

Integrase Strand Transfer Inhibitor Resistance in Integrase Strand Transfer Inhibitor-Naive Persons

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Abstract

There has been no systematic review of the prevalence of transmitted integrase strand transfer inhibitor (INSTI) resistance. We systematically searched the English-language PubMed database and GenBank to identify studies published since 2010 reporting 50 or more INSTI-naive HIV-1-infected adults undergoing integrase genotyping. We extracted data related to country, sample year, specimen type, sequencing method, and subtype. For studies with sequences in GenBank, we determined the prevalence of three categories of INSTI-associated resistance mutations: (1) nonpolymorphic INSTI-selected drug resistance mutations (DRMs) that we refer to as surveillance DRMs; (2) rarely selected nonpolymorphic INSTI-associated DRMs; and (3) common polymorphic accessory INSTI-selected DRMs. A total of 103 studies met inclusion criteria including 75 studies in GenBank containing integrase sequences from 16,481 INSTI-naive persons. The median sample year was 2013 (interquartile range: 2008–2014). The prevalence of INSTI surveillance DRMs, rarely selected DRMs, and common polymorphic accessory INSTI-selected DRMs were 0.5%, 0.8%, and 6.2%, respectively. There was no association between the presence of nonpolymorphic surveillance DRM and region, sample year, or subtype. Two surveillance DRMs, E138K and R263K occurred in 0.15% and 0.10% of naive sequences, respectively. Several lines of evidence suggested that the 0.5% prevalence of surveillance DRMs partly reflects the cumulative natural occurrence of these mutations in the absence of selective drug pressure. There was an unexplained temporal increase in the proportion of sequences with polymorphic accessory mutations. The prevalence of INSTI-associated surveillance DRMs is low even in regions where INSTIs have been a major component of antiretroviral therapy for several years. The presence of INSTI-associated surveillance DRMs in INSTI-naive persons likely results from actual cases of transmitted INSTI resistance and from a low background level reflecting the cumulative rare natural occurrence of several nonpolymorphic mutations.

Keywords: HIV-1, antiretroviral therapy, drug resistance, surveillance, integrase

Introduction

INTEGRASE STRAND TRANSFER INHIBITORS (INSTIs) have been increasingly used during the past 14 years since the approval of raltegravir in 2006 followed by the subsequent approvals of elvitegravir in 2011, dolutegravir in 2014, and bictegravir in 2018. INSTIs have been the most commonly preferred antiretrovirals for use in combination with two nucleoside reverse transcriptase inhibitors (RTIs) in the United States since 2014¹ and worldwide since 2018.² Although there has been no systematic review of the topic, the prevalence of INSTI-associated transmitted drug resistance (TDR) is considered to be low and routine genotypic resistance testing before starting an INSTI is not routinely recommended.^{3,4} We performed a systematic review of English-language

studies in PubMed and of submissions to the GenBank sequence database to quantify the extent to which INSTI-associated drug-resistance mutations (DRMs) have been reported in previously INSTI-naive populations.

Methods

Search criteria

Data relevant to INSTI TDR prevalence were obtained from systematic searches of the PubMed literature database and the GenBank sequence database. The PubMed search, last updated August 15, 2020, used the search terms “HIV” and “integrase” to identify studies published since January 1, 2010. Retrieved studies were reviewed in two stages. First, titles and/or abstracts were assessed to identify studies describing

integrase sequencing of samples obtained from HIV-1-infected persons. Following the title/abstract review, complete publications were reviewed to identify studies reporting integrase sequences from ≥ 50 INSTI-naive adult persons.

The GenBank search began with a BLAST search of the v. 239 database (released 2020-08-15) using the HIV-1 subtype B consensus integrase sequence. Retrieved HIV-1 group M integrase sequences with the same GenBank “Author” and “Title” fields were grouped into submission sets. GenBank annotations and associated publications were reviewed to identify submission sets containing integrase sequences encompassing positions 66–263 obtained from ≥ 50 INSTI-naive HIV-1-infected persons.

Studies for which the sequence data were not available in GenBank and that did not specify a list of DRMs used to define INSTI resistance were excluded.

