Systematic Review of HIV Drug Resistance in Southeast Asia

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Abstract

In 2010, 3.5 million people were living with HIV in the World Health Organization Southeast Asia Region (SEAR), giving this region the greatest burden of HIV after Africa. Scale-up of antiretroviral therapy has resulted in over 717,000 benefitting from it by the end of 2010. A systematic review of studies of HIV drug resistance in the SEAR published between 2000 and 2011 was performed. Of 10 studies of transmitted HIV drug resistance in recently infected patients, all but two reported low levels (< 5%) of transmitted resistance. Of 23 studies of HIV drug resistance in pretreatment populations initiating antiretroviral therapy, three reported moderate levels (5-15%) of HIV drug resistance and 20 reported low levels. Amongst 17 studies of acquired HIV drug resistance, levels of nucleoside reverse transcriptase inhibitor and nonnucleoside reverse transcriptase inhibitor resistance ranged from 52 to 92% and 43 to 100%, respectively, amongst those with virological failure. Overall, data included in this review suggest that currently recommended first- and second-line regimens are appropriate for the cohorts studied. However, data were only available from two of 11 Southeast Asia Region countries and studies largely examined urban populations. Results are unlikely to be representative of the region. Studies lacked standardized methods, which greatly limits comparability of data and their use for public health and antiretroviral therapy program planning. Routine, standardized, and nationally representative HIV drug resistance surveillance should be strongly encouraged in the Southeast Asia Region countries to best characterize population-level HIV drug resistance. National-level HIV drug resistance surveillance data may be used to optimize delivery of HIV care and treatment and minimize emergence of population-level HIV drug resistance, thus promoting the long-term efficacy and durability of available first- and second-line antiretroviral therapy regimens. **(AIDS Rev. 2013;15:162-70)**

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Key words

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Introduction

In 2010, an estimated 3.5 million people were living with HIV in the World Health Organization (WHO) Southeast Asia Region (SEAR)[1] (Fig. 1), giving this region the greatest burden of HIV after Africa. Five of 11 SEAR countries shoulder the region's HIV burden (India, Indonesia, Myanmar, Nepal, and Thailand), five countries carry less than 1% of the HIV burden (Bangladesh, Bhutan, Maldives, Sri Lanka, and Timor-Leste), and data are unavailable from the Democratic People's Republic of Korea (DPR Korea). In SEAR countries, the number of children living with HIV increased from 89,000 in 2001 to 140,000 in 2010, suggesting that mother-to-child transmission remains a significant mode of transmission[1]. Although the prevalence of HIV

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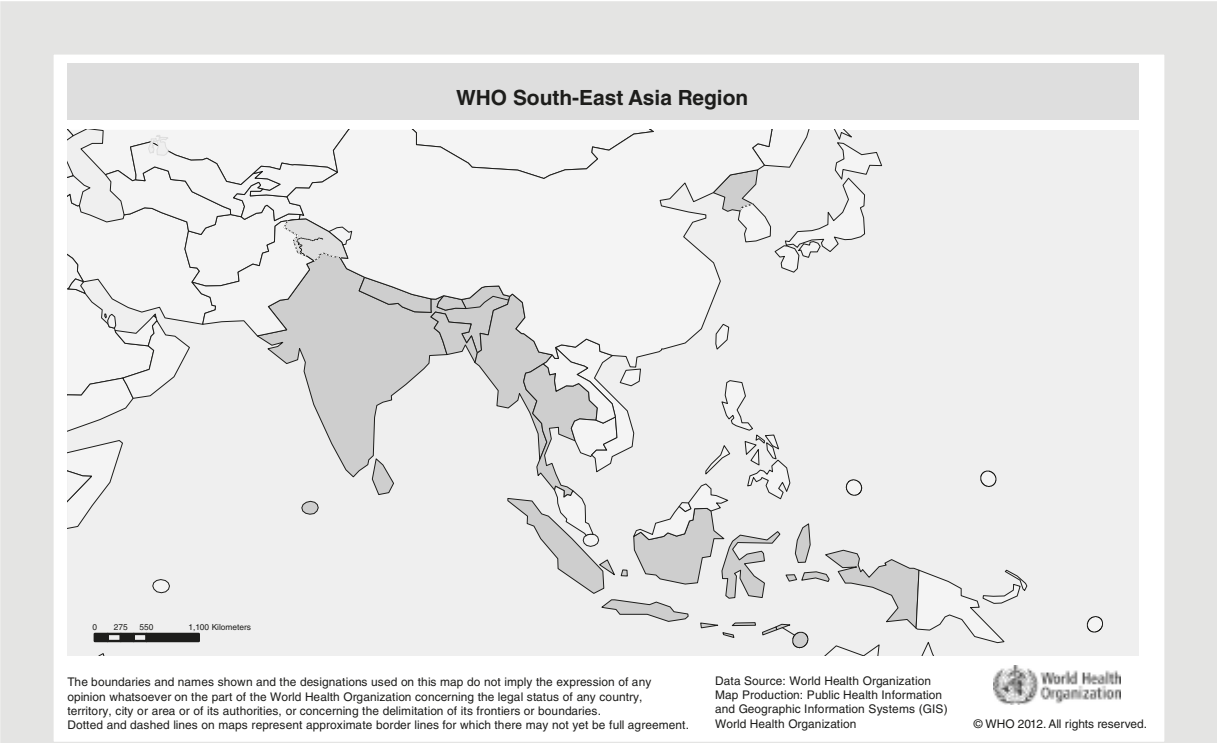


