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Drug Resistance Among Newly Diagnosed People Living with HIV-1 in Sichuan, China: A Large-Scale Population-Based Cross-Sectional Study

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Introduction: The increasing utilization of antiretroviral (ART) medications by people living with HIV(PLWH) poses a potential risk to the efficacy of standardized ART management protocols in resource-limited settings, particularly as it may accelerate the emergence of transmitted drug resistance (TDR). Our research aimed to elucidate the prevalence, risk factors, geographical variations, and transmission patterns of TDR to inform and enhance ART management strategies.

Methods: The study covered newly diagnosed PLWH in Sichuan from July 2022 to June 2023. We conducted sequencing - based resistance profiling and then analyzed TDR via the Stanford HIV Drug Resistance Database and logistic regression to identify associated factors via SPSS. ArcGIS and spatial statistics were used to create LISA maps showing the TDR distribution. HIV-TRACE's Tamura Nei 93 model was used to assess genetic clustering, and QGIS was used to analyze TDR intensity within and between cities.

Results: This study found that the prevalence of TDR in Sichuan had reached a moderate level, with a resistance rate of 10.91%. Resistance varied by drug class: 8.08% for NNRTIs, 1.34% for NRTIs, and 2.22% for PIs. Regions like LS, DY, SN, and LZ had higher TDR rates. Within NNRTIs, efavirenz (EFV) and nevirapine (NVP) resistance rates were 6.26% and 6.50%, respectively. Furthermore, risk factors for TDR included age under 15, HIV-1 subtypes, and residing in areas with high ART failure rates. In addition, spatial analysis showed significant clustering of TDR, especially for PIs and NNRTIs, with hotspots identified in certain regions. Among 1,486 drug-resistant PLWH, 52.76% formed resistance-linked networks, comprising 181 molecular clusters. Key resistance mutations included K103N, E138A, V179D, Y181C, and Q58E. The transmission intensity of TDR varied across regions, highlighting complex spatial dynamics.

Discussion: The prevalence of TDR in Sichuan had reached moderate epidemic levels, with certain cities already experiencing high epidemic rates. It is urgent that we urgently expand targeted genotypic drug resistance detection and implement interventions to improve medication adherence.

Keywords: HIV-1, transmitted drug resistance, antiretroviral therapy, nonnucleoside reverse transcriptase inhibitors, spatial analysis, molecular network

Introduction

Acquired immune deficiency syndrome (AIDS), as a type of infectious disease, has an important and far-reaching influence on Chinese society. To curb deaths and new infections, China actively responds to the "95–95-95" AIDS prevention and control goal. Since 2016, China's "universal treatment" program has enabled all people living with HIV (PLWH) to receive timely treatment upon request. Early treatment not only extends lifespan of PLWH but also prevents secondary HIV transmission. Detecting and treating it promptly is the most effective AIDS prevention and treatment approach. However, with all PLWH on antiretroviral therapy (ART), the spread of HIV-resistant strains poses a major challenge in AIDS prevention and treatment.[1–4] From the 2003 World Health Organization (WHO) guidelines advocating

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the use of nucleoside reverse transcriptase inhibitors (NRTIs) and non-nucleoside reverse transcriptase inhibitors (NNRTIs) to the present day, when more efficacious and better-tolerated integrase strand transfer inhibitors (INSTIs) are in use, it is clear that the evolution of ART drug utilization within the population has a direct bearing on the prevalence of drug resistance.[5] Transmitted drug resistance (TDR), detected in newly diagnosed PLWH without prior ART,[6] can affect ART treatment, increase mortality, and cause secondary HIV transmission.[7] Thus, assessing TDR levels is crucial for informing ART policies and ART services.