Data extracted

For each study, the following information was collected from its publication and/or GenBank annotations: (1) country or countries of virus isolation; (2) year or years of virus isolation; (3) recency of infection; (4) proportion of persons who were antiretroviral therapy (ART)-naive and who were ART-experienced but INSTI-naive; (5) proportion of persons undergoing integrase sequencing relative to those undergoing RT and/or protease sequencing; (6) specimen: plasma versus peripheral blood mononuclear cells (PBMCs); (7) sequencing method: Sanger dideoxynucleoside sequencing versus next-generation sequencing (NGS); (8) subtype distribution; and (9) mutations considered by the authors to be associated with INSTI-associated DRMs. If a range of sample years was provided for a study, all the samples in the study were assigned the median of the range. For three studies for which sample years were not provided, the samples were assigned the publication year minus 4 years, as this was the median time between sample year and publication year for studies with known sample years. NGS studies were excluded if the threshold for mutation detection was not reported or was either 15% or 20%, levels that are $>99\%$ concordant with Sanger sequencing.⁵

Analysis

The studies with sequences in GenBank were analyzed using the Calibrated Population Resistance tool, which determined the proportion of persons containing sequences with one or more of a list of DRMs we recently developed for the purposes of drug resistance surveillance: T66A/I/K, E92G/Q, G118R, F121Y, E138A/K/T, G140A/C/S, Y143C/H/R/S, S147G, Q148H/R/K, N155H, S230R, and R263K.⁶ This list of 24 INSTI surveillance DRMs was derived from an analysis of sequences from $\sim 17,300$ INSTI-naive and 2,450 INSTI-experienced persons using an approach similar to the one used to develop the widely used WHO list of RTI- and protease inhibitor (PI)-associated surveillance DRMs.⁷ In brief, we analyzed a list of 59 DRMs identified on one or more of four expert lists of INSTI-associated DRMs and identified 24 DRMs that were nonpolymorphic in INSTI-naive persons ($<0.2\%$ overall naive prevalence and $<0.5\%$ prevalence in the eight most common subtypes) and that occurred with significantly increased frequency in isolates from persons receiving an INSTI-containing regimen.⁶

Transmitted RT and/or protease resistance was defined as the presence of one or more WHO surveillance DRMs.⁷ For sequences in GenBank, those containing two or more signature APOBEC mutations were excluded from analysis.⁸ For the non-GenBank studies, we determined the proportion of persons containing sequences with one or more INSTI surveillance DRMs from the author reported mutations.

In addition to determining TDR prevalence using the INSTI surveillance DRMs, we determined the prevalence of two additional categories of mutations that were also defined in the publication that led to the development of the INSTI surveillance DRMs: (1) nonpolymorphic DRMs that were not classified as surveillance DRMs because they have rarely been selected in persons receiving an INSTI including A49G, H51Y, L74F, V75I, E92V, Q95K, H114Y, Y143A/G/K, P145S, Q146K/L/I/P/R, Q148E/G/N, G149A, V151L/A, S153F/Y, N155S/T, and S230G; and (2) polymorphic INSTI-selected DRMs including A49P, L74M, T97A, E157Q, G163R/K, and D232N.⁶ L74I was not included in this list because, although it is polymorphic, it was not found to be selected by INSTIs.⁶

Samples from different studies were pooled and a generalized linear-mixed model was used to assess the yearly change in the odds of a sequence containing INSTI surveillance DRMs, rarely selected nonpolymorphic INSTI-associated DRMs, or polymorphic INSTI-selected DRMs. To account for study heterogeneity, study was included in the model as a random effect using the R package lme4.

Results

Search results

The PubMed search yielded 2,141 publications of which 1,775 were excluded based on their title or abstract. Of the remaining 366 publications, 76 met inclusion criteria (Fig. 1). Of the 290 excluded publications, 126 described sequences solely from INSTI-experienced persons or from persons with insufficient treatment history; 96 did not report sequence data; 31 reported sequences from fewer than 50 INSTI-naive persons; 27 did not have sequences in GenBank and did not describe the mutations used to define INSTI-associated TDR; 7 used a low NGS mutation detection threshold; and 3 were duplicate studies.

The GenBank search yielded 75 publications meeting inclusion criterion including 48 that were also identified in the PubMed search and 27 not identified in the PubMed search (Fig. 1). Therefore, a total of 103 studies met study inclusion criteria, including 48 studies identified in both PubMed and GenBank searches, 28 identified only in the PubMed search, and 27 identified only in the GenBank search. Although 24 of the 27 studies identified only in the GenBank search were subsequently identified in PubMed, they were not retrieved by the original PubMed search as they included complete *pol* or genomic HIV-1 sequences performed primarily for defining regional subtype distributions and immune escape variants. The list of 103 included studies is provided in Supplementary Table S1.