Figure 1. *Southeast Asia Region (SEAR) of the World Health Organization (WHO). Countries represented in the SEAR are shown in grey. Dotted and dashed lines represent approximate borders which may not yet be in full agreement.*

in the general adult population is low (estimated at 0.3% in 2010), most-at-risk populations including sex workers and their clients, injecting drug users (IDU), men who have sex with men (MSM), and transgender populations carry a disproportionate HIV burden[1]. Across 269 sentinel sites in nine SEAR countries (excluding Bhutan and DPR Korea) for 2007-2010, HIV prevalence in female sex workers was < 1% in 33% of sites, 1-5% in 38% of sites, and 5-20% in 25% of sites, with the highest prevalence observed in south India. National HIV prevalence estimates in MSM in SEAR range from 5.2 to 28.8%[1]. However, these national estimates mask higher local estimates, i.e. 31 and 41% in MSM populations in Bangkok, Thailand and Hyderabad, India, respectively. Data regarding HIV prevalence in transgender populations is limited, but where data are available in MSM and corresponding transgender populations in the same geographic area, estimates in transgender populations are generally higher than in MSM populations. Five SEAR countries have significant HIV epidemics in IDU populations, including Myanmar, Indonesia, and Thailand where HIV prevalence was 26.5% (2010), 27% (2007), and 46% (2010), respectively[1].