Sichuan, situated in Southwest China (Figure 1A), comprises 21 prefecture-level cities (Figure 1B) and 183 counties (Figure 1C). Geographically, it is divided into the Chengdu Plain, Northeast Sichuan, South Sichuan, Panxi and West Sichuan (Figure 1B). With a permanent population of 83.675 million in the seventh census, it serves as Southwest China's economic hub. In terms of the AIDS epidemic, Sichuan had 220,000 cumulative and 17,000 newly diagnosed PLWH in 2023, indicating a severe epidemic situation that necessitates continuous TDR monitoring. In 2022, the Chinese Center for Disease Control and Prevention(CDC) conducted a drug-resistance survey on newly diagnosed PLWH in six provinces including Sichuan, revealing a 7.4%[8] resistance rate, higher than sub-Saharan Africa (6.0%) but lower than Greece (7.8%), Hungary (10.7%), Cameroon (10.4%), Namibia (12.7%), Israel (12.1%), Mozambique (14.4%), Mexico (14.8%), the US (22.5%), and Japan (12.5%).[9–18]Integrating 2014 and 2019 surveillance results in Sichuan, a significant

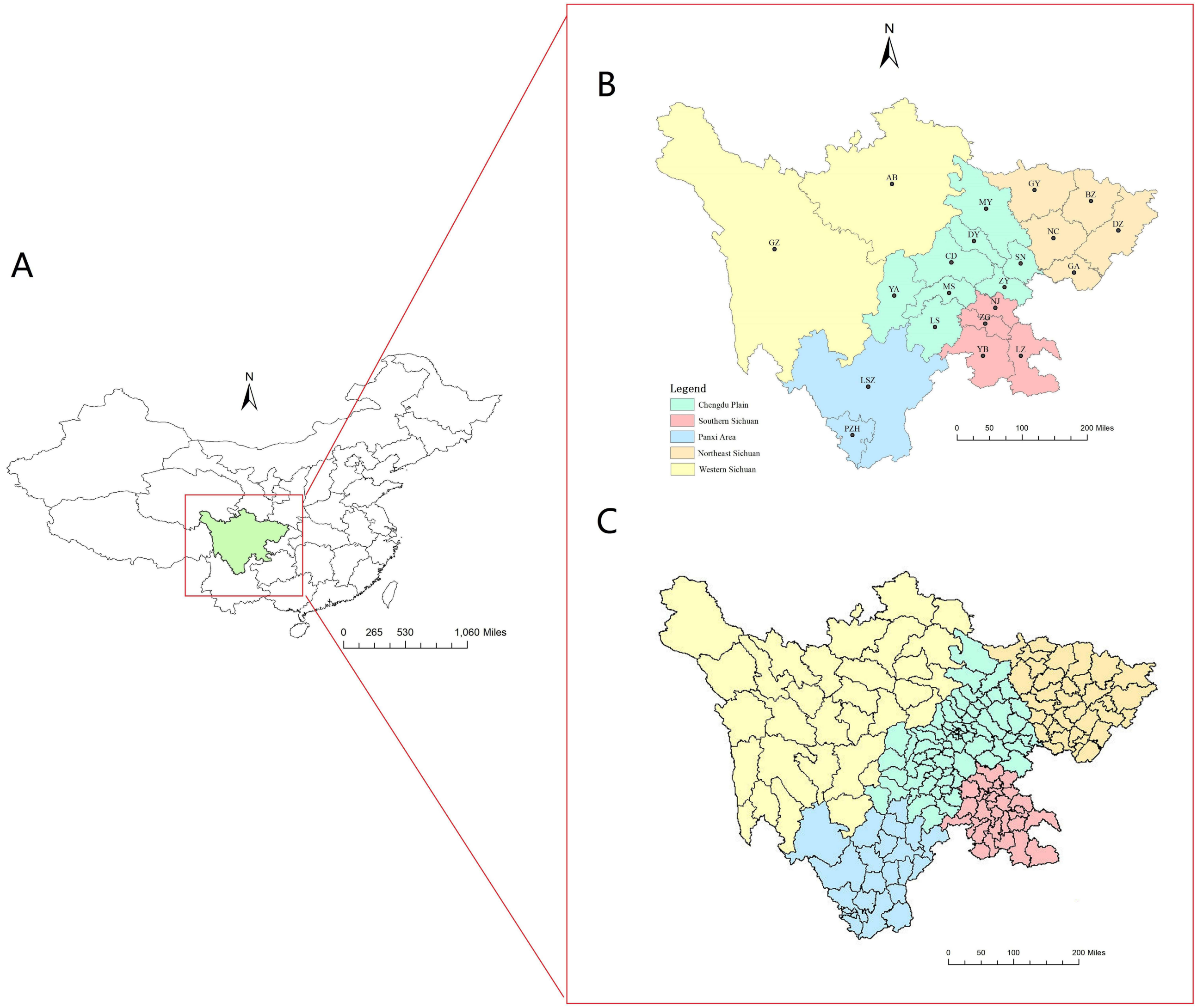

Figure 1 (**A–C**) The geographical distribution of Sichuan in China.