Study populations

Table 1 summarizes the 103 studies and their 31,310 sequences by study size, geographic region, median sample

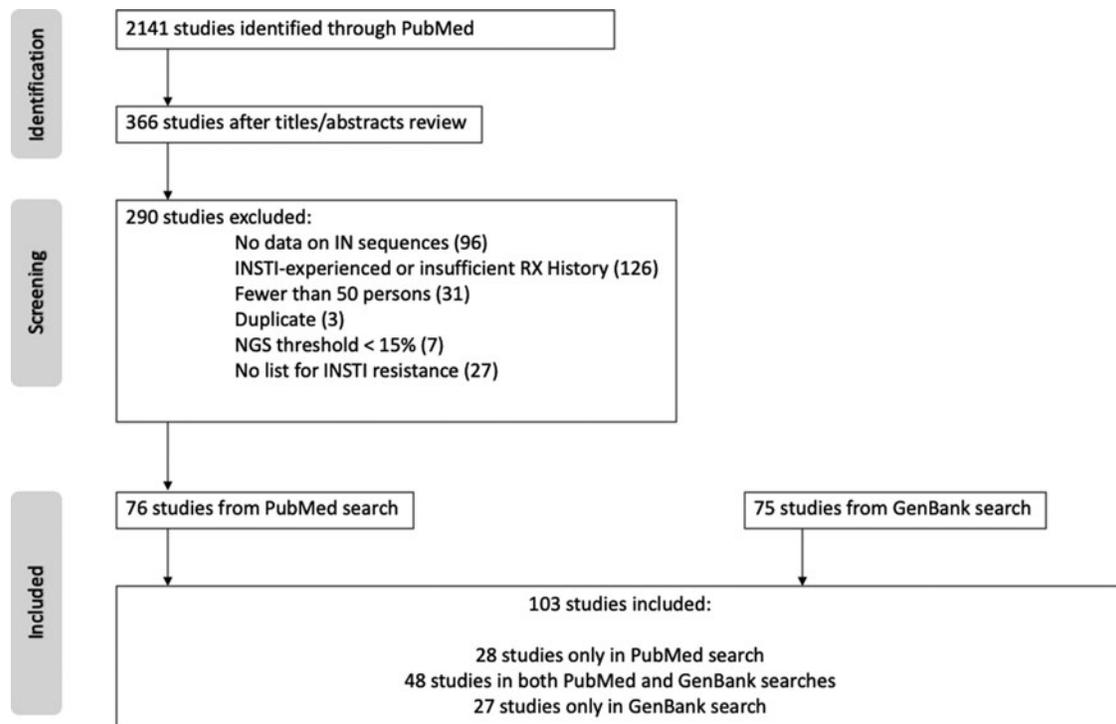


FIG. 1. Flow chart of the study selection process. Of 2,141 studies identified through the PubMed search performed in August 2020, 1,775 studies were excluded based on a review of the study title and/or abstract. Of the remaining 366 studies, 290 were excluded for one of the listed exclusion criteria. Of the 75 additional studies identified through the GenBank search that met study inclusion criteria, 27 had not been identified in the PubMed search.

year, and proportion of sequences in GenBank. Sixty-four studies included persons from the predominantly low- and middle-income country regions of Sub-Saharan Africa, South/Southeast Asia, and Latin America and 51 studies included persons from the predominantly upper-income regions of Europe and North America. The median sample year was 2013 (interquartile range: 2008–2014) and the distribution of samples by year is given in Figure 2. The large number of sequences from 2013 were baseline sequences from clinical trials of a first-line elvitegravir-containing regimen.⁹ The 75 studies containing sequences in GenBank reported 16,481 sequences.

The 28 studies not containing sequences in GenBank reported 14,829 sequences. The 16,481 GenBank sequences included 4,679 (28.4%) published in the 18 months since the list of surveillance DRMs was developed. Therefore, this set of sequences was not analyzed before this study.

Of the 103 studies, INSTI-naive sequences were obtained from ART-naive persons in 65 (63.1%) studies, from ART-experienced (but INSTI-naive) persons in 19 (18.4%) studies, and both ART-naive and ART-experienced INSTI-naive persons in 19 (18.4%) studies. Seven (6.8%) studies included only persons who were recently infected.