Southeast Asia region antiretroviral treatment scale-up and guidelines

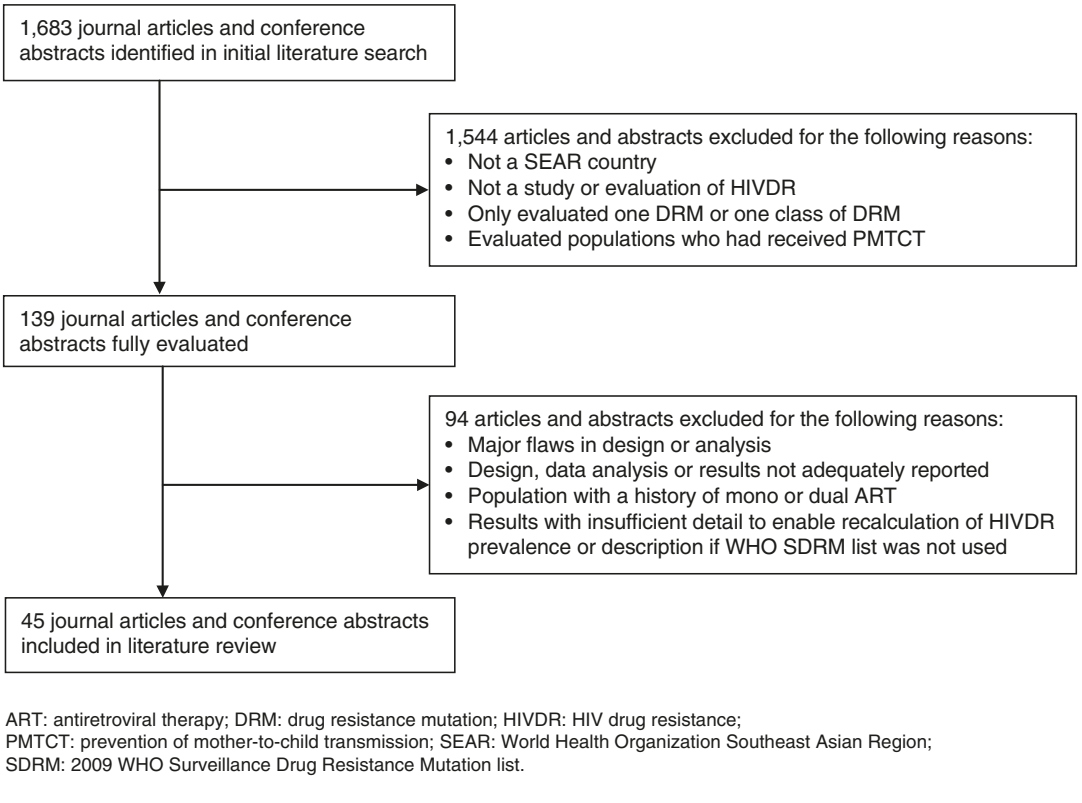
Globally, over eight million people were receiving antiretroviral therapy (ART) in low and middle-income countries as of the end of 2010, representing a 26-fold increase since 2003[2]. As in other regions of the world, SEAR has experience rapid expansion of ART, with over 717,000 individuals benefitting from it at the end of 2010[1]. Successful ART scale-up in SEAR has been largely due to the use of a public health approach to ART delivery supported by standardized protocols and simplified patient monitoring[3]. The ART guidelines from all 10 SEAR countries recommend use of nonnucleoside reverse transcriptase inhibitor (NNRTI)-based first-line regimens in combination with two nucleoside reverse transcriptase inhibitors (NRTI)[4-13]. In all ten countries, tenofovir is a component of first-line regimens[4-13]. In all SEAR countries, ritonavir-boosted protease inhibitors (PI) in combination with two NRTI are reserved as second-line agents for patients with virological failure or toxicity to NNRTI[4-13]. Treatment guidelines from the DPR Korea were unavailable.

HIV drug resistance

In the presence of drug selective pressure, emergence of some HIV drug resistance (HIVDR) is inevitable due to the error-prone replication of HIV, its high mutation rate, and the need for lifelong treatment[14,15]. Given the inevitability of some HIVDR, it is not surprising that a 2012 analysis showed that higher levels of HIVDR were observed in areas with greater ART coverage[16] (defined as the number of people on ART divided by the number of people with HIV). Moreover, increased levels were observed with increasing time since treatment roll-out, a finding particularly notable in southern and east Africa, where HIVDR was estimated to have increased at a rate of almost 15 and 30% per year, respectively, since ART roll-out. This increase in HIVDR was driven almost exclusively by NNRTI. Although similar increases were not observed in other regions, this increase may reflect lack of data rather than differences in levels of HIVDR[17]. As in most resource-limited settings, access to patient-level HIVDR testing, viral load monitoring, and second-line and salvage regimens is often restricted in SEAR. Therefore, reliable information about HIVDR which can inform public health and ART program decision making is required in the region to support the choice of treatment regimens and optimization of patient care[18].

Broadly, population-level HIVDR may be divided into three main categories: transmitted HIVDR, acquired HIVDR, and HIVDR in pretreatment populations.

– Transmitted HIVDR is HIVDR detected in recently infected populations who are unexposed to antiretroviral (ARV) drugs. In the absence of drug selective pressure, certain drug resistance mutations will revert to "wild-type" at varying rates after initial infection[19]. When patients with transmitted HIVDR initiate ART, drug selective pressure may result in rapid re-emergence of clinically relevant mutations, leading to rapid virological failure.



