines. Countries with gradual adoption of InSTI for first line therapy, mostly raltegravir (RAL), include Chile, Mexico, Colombia, Venezuela, and Argentina. Brazil in 2017 incorporates them in their preferred options supported with a large-scale purchase of dolutegravir (DTG) [2]. Overall, the use of InSTI in Latin America remains very limited and scarce in real practice because, mainly, of price barriers. In this context, in Argentina, access to InSTI-based antiretroviral therapy has been historically restricted to pretreated patients, mainly in the context of virologic failure or comorbidity in which more accessible non-nucleoside reverse transcriptase inhibitor or protease inhibitor-based antiretroviral therapy cannot be prescribed.

Despite its potency and favorable virological profile, HIV can become resistant against InSTIs through the emergence of mutations within the integrase coding region. Different pathways against first-generation InSTIs were identified whose primary mutations include the substitutions N155H, Q148K/R/H, and Y143R/C [3–10].

In Argentina, a considerable prevalence of acquired and transmitted drug resistance has been described [11–14]. However, these epidemiological surveys included exclusively sequencing of protease and retrotranscriptase coding regions as access to HIV-1 integrase resistance sequencing has been, until recently, extremely limited.

Considering that information regarding prevalence of resistance is a key element in order to better define the public health approach to antiretroviral drug access, we aimed to describe the clinical and virological profile of patients who underwent an integrase resistance test and the prevalence of InSTI resistance mutations in Buenos Aires city, Argentina.

PATIENTS AND METHODS

We underwent a retrospective multicentric pilot survey from January 2011 to November 2017 of InSTI-failing patients assisted at two private and one public healthcare institutions located in Buenos Aires city, Argentina. Virologic Failure was defined as the inability to achieve or maintain suppression of viral replication to an HIV RNA level <200 copies/mL [1]. RNA was extracted from plasma samples of InSTI-exposed patients by the automated system MagnaPure Compact Nucleic Acid Isolation kit Large Volume, Roche (Mannheim, Germany), amplified by a validated in house RT-PCR of the entire integrase HIV-1 gene (corresponding to 288 amino acids) [15,16] and

sequenced by Sanger sequencing in ABI 3500 Applied Biosystems™. The ChromasPro (version 2.0.0) and RECall (beta v3.04) programs were used to assemble and edit sequences. The consensus sequence was interpreted using the Stanford University HIV Drug Resistance Database. Integrase resistance mutations were classified as "major" or "accessory" and, considering the genotype interpretation system (GIS) of Stanford HIVdb program (version 8.4), the predicted efficacy of RAL, DTG and elvitegravir (EVG) was classified within five categories: from susceptible to high-level resistance. The GIS categories "susceptible" and "potential low level resistance" were grouped together as "susceptible", whereas "low level resistance" and "intermediate resistance" were grouped as "intermediate" and the remaining as "resistant".

RESULTS

A total of 67 patients were included. Of them, 64.2% were male. The median (interquartile range IQR) of age, pre-genotype viral load, and CD4 T-cell count were: 43 (23-52) years, 4465 copies/mL (859-27812) and 306/µL (153-499), respectively. Predominant HIV subtype was B/F (52.4%). Most of the patients had been exposed to several antiretroviral regimens (with most of the changes attributable to virologic failure) and a considerable proportion of them had HIV acquired perinatally. All of them had ongoing InSTI-containing regimens with a median of exposure of 22.5 (10-51) months; 94% were under RAL therapy (only one patient was under DTG therapy, but had prior exposure to RAL). Median time from diagnosis of virologic failure to genotype was 6 (3-13) months. Most frequent accompanying antiretrovirals included a boosted protease inhibitor + nucleos(t)ides reverse transcriptase inhibitors (NRTIs) or NRTIs exclusively. Resistance to other antiretroviral drug classes was highly prevalent. Patients had a median of 3 (0-9) and 4 (1-8) mutations in protease and retrotranscriptase genes, respectively. Ninety seven percent of patients had irregular adherence to the current antiretroviral regimen. A detail of the clinical and immunovirological profile of the patients is shown in table 1. Integrase gene sequencing was successful in 57 of them (85.1%): 71.9% (n = 41) had resistance mutations with a median of 2 per patient (50.8% had combined mutations, n = 29). Predominant major integrase resistance mutations were N155H (35.1%), Q148H/R (15.8%) and G140A/S (14%). Predominant combinations of mutations were 140A/S + Q148H/R and N155H + G163K/R. A detail of major, accessory and combined mutations is shown in table 2. We found no association between development or integrase resistance mutations and time on virologic failure, time of exposure to raltegravir and number of prior antiretroviral regimens.