TABLE 1. GEOGRAPHIC DISTRIBUTION OF STUDIES CONTAINING INTEGRASE SEQUENCES FROM 50 OR MORE INTEGRASE STRAND TRANSFER INHIBITOR-NAIVE HIV-1-INFECTED PERSONS

	<i>Asia</i>	<i>Africa</i>	<i>Latin America</i>	<i>Europe</i>	<i>North America</i>
No. of studies	36	20	8	38	13
Total no. of persons	8,154	3,541	530	12,968	4,538
Median no. of persons per study (IQR)	110 (58–237)	92 (77–284)	69 (41–84)	291 (80–497)	132 (54–283)
Median sample year (IQR)	2013 (2010–2015)	2010 (2007–2014)	2008 (2003–2008)	2011 (2008–2014)	2009 (2007–2013)
No. of countries	13	15	5	17	3
Number (%) studies in GenBank	32 (89)	20 (100)	8 (100)	18 (47)	11 (85)
Number (%) sequences GenBank	6,293 (77)	3,541 (100)	530 (100)	3,922 (30)	2,195 (48)

Studies containing sequences from more than one region were divided by region if the numbers of sequences per region were known. Two studies with sequences from multiple regions are not shown because the number of sequences from each region was not provided. IQR, interquartile range.

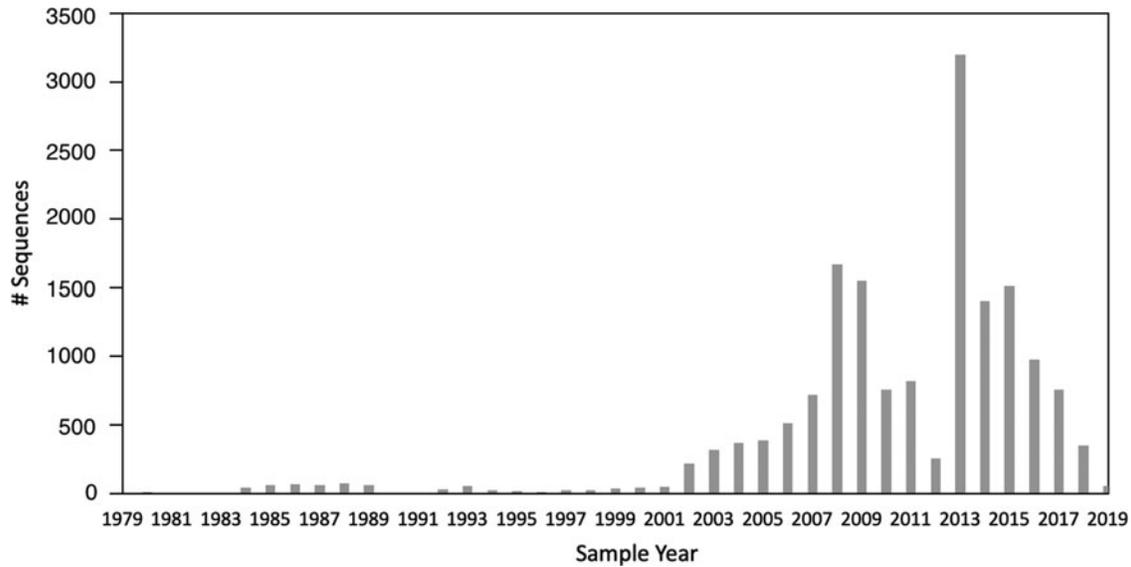


FIG. 2. Distribution of the 16,481 sequences in GenBank according to sample year.

Virus sequences

In 88 (85.4%) studies, all persons underwent integrase sequencing whereas in 15 (14.6%) studies, integrase sequencing was performed on a subset of samples from persons undergoing protease and/or RT sequencing. There was no evidence, however, that integrase sequencing in these studies was preferentially performed on samples from persons considered to be at higher risk of harboring transmitted INSTI resistance such as those with transmitted RT and/or protease resistance. Sequences were obtained from plasma in 93.2% of studies and PBMC in 6.8% of studies. NGS was performed in nine studies.

For the GenBank sequences, the most common subtypes were B (47.3%), C (20.1%), CRF01_AE (10.8%), and A (9.8%). For the 28 non-GenBank studies, subtype B was reported to be the predominant subtype in 25 studies and subtype A was the predominant subtype in 1 study; for 2 studies the predominant subtype was not reported. Of the 16,481 persons in the GenBank studies, 6,816 (41.4%) also had RT and/or protease sequences.

INSTI surveillance DRMs in studies with sequences in GenBank

Thirty-two of the 75 studies with sequences in GenBank reported one or more persons with an INSTI surveillance DRM. The overall proportion of persons with a surveillance DRM was 0.5% (77/16,481) including 75 persons with exactly 1 surveillance DRM, 1 person with 3 surveillance DRMs, and 1 person with 4 surveillance DRMs. It was similar in all regions ranging from a low of 0.4% in Latin America to a high of 0.5% in Asia. There were no meaningful differences in the proportion of persons with a surveillance DRM by subtype: A (0.5%), B (0.6%), C (0.4%), D (0.8%), F (0.8%), G (0%), CRF01_AE (0.3%), and CRF02_AG (0.3%).