upward trend in TDR was found, reaching a medium epidemic level in 2019.[19,20] Compared to the 2022 cluster-sampling drug-resistance survey conducted by the Chinese CDC, this study provided excellent population coverage and covered high-epidemic areas, offering a more representative view of HIV drug-resistance prevalence in Sichuan. To establish a scientific and reliable ART program for PLWH, the Sichuan CDC conducted HIV resistance surveillance in all newly diagnosed PLWH from July 2022 to June 2023. This aimed to determine the prevalence, influencing factors, geographical differences and transmission patterns of HIV-resistant strains. These research findings provide a solid and evidence-based scientific basis for AIDS prevention and control initiatives in Sichuan.

Materials and Methods

Research Subjects

A cross-sectional survey was conducted in Sichuan to evaluate TDR. The study enrolled individuals newly diagnosed PLWH from July 2022 to June 2023. The inclusion criteria included individuals did not accept prior ARV exposure.All respondents needed to have a unique card ID and could provide written oral informed consent, while PLWH<18 years old required written or oral informed consent from their guardians. The Ethics Committee of the Sichuan Provincial Center for Disease Control and Prevention approved both the informed consent process and the research. The study followed the Declaration of Helsinki.

Sample Collection and Preservation

Plasma were collected from HIV antibody screening-positive patients. For each newly diagnosed PLWH, 5 mL of venous whole blood was collected using an 8-mL vacuum blood collection tube containing EDTA-K2. Two 1-mL plasma samples and one 1-mL lymphocyte-enriched sample were separated within 6 hours. One plasma sample was stored at 4° C for HIV supplementary tests. The other plasma sample and the lymphocyte-enriched sample from research subjects meeting the inclusion criteria were stored at −70°C for HIV genotype drug resistance testing. The investigation and sampling were carried out in a standardized manner by municipal and county-level medical institutions or centers for disease control and prevention.

Laboratory Detection and Drug Resistance Analysis

HIV complementation tests were completed by an HIV confirmation laboratory approved by the Sichuan Provincial Health Commission or a registered nucleic acid laboratory. The CD4+ T lymphocyte test was completed in a laboratory that passed the provincial or national assessment of quality control. The Center of HIV Confirmation Laboratory of Sichuan Province was responsible for HIV nucleic acid extraction, PCR amplification and HIV-1 pol gene sequencing of PLWH plasma samples. Nested PCR was used to amplify the PR-RT region of the HIV-1pol gene. The process of resistance testing, PCR amplification, primer sequence, and data analysis referred to the HIV Resistance Monitoring Strategies and Detection Technologies[6] compiled by Shao Yiming and the Guide for Resistance Testing and Quality Assurance of HIV-1 genotyping[21] (2013 edition) compiled by the Chinese CDC.

Sequence and Genotyping

The Sequencer 5.1 was used to edit, clean and splice the sequences, and then these sequences were corrected with the BioEdit 7.0 and compared with the reference sequences in the HIV database.[22] The neighbor-joining method of Mega7.0 was used to construct the phylogenetic tree with the Kimura2-parameter model, and the phylogenetic tree was constructed 1000 times. The accuracy of the genotyping was verified by a bootstrapvalue >70%. Finally, genotyping was identified in combination with the HIV BLAST tool in the HIV Database.[22] All possible sample cross-contamination, laboratory contamination, and sequence quality control methods were validated by constructing adjacent phylogenetic trees and using QC tools in the HIV Database.[22]