Considering Stanford HIVdb program GIS modified categories for each mutation profile, RAL and EVG were classed as susceptible, intermediate resistance and resistant in 2.4%, 21.9% and 75.6% of patients, respectively. For DTG, 68.4%, 29.3%, 2.4% of mutational profiles were classified as susceptible, intermediate resistance and resistant, respectively.

Table 1	Demographic and immunovirological profile of 67 HIV-infected patients who underwent an integrase resistance genotypic test in Buenos Aires, Argentina. Values are number (percentages) unless otherwise stated.

	N (%)
Male sex	43 (64.2)
Mode of infection	
Sexual	45 (67.2)
Perinatal	18 (26.9)
CDC C3 category	41 (61.2)
Age, median (IQR)	43 years (26-52)
Viral load, median (IQR)	4465 copies/ml (859-27812)
CD4 T-cell count, median (IQR)	307 cells/µL (153-499)
HIV-1 subtype (n = 50)	
B/F	27 (54)
B	23 (46)
Number of prior antiretroviral regimens, median (IQR)	5 (4-7)
Concomitant resistance in retrotranscriptase or PR genes (n = 62)	51 (82.3)
Single NRTI resistance	8 (13)
Single NNRTI resistance	5 (8)
Single PR resistance	6 (9.5)
Resistance to 2 drug classes[a]	13 (30)
Resistance to 3 drug classes[a]	15 (24)
Accompanying ART (n = 54)	
NRTIs only	13 (24)
Boosted PI + NRTIs	17 (31.5)
Boosted PI + NRTIs + other drug[b]	12 (22.2)
Boosted PI + other drug[b]	4 (7.4)
Boosted PI only	2 (3.7)
Other	6 (11)

ART: antiretroviral therapy; IQR: interquartile range; NRTI: nucleos(t)ide reverse transcriptase inhibitor; NNRTI: non-nucleos(t)ide reverse transcriptase inhibitor; PR: protease
[a]Excluding integrase mutations; [b]Either NNRTIs or maraviroc

DISCUSSION

We present the first interim epidemiological study of resistance to InSTIs in Argentina and one of the first surveys of its kind in Latin America. Several aspects of this study should be highlighted.

The clinical profile of the patients reflects that access to InSTIs (almost exclusively RAL) has been limited to heavily pretreated patients, with limited therapeutic options, as shown by the high number of prior antiretroviral regimens and the prevalence of concomitant resistance to other drug classes. In this context it not surprising that, in this first "wave" of integrase failing patients, a considerable proportion of them

had HIV acquired perinatally and advanced disease. It is expected, with evolving guidelines in Latin America, that naive patients (or, at least, less exposed patients) will have access to first and second generation InSTIs in the region. With this perspective, a change may be expected in the clinical and mutational profile of patients failing InSTIs in the upcoming years.