The probability of a sample containing a surveillance DRM according to logistic regression was independent of sample

year (OR=0.98; 95% confidence interval [CI]: 0.92–1.05; $p=.6$). The proportion of persons with a surveillance DRM was similar (0.5%) for samples obtained before and after 2006 (Fig. 3).

Among the 6,816 persons who also underwent RT and/or protease sequencing, 679 (10.0%) had one or more RT and/or protease surveillance DRMs.⁷ INSTI surveillance DRMs were not significantly more common in the 679 samples with an RT and/or protease surveillance DRM (5/679 [0.7%]) compared with those without an RT and/or protease surveillance DRM (31/6,137 [0.5%]; $p=.4$, Fisher's exact test). The study with the highest prevalence of INSTI surveillance DRMs was conducted in Thailand between 2004 and 2009.¹⁰ It reported that among 121 ART-naïve persons, 3 (2.5%) contained a surveillance DRM: T66I, E138K, and S147G each in 1 person.

Of the 24 INSTI DRMs, 15 were present in one or more persons: E138A/K/T (29 persons in 14 studies), R263K (16 persons in 11 studies), T66I/A (11 persons in 11 studies), S230R (6 persons in 5 studies), Q148H/R (6 persons in 6 studies), S147G (6 persons in 6 studies), G140A (3 persons in 3 studies), E92G (2 persons in 2 studies), F121Y (1 person in 1 study), N155H (1 person in 1 study), and Y143S (1 person in 1 study) (Table 2). Although the two most commonly occurring surveillance DRMs, E138K (GAR=>AAR; $n=24$; 0.15%), and R263K (AGR=>AAR; $n=16$; 0.10%) occur in an APOBEC3F or 3G dinucleotide context, the sequences with these mutations had no evidence of APOBEC-mediated G-to-A hypermutation.

Additional INSTI-associated DRMs in studies with sequences in GenBank

In addition to the surveillance DRMs, we assessed the prevalence of two other categories of INSTI-associated mutations: (1) nonpolymorphic INSTI-associated mutations that are rare in INSTI-treated persons; and (2) common polymorphic INSTI-selected mutations. Of the 16,481 GenBank

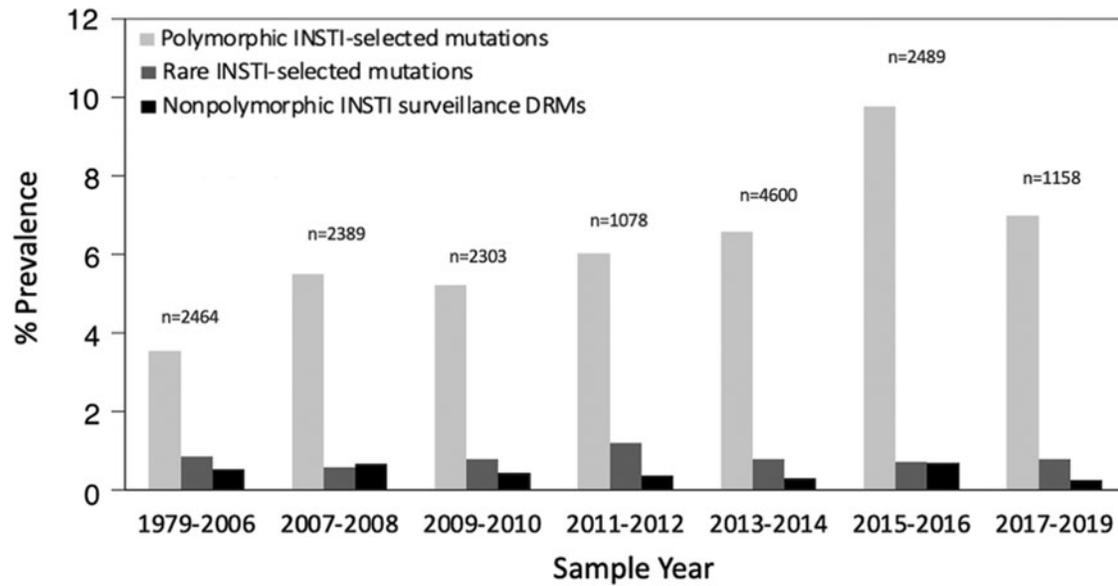


FIG. 3. The prevalence of nonpolymorphic INSTI surveillance DRMs, nonpolymorphic mutations rarely selected during INSTI therapy, and common polymorphic INSTI-selected mutations by sample year. The total number of sequences during each time period is shown above each set of histograms. DRM, drug-resistance mutation; INSTI, integrase strand transfer inhibitor.

sequences, 129 (0.8%) had one or more nonpolymorphic rarely selected INSTI-associated mutations and 1,029 (6.2%) had one or more polymorphic mutations.