Resistance Analysis

The results were interpreted by using the HIV dbdatabase (https://hivdb.stanford.edu) developed by Stanford University in the United States for the resistance analysis among PLWH.[23] Twenty antiviral drugs, including NNRTIs such asdoravirine(DOR), efavirenz(EFV), etravirine(ETR), nevirapine(NVP), and rilpivirine(RPV), were used in the HIV dbdatabase; NRTIs, such asabacavir(ABC), zidovudine (AZT), emtricitabine(FTC), lamivudine(3TC), tenofovir(TDF), stavudine(D4T), and dehydroxyinosine(DDI); and PIs, such asazanavir(ATV), darunavir(DRV), lopinavir(LPV), fosanavir(FPV), indinavir(IDV), nelfinavir(NFV), sadequinavir(SQV), and tipranavir (TPV), were used. If resistance to one or more drugs reaches a low-level or higher degree, it is classified as drug resistance. Based on this criterion, the overall and categorized (NNRTI, NRTI and PI) as well as various types of drug resistance were calculated. According to the WHO's definitions of drug resistance prevalence levels, a resistance rate of <5% indicates low prevalence, 5–15% indicates moderate prevalence, and >15% indicates high prevalence.

Data Collection

The clinical and demographic data was obtained and exported using the unique card number of each PLWH from the AIDS Prevention and Control Information System of the Chinese CDC. Among the analytical variables, the regional viral suppression failure rates over the past three years were calculated as the proportion of treated PLWH with an HIV-1 viral load(VL) >1000 copies/mL, based on test results collected by all nucleic acid testing labs under the Sichuan CDC.

Statistical Analysis

SPSS 19.0 was used for the statistical analysis. Univariate and multivariate logistic regression were used to screen for factors correlated with resistance. Variables with $P<0.05$ in the univariate analysis were further entered into a multivariate stepwise logistic regression model, and variables significant at $P<0.05$ were retained in the final model. The detection rate of mutation sites among each HIV subtype was tested by the chi-square test (test level $\alpha=0.05$).

Construction of the Molecular Network of HIV-1

Gene distances between genes were calculated via HIV-TRACE under the model of Tamura Nei 93. According to the Technical Guidelines for Monitoring and Intervention of the HIV Transmission Network (2021PilotEdition),[24] a 1% gene distance was selected as the cutoff for the construction of the molecular network. The transmission network was subsequently visualized via Cytoscape 3.6.1,[25] and several molecular clusters with potential connections were identified. The network characteristics include nodes (individuals in the network), edges (links between two nodes that represent potential transmission relationships between two individuals), degrees (the number of edges connecting one node to other nodes), network size (the number of individuals in a cluster), and clusters (groups of link sequences). The gene sequences with drug resistance mutation sites were screened to construct a molecular network that was composed of only resistance gene sequences.

Spatial Analysis

The prevalence of total TDR, NNRTI, NRTI, and PI in183 counties in Sichuan was visualized via ArcGIS10.3.[26] Spatial autocorrelation analysis was used to determine the spatial distribution by combining the Z value, P value and Moran's I index. When Moran'sI is positive, the spatial data are positively correlated, indicating a clustering trend. Conversely, when the Moran I index is negative, there is a negative correlation in the spatial data, suggesting a discrete trend. When the global Moran'sI index is 0, the correlation of spatial data is not significant, indicating that the spatial data are randomly distributed. Local Moran's I and Getis–OrdGi* statistics were used for local autocorrelation analysis, and local spatial8uassociation index (LISA) clusters and cold and hot spot maps were drawn. Five patterns were included in the LISA cluster map. "High-high" means that areas with high TDR prevalence are surrounded by areas with high TDR prevalence; "low-low" means that areas with low TDR prevalence are surrounded by areas with low TDR prevalence; "low-high" means that areas with low TDR prevalence are surrounded by areas with high TDR prevalence; "high-low" means that areas with high TDR prevalence are surrounded by areas with relatively low TDR prevalence; and a value of zero means that TDR prevalence is randomly distributed across regions,namely, there is no spatial clustering pattern.

Individual links in the molecular network clusters were used to reflect whether the TDR of HIV was more prevalent within or between the regions. To show more granular transmission of TDR between cities, each transmission network was visualized, and the links between regions are represented by lines of different colors; the darker the color is, the thicker the lines are, indicating that the relationship between the regions of TDR transmission is stronger. Links within a region are indicated by blue circles, and the larger the circle is, the stronger the relationship between the TDR of HIV transmission within regions. The transport diagram was created in QGIS (version 3.10).[27]