ART: antiretroviral therapy; DRM: drug resistance mutation; HIVDR: HIV drug resistance; PMTCT: prevention of mother-to-child transmission; SEAR: World Health Organization Southeast Asian Region; SDRM: 2009 WHO Surveillance Drug Resistance Mutation list.

Figure 2. *Flowchart of studies included in the review of HIV drug resistance in World Health Organization Southeast Asian Region countries.*

- Acquired HIVDR is HIVDR which emerges in response to drug selective pressure. Acquired HIVDR may emerge even when optimal regimens are provided and adherence is supported.
- Pretreatment HIVDR is HIVDR detected in populations initiating ART for the first time. Pretreatment HIVDR may have been acquired due to ARV drug exposures including prevention of mother-to-child transmission (PMTCT), pre-exposure prophylaxis (PrEP) or post-exposure prophylaxis (PEP), previous combination ART, or may have been present since time of infection (transmitted)[18,19].

The purpose of this review is to summarize the published literature assessing transmitted, acquired, and pretreatment HIVDR in the SEAR countries and identify gaps in knowledge required for public health decision making, and to suggest directions for future research. A systematic review of the literature was performed to identify reports published in English between January 2000 and August 2011 that documented transmitted, pretreatment, and acquired HIVDR in SEAR countries (Fig. 2; see Supplementary data).

Definition of categories of drug resistance

Between 2005 and 2012, the WHO recommended a method to categorize transmitted HIVDR into three prevalence classifications: low (< 5%), moderate (5-15%), or high (> 15%)[15]. For the purpose of this review, these prevalence classifications were applied to results from pretreatment populations to facilitate comparison between studies and discussion. Accurate estimation of levels of transmitted HIVDR requires genotyping of specimens from populations likely to be recently infected and ARV naive. Commonly used WHO criteria to maximize inclusion of individuals likely to be recently infected include age < 25 years, no previous pregnancy (if female), CD4 cell count > 500 cells/mm^3, and first HIV risk-defining event within the past three years, if available. Recent data suggest that under certain circumstances, and if resources permit, LAg-Avidity EIA or Multi-Assay Algorithm assay (MAA) may be used to identify individuals likely to be recently infected[21,22]. Studies evaluating the rate of CD4 cell count decay from time of seroconversion facilitate estimation of the duration of infection[23,24]. Lodi, et al. document that the median time from seroconversion to a CD4 count < 350 cells/mm^3 is 4.19 years. In this review, to standardize results and facilitate discussion, cohorts with median CD4 cell counts > 350 cells/mm^3 were classified as describing transmitted HIVDR, and cohorts with median CD4 cell counts < 350 cells/mm^3 were classified as describing pretreatment populations.

Transmitted HIV drug resistance

A total of 10 studies of transmitted HIVDR were included (Table 1, supplementary data). Nine of the 10 studies reported low (< 5%) levels of transmitted HIVDR when WHO classifications were applied. Two studies documented moderate levels (5-15%) of transmitted NNRTI resistance. One study described a cohort of 18 voluntary counseling and testing (VCT) attendees in Chennai, India and reported 11% NNRTI HIVDR[30]. The other described a cohort of 303 sexually transmitted infection (STI) clinic attendees in Mumbai, India, of which 62 of 303 had a genotype performed and reported 5.7% HIVDR[31]. Overall, the findings suggest that resistance transmission remains minimal in the cohorts assessed and that current recommendations for first-line ART are likely to be appropriate for the majority of patients included in these studies when they require therapy in the future.

The accurate estimation of transmitted HIVDR requires assessment of recently infected individuals. As the time period between seroconversion and HIVDR testing increases, the likelihood that transmitted HIVDR is underestimated due to reversion to wild-type virus increases[19]. In addition, undisclosed ARV exposure between seroconversion and the time of HIVDR testing may result in acquired HIVDR, which can lead to falsely elevated estimates of transmitted HIVDR.

In the studies included in this review, various criteria were used to define recently infected populations, including: women with no previous pregnancy[32], BED capture enzyme immunoassay analysis[30,31,34], or serial HIV testing to document seroconversion[34-37].