There was no significant temporal trend in the likelihood that a sequence contained a rare nonpolymorphic mutation (OR = 0.99; 95% CI: 0.95–1.04; $p = .8$; Fig. 3). There were also no meaningful differences in the proportion of persons with rare INSTI-associated mutations by subtype: A (1.0%), B (0.7%), C (0.8%), D (0.8%), F (0.8%), G (0.9%), CRF01_AE (0.7%), and CRF02_AG (1.0%).

TABLE 2. PREVALENCE OF INTEGRASE STRAND TRANSFER INHIBITOR SURVEILLANCE DRUG RESISTANCE MUTATION IN 75 STUDIES WITH SEQUENCES IN GENBANK AND 28 STUDIES WITHOUT SEQUENCES IN GENBANK

Surveillance DRM	GenBank Studies (N = 16,481), n (%)	Non-GenBank Studies (N = 14,829), n (%)
E138A/K/T	29 (0.18)	14 (0.09)
R263K	16 (0.10)	2 (0.01)
T66I/A	11 (0.07)	6 (0.04)
S230R	6 (0.04)	3 (0.02)
Q148H/K/R	6 (0.04)	17 (0.11)
S147G	6 (0.04)	1 (0.01)
G140A/S/C	3 (0.02)	8 (0.05)
E92G/Q	2 (0.01)	10 (0.07)
F121Y	1 (0.01)	11 (0.07)
N155H	1 (0.01)	1 (0.01)
Y143R/S	1 (0.01)	2 (0.01)

Several of the INSTI surveillance DRMs were inconsistently recorded as DRMs by study authors. This was most often the case for R263K, which was not recognized as an important DRM until 2012.

DRM, drug-resistance mutation; INSTI, integrase strand transfer inhibitor.

The likelihood of a sequence containing a polymorphic INSTI-selected mutation exhibited a significant yearly increase (OR: 1.05; 95% CI: 1.01–1.09; $p = .007$) (Figs. 3 and 4). The most common polymorphic mutations were E157Q (2.5%), L74M (1.5%), and T97A (1.4%). There was some variability in the proportions of persons with one or more of the common polymorphic INSTI-selected mutations by subtype: A (7.3%), B (5.7%), C (6.3%), D (9.7%), F (12.0%), G (9.8%), CRF01_AE (2.5%), and CRF02_AG (13.1%). The complete summary of the prevalences of INSTI surveillance DRMs, rare INSTI-associated DRMs, and common polymorphic INSTI-selected mutations by subtype is given in Supplementary Table S2.

INSTI-associated mutations in non-GenBank studies

Sixteen of the 28 non-GenBank studies reported one or more persons with an INSTI surveillance DRM including 65 persons with 1 DRM and 5 with 2 DRMs. The prevalence of INSTI surveillance DRMs in these studies was 0.5% and therefore the same as in the studies with sequences in GenBank. Although it was not possible to perform individual level analyses, there did not appear to be significant differences in the proportions of persons with a surveillance DRM according to study region, median sample year, or non-INSTI ART exposure.

Twenty distinct surveillance DRMs were reported of which the most common were Q148H/K/R (17 persons in 5 studies), E138A/K (14 persons in 9 studies), F121Y (11 persons in 3 studies), E92G/Q (10 persons in 4 studies), G140A/S/C (8 persons in 6 studies), T66I/A (6 persons in 5 studies), S230R (3 persons in 2 study), R263K (2 persons in 2 studies), Y143R/S (2 person in 2 study), S147G (1 person in 1 study), and N155H (1 person in 1 study) (Table 2). The study with the highest TDR prevalence was conducted in Italy between 2009 and 2018.¹¹ It identified 3 (5.8%) surveillance