Results

Demographic and Epidemiological Characteristics of the Subjects

The survey covered 13,619 qualified sequences. Of these, 9,963 (73.16%) were male, 6,781 (49.79%) were aged 50–69, and 8,394 (61.63%) were farmers. In terms of education, 6,366 (46.74%) had a primary school education. Marital status revealed that 6,889 (50.58%) were married or had spouses. With respect to transmission routes, 12,385 (90.94%) were heterosexual transmission cases. Additionally, 8,996 (66.05%) were HIV-infected patients, whereas 10,057 (73.85%) had no history of sexually transmitted diseases. Before treatment, 47.10% (6,415) of the patients had CD4+ T lymphocyte counts ranging from 200 to 499/mm³. The failure rate of HIV ART-induced viral suppression in the areas where 8673 (63.68%) PLWH reside ranged from 5% to 14% in the past three years. A total of 21 genotypes were detected in this study, including 7,986 cases (58.64%) of CRF07_BC, 2,955 cases (21.70%) of CRF01_AE, 1,259 cases (9.24%) of CRF08_BC, 602 cases (4.42%) of CRF85_BC, 189 cases (1.39%) of the B subtype, 165 cases (1.21%) of CRF55_01B, 57 cases (0.42%) of CRF105_108, 109 cases (0.80%) of other subtypes, and 297 cases (2.18%) of the unclassified recombinant form (URF) (Table 1).

Table 1 The Demographic and Clinical Characteristics of Newly Diagnosed HIV-Infected Individuals in Sichuan From July 2022 to June 2023

Variables	Cases	%
Demographic characteristics		
Sex		
Male	9963	73.16
Female	3656	26.84
Age at diagnosis (Years)		
15-29	1370	10.06
30-49	3338	24.51
50-69	6781	49.79
≥70	2014	14.79
<15	116	0.85
Occupation		
Farmer	8394	61.63
Housekeeping and job-waiting	2072	15.21
Others	3153	23.15
Marital status		
Unmarried	2804	20.59
Married	6889	50.58
Divorced/widowed	3926	28.83
Ethnicity		
Han	12361	90.76
Yi	1068	7.84
Others	190	1.40

(*Continued*)

Table 1 (Continued).

Variables	Cases	%
Educational level		
No Formal Education	1721	12.64
Primary school	6366	46.74
Middle school	3405	25.00
High school/technical secondary school	1000	7.34
College and above	1127	8.28
Epidemic characteristics		
Risk of HIV infection		
Heterosexual	12385	90.94
Homosexual	1088	7.99
Others	146	1.07
Clinicalcharacteristics		
Disease progression		
HIV infection	8996	66.05
AIDS	4623	33.95
Sexual Health History		
No	10057	73.85
Yes	882	6.48
Unknown	2680	19.68
Laboratory results		
CD4+ T cell count, cell/mm3 (before treatment)		
<200	4305	31.61
200-499	6415	47.10
≥500	1505	11.05
Unknown	1394	10.24
HIV subtype		
CRF01_AE	2955	21.70
CRF07_BC	7986	58.64
CRF08_BC	1259	9.24
CRF85_BC	602	4.42
CRF105_108	57	0.42
CRF55_01B	165	1.21
B	189	1.39
Others	109	0.80
URF	297	2.18
HIV Antiviral Treatment Outcomes		
The failure rate of HIV ART viralsuppression in residential area*		
<5%	3876	28.46
5–14%	8673	63.68
≥15%	1070	7.86

Notes:*was the proportion of people with a viral load greater than 1000 copies/mL among the total number of infected individuals during treatment.

Analysis of TDR and Mutation

Among 13,619 individuals, 1,486 were found to be drug resistant, with a total TDR prevalence of 10.91%. A total of 59 sites with drug resistance mutations were identified. In terms of ARV, the prevalence of TDR among NNRTIs was 8.08%, whereas the prevalence of TDR, specifically for NVP, EFV, RPV, DOR, and ETR, was 6.50%, 6.26%, 3.69%, 1.82%, and 1.42%, respectively. The primary drug resistance mutations were V179D/E/X/Q/N/A/I/T/L/K (8.38%) and K103N/S/R/Q (4.32%). For NRTI, the TDR incidence was 1.34%, and the prevalence of ABC, D4T, FTC, 3TC, DDI, and AZT were 0.84%, 0.81%, 0.67%, 0.67%, 0.50%, 0.47%, and 0.35%, respectively. The primary drug resistance mutations were