Four studies were reclassified as studies reporting HIVDR in pretreatment populations[38-41]. Two[38,39] were reclassified because the median CD4 cell counts of the study cohort was < 350 cells/mm^3, suggesting the majority were chronically rather than recently infected with HIV[23,24]. One was reclassified because the frequency of serial testing was inadequately described and the median CD4 cell count of the cohort was not reported[40]. The remaining study evaluated a VCT population of which 71% were female, but no information was provided concerning previous or current pregnancy or median CD4 cell count of the study population[41]. For these reasons, this publication was reclassified as a study in a pretreatment population.

The overall few studies successfully achieving their stated aim of assessing transmitted HIVDR underscores challenges associated with identifying recently infected populations. To facilitate surveillance of transmitted HIVDR, the WHO has recently developed new methods and updated epidemiological criteria used to define recently infected populations in resource-limited settings[15,42,43].

Acquired HIV drug resistance

Nine cross-sectional studies of acquired HIVDR and eight studies of HIVDR in populations identified as failing ART by clinical, immunological, and/or virological criteria were included. Levels of NRTI and NNRTI resistance amongst those failing with detectable viral load across all studies of acquired drug resistance ranged from 52 to 92% and 43 to 100%, respectively (Table 2, supplementary data).

In cross-sectional studies of acquired HIVDR, the duration on ART at time of viral load and genotyping ranged from six to 50 months. Overall, eight of nine cross-sectional studies reported low levels of PI resistance in patients failing PI-based first-line ART. However, in one study, two of four (50%) patients on PI-based ART were failing with major PI mutations[27]. Specific ART regimens were not described and use of un-boosted PIs cannot be excluded. Moreover, this study's small sample size renders uncertain any interpretation. In all nine cross-sectional studies, NNRTI and NRTI resistance predominated amongst patients with virological failure and detected HIVDR. The levels of HIVDR and complex NRTI patterns observed in some reports suggest a long duration of virological failure in the setting of ongoing drug selective pressure prior to detection of treatment failure.

Eight studies of acquired HIVDR evaluating populations failing first-line ART by clinical, immunological, or virological criteria were included (Table 1, supplementary data). Amongst these eight studies, the prevalence of Q151M, which confers broad NRTI cross resistance, ranged from 0-15%. One study reported 11% K65R, which was considerably higher when compared to other reports in this review (range 0-6%)[26]. In 50% of studies of acquired HIVDR performed in populations detected as failing ART by clinical, immunological, or virological criteria, higher levels of thymidine analogue mutations (TAM)[44], Q151M[45], K65R in combination with Q151M[26], or any mutations conferring resistance to tenofovir (TDF)[46] were reported. Q151M confers broad high-level resistance to most NRTI and low-level resistance to TDF; K65R confers high-level resistance to TDF[47], an important component of recommended ART regimens[48]. Studies reporting higher levels of Q151M suggest prolonged duration of virological failure on NRTI-containing regimens[26,45].

Due to marked heterogeneity of data, it was not feasible to assess the proportion of patients with NRTI-only resistance or the proportion with one or more TAM (Table 2, supplementary data).

Although results indicate that the majority of those assessed who were failing a NNRTI-based first-line regimen in these studies would achieve virological suppression on PI-based second-line regimens, anticipated levels of resistance to second-line NRTI components of commonly used regimens supports scale-up of routine viral load testing. Specifically, routine viral load testing would permit early detection of virological failure, thus allowing for reinforcement of patient adherence to ART and timely switch to PI-based ART, if required.

In most cases, patient-level data were unavailable, thus precluding assessment of the anticipated clinical relevance of TAM to second-line ART regimens used in the region. One study of acquired HIVDR in patients with clinical, immunological, or virological failure reported a 20% prevalence of PI resistance[49] and one cross-sectional study of acquired HIVDR reported an 8% prevalence of PI resistance[50]. The remaining studies of acquired HIVDR in patients with clinical, immunological, or virological failure[26,45,51] and cross-sectional studies of acquired HIVDR[28,52] found little or no PI resistance. Possible explanations for this difference include variable previous PI exposures, concurrent NRTI resistance, and use of un-boosted PI or differences in levels of adherence.