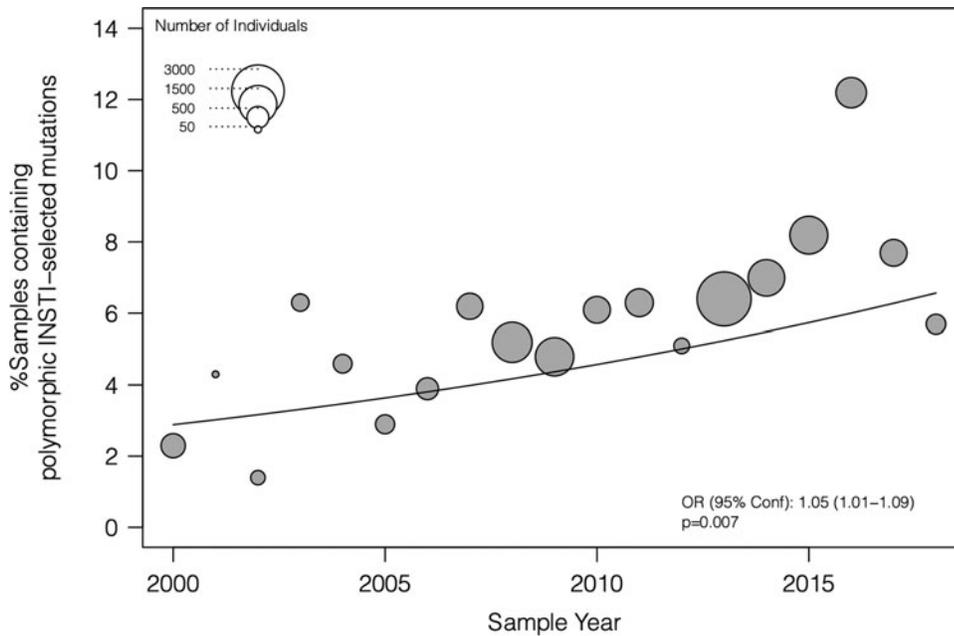


FIG. 4. Temporal trend in the yearly proportion of sequences with one or more polymorphic INSTI-selected mutations. The diameter of each circle is proportional to the number of samples sequenced that year. The polymorphic INSTI-selected mutations include A49P, L74M, T97A, E157Q, G163R/K, and D232N. The fitted line shows the fixed effect of sample year in generalized linear mixed model logistic regression. The yearly change in the odds (odds ratio [OR]), 95% CI and p -value of a sequence containing polymorphic INSTI selected mutations are indicated. CI, confidence interval.

DRMs in 52 ART-naive persons (G140S+Q148H in 2 persons and E138K in 1 person). Another study conducted in Korea between 2014 and 2017 identified 3 (5.2%) surveillance DRMs in 58 INSTI-naive ART-experienced persons (E92Q in 2 persons and S230R in 1 person).¹²

Discussion

This systematic review supports the prevailing concept that INSTI-associated TDR is infrequent.⁴ The 103 studies reviewed included ~31,000 INSTI-naive persons from 53 countries. The overall median surveillance DRM prevalence for the 75 studies with sequences in GenBank was 0.5%. There was no association of INSTI resistance with region, year, or ART experience with drugs other than INSTIs. There was also no difference in the proportion of persons with a surveillance DRM before and after the approval of the first INSTI in late 2006.

We hypothesize that a significant proportion of INSTI surveillance DRMs may reflect the cumulative rare natural occurrence of these mutations rather than transmission of resistant variants from INSTI-treated persons. E138K and R263K, the two most commonly occurring surveillance DRMs occurred in ~0.25% of samples accounting for approximately one-half of all surveillance DRMs and was unchanged before and after the introduction of INSTIs. Moreover, several additional surveillance DRMs have nonzero INSTI-naive prevalences ranging from 0.01% to 0.05%, which when combined with E138K and R263K result in an aggregate prevalence of 0.5%.⁶ Thus, 0.5% approximates the background surveillance DRM prevalence in the absence of INSTI selective drug pressure.

Of the 103 studies identified in this review, nearly all reported that transmitted INSTI resistance has been low even in regions where INSTI use has been widespread. For example, in Swiss HIV cohort, only 6 (0.5%) of 1,316 integrase samples obtained from INSTI-naive persons between 2008 and 2014 had a nonpolymorphic INSTI-associated DRM.¹³ In the Spanish CoRIS cohort, only 1 (0.1%) of 1,109 integrase

samples obtained from ART-naive persons between 2012 and 2017 had a nonpolymorphic INSTI-associated DRM.¹⁴ In a study from North Carolina between 2010 and 2016, only 3 (0.4%) of 840 samples obtained from ART-naive persons had a sample with an INSTI surveillance DRM.¹⁵ In addition, a study published in 2020 following the completion of this systematic review, reported that none of 640 ART-naive persons in the United Kingdom tested between 2014 and 2016 had an INSTI-associated DRM above the 20% threshold used for Sanger sequencing.¹⁶

In contrast, several studies have demonstrated that transmitted INSTI drug resistance can be identified at the population level. Chang *et al.* reported that 12 (0.9%) of 1,307 ART and/or INSTI-naive persons from Taiwan had nonpolymorphic INSTI-associated DRMs between 2006 and 2015.¹⁷ Their study was notable for phylogenetic analyses that identified several sequence clusters that included viruses from INSTI-experienced persons and ART-naive persons sharing the same INSTI-associated DRMs.