S68G/N (3.73%) and M184V/I (0.58%). For PI, the TDR prevalence rate was 2.22%, and the prevalence rates of TPV, NFV, FPV, IDV, LPV, ATV, SQV, and DRV were 1.49%, 0.81%, 0.31%, 0.14%, 0.12%, 0.09%, 0.07%, and 0.04%, respectively. The major drug resistance mutation was M46I/L (0.37%), and the accessory mutation was Q58E (1.42%). Regionally, the total TDR prevalence rates in all regions exceeded 5%, except for BZ and AB. Among them, the TDR prevalence rates in MY, SN, TA, and ZY of the Chengdu Plain; ZG, LZ, and YB of southern Sichuan; GY and GA of northeastern Sichuan; and PZH of the Panxi area exceeded 10%. DY in the Chengdu Plain and LSZ in the Panxi area exceeded 15%. For NNRTI, the TDR prevalence rate in all regions exceeded 5%, except AB and BZ. The TDR prevalence rates in LZ (southern Sichuan), SN (Chengdu Plain), and LSZ (Panxi Area) exceeded 10%. For EFV, the TDR incidence rates in CD, DY, MY, SN, YA, ZY, and MS of the Chengdu Plain; ZG, LZ, NJ, and YB of southern Sichuan; and PZH of the Panxi area exceeded 5%. The LSZ (Panxi area) exceeded 10%. The TDR incidence of NVP in various regions was similar to that of EFV. For RPV, the TDR incidence rates in DY, SN, and YA of the Chengdu Plain, LZ (southern Sichuan), and GA (northeastern Sichuan) exceeded 5%. Among the common drug-resistant mutation sites of various virus subtypes, except for the detection rates of T215, D67, and K65 in NRTI and G190, A98, and Y188 in NNRTI, which showed no statistically significant differences (P≥0.05), other drug-resistant mutation sites with higher detection rates were all statistically significant.(Tables 2–4).

Table 2 Antiviral Medications and Drug Resistance Mutation Sites of Newly Diagnosed PLWH in Sichuan Province From July 2022 to June 2023

ART drugs	Number	%	Drug Resistance Mutations	Number	%
Total	1486	10.91			
NNRTI	1101	8.08			
NVP	885	6.50	V179D/E/X/Q/N/A/I/T/L/K	1141	8.38
EFV	852	6.26	K103N/S/R/Q	588	4.32
RPV	502	3.69	E138A/G/K	271	1.99
DOR	248	1.82	V106M/I	193	1.42
ETR	193	1.42	P225H	53	0.39
			Y181C/S	53	0.39
			K101E/P/H/Q	47	0.35
			G190A/Q/S	44	0.32
			V108I/A/T	32	0.23
			Y188L/F	26	0.19
			A98G	25	0.18
			H221Y	20	0.15
			F227L	12	0.09
			L100I/F/V	10	0.07
			M230L/I	8	0.06
			K238N/T	6	0.04
			P236L	2	0.01
			Y318F	1	0.01
			L234I	1	0.01
NRTI	183	1.34			
ABC	115	0.84	S68G/N	508	3.73
D4T	110	0.81	M184V/I	79	0.58
FTC	91	0.67	K219E/N/Q/R	34	0.25
3TC	91	0.67	D67N	30	0.22
DDI	68	0.5	K70Q/T/N/E/K/R	29	0.21
AZT	64	0.47	K65R	22	0.16
TDF	48	0.35	T215I/A/D	22	0.16
			M41L	22	0.16
			A62V	17	0.12

(*Continued*)

Table 2 (Continued).

ART drugs	Number	%	Drug Resistance Mutations	Number	%
			L210W	10	0.07
			V75M/I	8	0.06
			L74I/V	7	0.05
			T69D	5	0.04
			Y115F	5	0.04
			F77L	3	0.02
			F116Y	1	0.01
			E44D	1	0.01
			Major mutations		
PI	303	2.22	M46I/L	50	0.37
TPV	203	1.49	I47A/V	7	0.05
NFV	110	0.81	D30N	3	0.02
FPV	42	0.31	V82A/L	3	0.02
IDV	19	0.14	L90LM	2	0.01
LPV	16	0.12	V32I	2	0.01
ATV	12	0.09	I54V	1	0.01
SQV	9	0.07	L76V	1	0.01
DRV	5	0.04	I84V	1	0.01
			N88S	1	0.01
			Accessory mutations		
			Q58E	193	1.42
			L33F	46	0.34
			K43T	32	0.23
			L10F/I	27	0.2
			K20T	9	0.07
			N88D	8	0.06
			M46V/T	7	0.05
			L89M/V	6	0.04
			L24F	4	0.03
			G73S/D	2	0.01
			T74P	1	0.01
			G48R	1	0.01
			F53Y	1	0.01