In India, the private sector provides healthcare for up to 70% of the population[50] and differences in ART delivery and HIV care in the private sector may be related to the country's higher reported levels of acquired HIVDR. For example, Shet, et al. found that patients receiving ART in the private sector in south India had a lower level of self-reported adherence, a lower level of virological suppression (defined as viral load < 100 copies/ml), and a higher prevalence of HIVDR when compared to patients in the public/private and public sectors[53]. Another study from India reported that patients receiving ART in the private sector were 2.7 times more likely to experience a treatment interruption when compared to those in the public sector[54]. Poor adherence and treatment interruptions are well documented to increase the likelihood of treatment failure and selection of HIVDR[55,56]. Other barriers to

adherence described in India's private sector include the cost of ART, drug side effects or toxicities, lack of prescriber knowledge about ART, inability to mitigate ART side effects/toxicities, and drug stock outs[54,57].

Pretreatment HIV drug resistance

Twenty-three studies of HIVDR in pretreatment populations were included (Table 1, supplementary data). Twenty studies reported low levels of HIVDR and three studies reported moderate levels of NRTI HIVDR. Among the studies of HIVDR in pretreatment populations, the prevalence of NRTI, NNRTI, and PI HIVDR was 0-8, 0-8, and 0-4.3%, respectively (Table 2, supplementary data).

The finding that the majority of studies report low levels of HIVDR suggests that amongst the cohorts studied, levels of HIVDR would not preclude successful virological suppression when currently recommended first-line regimens are used in the populations studied.

Patterns of HIV drug resistance detected

Across all studies of transmitted HIVDR, a total of 17 drug resistance mutations (DRM) were detected. Eight DRM conferring resistance to NRTI were reported: M41L, T69D, K70R, V75M, F77L, Q151M, M184V and K219R, of which M41L occurred twice and was the only NRTI DRM reported more than once in more than one sequence in one or more studies. Five DRM conferring resistance to NNRTI were reported: K101E, K103N, V106M/V, Y181C, and G190E, of which Y181C, K103N and V106M/V occurred twice and were the only NNRTI DRM reported more than once in more than one sequence in one or more studies. One study detected K101E and M184V but did not report their frequency; therefore, they are not included in this summary[31]. No PI DRM were reported.

In cross-sectional studies of acquired HIVDR, most detected resistance was to NRTI or NNRTI. Among NRTI mutations, M184V was most common, occurring in 50-90% of genotypes with any HIVDR. Of the studies which reported on the prevalence of TAM, any TAM was described in 3-42% of specimens and included M41L, D67N, K70R, T215F/Y and L210W[27-29,50,58]. The most commonly detected NNRTI DRM included K103N, K101E, G190A, and Y181C, which were detected in 24-44, 14-22, 18-35, and 23-37% of studies, respectively[27-29,50,58]. Only three studies detected PI DRM, with the most commonly observed mutations being I54M (0-6%), V82A (3-5%), and L90M (3-5%)[17,27,29].

All studies of acquired HIVDR in patients with known clinical, immunological, or virological failures reported NRTI and NNRTI resistance. Among NRTI DRM, TAM were found in up to 65% of patients failing with HIVDR within any single study. Frequently observed non-TAM included M184V detected in 33-85% of genotypes and Q151M detected in 5-11% of genotypes with HIVDR. Among NNRTI DRM, K103N, Y181C, and Y181C were detected in 25-48, 10-41, and 10-28% of genotypes, respectively.

In studies of HIVDR in pretreatment populations, six reported no HIVDR[27,28,38,40,41,59] and three did not report which DRM had been detected[25,39,60]. Of the remaining 14 studies, the most commonly observed DRM was the M184V reported in seven of 14 studies. TAM were present in six[29,61-65] of 14 studies and accounted for up to 47% of all DRM in an individual study[61]. K103N was present in three of 14 studies[61,64,66], while Y181C was reported in three of 14 studies[61,65,67]. Five of 14 studies reported PI DRM[62,65,68-70], the most common being M46I, which was present in three of the five studies reporting PI DRM[65,68,69] and which represented up to 33% of all DRM described in any individual study[68,69].