In addition, three recent studies from the United States presented at the Conference on Retroviruses and Opportunistic Infections in 2019 and 2020¹⁸⁻²⁰ also reported prevalences of nonpolymorphic INSTI-associated DRMs of ~1%. Specifically, two nationwide studies published by the U.S. Centers for Disease Control and Prevention reported rates of INSTI-associated DRMs of 0.8% in newly diagnosed persons both between 2013 and 2016 and in 2018,^{18,19} and a statewide study from Florida reported rates of 1.4% in 2015 and 2.2% in 2016 in ~4,000 newly diagnosed persons.²⁰ This relatively high rate of INSTI-associated DRMs in this population is consistent with the increasing proportion of acquired INSTI-associated resistance in Florida.²¹ Based on our hypothesis that the background prevalence of INSTI-associated surveillance DRMs in INSTI-naive persons is ~0.5%, these reports are consistent with a recent increase in INSTI-associated TDR in the United States.

This study has two major limitations. This first limitation is that the analyzed dataset overlaps with the dataset used to

develop the list of surveillance DRMs. Because the surveillance DRMs were selected based on being nonpolymorphic, one could argue that this is responsible for the low prevalence of surveillance DRMs in this study. Four factors, however, argue that there are independent data supporting the study's conclusions. First, the prevalence of the surveillance DRMs was also 0.5% in the 28 studies reporting 14,829 sequences that were not in GenBank and therefore not used to derive the surveillance DRMs. Second, ~30% of the sequences in this study were published between February 2019 and August 2020 and these were also not used to derive the surveillance DRMs. Third, essentially identical results are obtained using the complete list of 2019 IAS-USA list of 25 INSTI resistance mutations with the exception of the known polymorphic mutations L74M and T97A²² (data not shown). These mutations include 22 of the 24 INSTI surveillance DRMs plus three additional rare mutations listed for the investigational INSTI cabotegravir, which were not present in this dataset. Finally, the lack of a temporal increase in the prevalence of INSTI surveillance DRMs is consistent with our hypothesis that there is a background prevalence of these mutations of ~0.5% in the absence of selective drug pressure.

The second major limitation of the study is that the median sample year was 2013, which resulted in part from the median 4-year delay between sample collection and publication date. Therefore, ongoing surveillance is required to determine the more recent regional prevalences of transmitted INSTI resistance.

A secondary goal of this study was to examine the prevalence in INSTI-naive persons of two categories of mutations that were not included in the list of INSTI surveillance DRMs including the nonpolymorphic mutations that are rarely observed in INSTI-treated persons and the common polymorphic accessory INSTI-selected mutations. Of interest, we found that polymorphic accessory INSTI-selected mutations particularly L74M, T97A, and E157Q increased over time. One intriguing possible explanation for this finding is that in some persons, these mutations emerged in combination with nonpolymorphic INSTI DRMs but that the nonpolymorphic DRMs are more likely to fade from detectability after virus transmission because of their reduced fitness in the absence of selective drug pressure. Indeed, several of the most common clinically significant mutations including Q148H/R and N155H have markedly reduced fitness *in vitro* with replication levels even below those of M184V.^{23–25}

Conclusions

Our study supports the prevailing concept that INSTI-associated TDR has been very uncommon. Our analysis also indicates that even in the absence of selective drug pressure there is an ~0.5% background prevalence of INSTI surveillance DRMs. Recognition of this background makes it possible to conclude that ART-naive populations with prevalences above this background are likely undergoing INSTI-associated TDR caused by viruses emerging under INSTI selective drug pressure.

Authors' Contributions

R.W.S. conceived, designed the study and the primary writer of the article. A.J.B. and S.-Y.R. screened titles, abstracts, and full text articles for inclusion and extracted data

from included studies. A.J.B. and S.-Y.R. analyzed data and performed statistical analysis. All authors contributed to writing the article and approved the final version.

Disclaimer

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Author Disclosure Statement

No competing financial interests exist.

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Supplementary Material

Supplementary Table S1
Supplementary Table S2

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