In this cohort of RAL-exposed failing patients, integrase resistance mutations were detected in the majority of them, reflecting the low genetic barrier of this first generation drug. Fourati et al., in a French national study of RAL-experienced HIV-1 infected patients, described that 39% of viruses of patients experiencing virological failure harbored at least one major InSTI resistance mutation. In this dataset, Q148 and N155 pathways predominated (observed in 15.4% and 19.1% of patients, respectively), whereas Y143 was detected in 6.7%. This prevalence is comparable to the one observed in our cohort for Q148 and Y143 pathways, but a higher prevalence of N155 substitutions was described in our population (35.1%) [17]. Further research is needed in order to define factors associated with selection of different mutational pathways among Latin American population. Considering RAL´s low genetic barrier, the irregular adherence that the vast majority of patients had to the therapy, in the context of high prevalence of, at least, some level of resistance to accompanying drugs may contributed to development of InSTI mutations. Of note, median time of exposure to RAL before integrase genotype in our study almost doubled the one documented in the French survey (22.5 vs 11 months). This reflects logistical barriers to the access to integrase genotype at the local level (median of 6 months) rather than a longer time to virologic failure on RAL-containing therapy in our setting. It is expected, in the upcoming years, that integrase genotype will be more accessible in Buenos Aires city (as in the rest of Argentina), facilitating an opportune evaluation of InSTI-failing patients.

Considering Stanford HIVdb program GIS, extremely low and identical activity of RAL and EVG is described, confirming extensive cross-resistance in our population. In contrast, second generation InSTI DTG remains either partially or fully active in 97.7% of patients, constituting and extremely important therapeutic option for future regimens in this heavily pretreated patients as shown in VIKING trials [18, 19]. Of note, in our survey, R263K signature mutation that affects the efficacy of DTG [20] was not observed. As high-level DTG resistance requires multiple first-generation InSTI-resistance mutations, a timely interruption of RAL would prevent accumulation of resistance and should be considered in order to maximize the effect of DTG in our population [3, 17]. In this context, access

Table 2	Prevalence of individual major and accessory integrase resistance mutations and combination of them in a cohort of raltegravir failing patients in Buenos Aires, Argentina (N = 57).

	N (%)
Major mutations	
N155H	20 (35.1)
Q148HR	9 (15.8)
G140AS	8 (14)
E138A	3 (5.3)
Y143R	3 (5.3)
T66A	1 (1.8)
E92Q	1 (1.8)
Accesory mutations	
G163KR	18 (31.6)
T97A	11 (19.3)
V151I	8 (14)
L74IM	5 (8.8)
E157Q	2 (3.5)
A128T	1 (1.8)
Combined mutations	
G190A/S + Q148H/R	6 (10.5)
N155H + G163K/R	4 (7)
A128T + V151I + N155H	1 (1.7)
E138A/E + G140A/S + Q148H/R	1 (1.7)
E92A/E + T97A + V151I + N155H	1 (1.7)
L68V + L74M + E92Q + T97A + N155H	1 (1.7)
L74I + V151I + N155H + G163R	1 (1.7)
L74I + Y143R + V151I + N155H	1 (1.7)
L74M + T97A + Y143R + G163R	1 (1.7)
Q148H/R + G163R	1 (1.7)
T66A/T + L74I + T97A + E138A/E	1 (1.7)
T79A + G163R	1 (1.7)
T97A + E138A/E + Y143R + G163K	1 (1.7)
T97A + G140AS + Q148HR	1 (1.7)
T97A + G163R	1 (1.7)
T97A + N155H	1 (1.7)
T97A + N155H + G163R	1 (1.7)
V151I + N155H + E157Q	1 (1.7)
V151I + N155H + E157Q +G163R	1 (1.7)
V151I + N155H	1 (1.7)

to DTG should be warranted in our setting.

In conclusion, this first interim survey of resistance to InSTIs in Argentina reflects that, to date, integrase resistance test was prescribed almost exclusively in heavily pretrated RAL-exposed patients. The three main mutational pathways were described, with a clear predominance of N155H. Despite almost null susceptibility and extensive cross resistance was shown among RAL and EVG, DTG remains active in the vast majority of patients. Further monitoring is needed to describe evolving trends in the clinical, virological and mutational profile of InSTI-failing patients as long as first and second generation InSTIs become more accessible in Argentina as other Latin American countries in the near future.

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CONFLICT OF INTERESTS

The authors declare that they have no conflict of interest

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