Current HIV drug resistance status in the WHO Southeast Asia region

At the end of 2010, the WHO SEAR accounted for 10% of the global population of people living with HIV[1]. In the setting of ongoing ART scale-up in SEAR countries, emergence of HIVDR is inevitable and necessitates population-based HIVDR surveillance to guide ART programs in the selection of appropriate and effective regimens for first- and second-line ART, PMTCT, PrEP, and PEP. Moreover, when combined with data obtained from routine ART program monitoring and evaluation activities, HIVDR surveillance data supports identification of ART program and clinic-level factors requiring optimization in order to minimize the preventable emergence and transmission of HIVDR.

In this review of HIVDR studies published in SEAR, overall reported levels of HIVDR were low. Based on results from studies assessing transmitted and pretreatment HIVDR, currently recommended first-line ART regimens appear appropriate for the majority of study participants. Moreover, the frequency and pattern of DRM described in studies of acquired HIVDR supports currently recommended PI-based second-line ART regimens for the majority participating in these studies. However, relatively high levels of acquired

resistance to one or more of the NRTI used in second-line regimens support use of routine viral load testing to detect virological failure early and support interventions to improve adherence or earlier switch to second-line. Finally, HIVDR described in studies of pretreatment populations support currently recommended first-line ART regimens for those assessed in the studies included in this review.

This review has several important limitations. The interpretation of four of 10 studies of transmitted HIVDR is limited due to the studies' small sample sizes, wide confidence intervals, mixed populations (VCT and antenatal care), and multiple years of pooled data[30,31,61,71]. In addition, a wide range of genotyping amplification rates (52-94%) raises concerns regarding specimen quality, reproducibility, and the sensitivity/specificity of genotyping assays used.

Amongst studies of acquired HIVDR, interpretation of eight studies was limited due to their small sample size, limited description of epidemiological methods, HIVDR prevalence estimates with wide confidence intervals, heterogeneity of previous ARV experience, or partially missing genotypic information, specifically no information about TAM[26,28,29,44,50,52,72,73]. The study characteristics greatly limit the generalizability and interpretation of results beyond the cohorts assessed. Finally, interpretation of two of 23 studies of HIVDR in pretreatment populations was limited due to high rates of amplification failure[62,65]. In almost all studies, small conveniently chosen samples limit generalization of results, thus greatly limiting the utility of results for public health planners in the region. Finally, very few studies reported prevalence estimates with corresponding confidence intervals, greatly limiting data interpretation.

The absence of reports from nine of 11 SEAR countries combined with the fact that all data available were obtained from urban areas further highlights important gaps in our knowledge about HIVDR in the SEAR area. The magnitude and possible impact of HIVDR on ART treatment outcomes in the remaining nine SEAR countries and in non-urban areas remains unknown. The paucity of data and the limited applicability of available data for public health planning underscore the urgent need for routine national HIVDR surveillance in SEAR countries. To support population-level statements about HIVDR and provide needed information to ART program planners and Ministries of Health, the WHO recommends standardized nationally representative methods to assess acquired, transmitted, and pretreatment HIVDR within defined populations[15].

Conclusions

In this systematic review of HIVDR in the WHO SEAR region, most studies reported low levels of HIVDR, which is reassuring. However, limited generalizability of results, heterogeneity of study designs, and biases introduced by high and possibly non-random rates of genotyping failure diminish the strength of findings to support public health and ART program recommendations and actions.

Routine, standardized, and nationally representative HIVDR surveillance should be strongly encouraged in SEAR countries to best characterize population-level HIVDR. In countries with low-prevalence and concentrated epidemics, surveillance activities should be extended to poorly characterized most-at-risk populations, as well as to geographic areas where information is limited or nonexistent. Results of HIVDR surveillance activities should be actively used to optimize delivery of HIV care and treatment and promote the long-term efficacy and durability of available first- and second-line ART regimens in the region.

Disclosures

All authors report no conflict of interests. Some authors are employees of the WHO. Views expressed in this manuscript are the views of the authors and not the WHO.

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Supplementary Data

Supplementary data is available at AIDS Reviews journal online (http://www.aidsreviews.com). This data is provided by the author and published online to benefit the reader. The contents of all supplementary data are the sole responsibility of the authors.

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