Table 3 Drug Resistance Rate of Newly Diagnosed PLWH in Sichuan Province From July 2022 to June 2023

Area	Region	Number	Resistance%	PI%	NRTI%	NNRTI%					
						Total%	EFV%	NVP%	RPV%	DOR%	ETR%
Chengdu Plain	CD	3876	9.26	2.19	1.24	6.4	5.26	5.65	2.76	1.73	1.6
	DY	982	15.07	4.89	0.92	9.67	6.62	6.62	5.3	1.02	2.04
	MY	604	11.26	2.15	0.83	8.61	7.62	7.78	2.98	1.32	0.83
	SN	405	14.81	3.7	1.23	10.37	6.67	6.67	5.19	1.98	0.25
	LS	592	7.26	1.18	1.18	5.41	3.55	3.72	2.87	1.52	1.18
	MS	541	8.32	1.66	1.29	6.47	6.1	6.28	3.33	3.14	1.48
	YA	218	10.55	1.38	2.29	8.26	7.8	8.26	5.96	2.29	1.38
	ZY	404	11.14	0.99	1.73	9.41	7.67	7.67	2.97	0.74	1.24

(*Continued*)

Table 3 (Continued).

Area	Region	Number	Resistance%	PI%	NRTI%	NNRTI%					
						Total%	EFV%	NVP%	RPV%	DOR%	ETR%
Northeast Sichuan	GY	118	11.02	4.24	1.69	5.08	4.24	4.24	0.85	0.85	0
	NC	203	7.88	2.46	0.99	5.42	4.93	4.93	0.99	1.48	0.49
	GA	688	12.21	1.74	1.31	9.16	4.22	4.22	6.54	0.87	1.31
	DZ	483	6.83	0.83	0.62	5.59	3.93	3.93	3.52	1.86	1.24
	BZ	91	2.2	1.1	0	1.1	0	0	1.1	0	0
South Sichuan	ZG	579	12.61	3.11	1.04	9.15	5.01	5.7	4.15	1.73	1.38
	LZ	811	14.06	1.85	2.34	10.85	9	9.12	5.3	2.84	2.59
	NJ	736	8.02	1.09	2.04	6.39	5.3	5.43	2.85	2.04	0.82
	YB	966	10.56	1.97	0.83	8.8	6.73	7.04	4.45	2.28	1.55
West Sichuan	AB	18	0	0	0	0	0	0	0	0	0
	GZ	51	5.88	0	0	5.88	3.92	3.92	1.96	0	0
Panxi	PZH	183	10.93	1.09	1.64	8.74	7.1	7.65	4.92	3.28	2.19
	LSZ	1070	16.45	2.8	2.15	13.18	11.59	11.96	3.46	2.43	1.12
Total		13619	10.91	2.22	1.34	8.08	6.26	6.5	3.69	1.82	1.42

Factors Influencing TDR

The results of the multi-factor logistic regression revealed that PLWH under 15 years of age had a higher TDR incidence than those aged 15–29 years did (OR=1.64, 95% CI=1.01~2.66). Additionally, compared with PLWH infected with CRF01_AE, individuals infected with CRF08_BC (OR=1.65, 95% CI=1.36~2.01), CRF55_01B (OR=2.06, 95% CI=1.37~3.12), the B subtype (OR=2.23, 95% CI=1.53~3.26), or other subtype viruses (OR=1.90, 95% CI=1.14~3.16) had a greater risk of TDR. Moreover, compared with regions with antiretroviral therapy viral suppression failure rates less than 5% in the past three years, areas with failure rates ranging from 5% to 14% and 15% or higher had a greater prevalence of TDR (Table 5).

Spatial Autocorrelation Analysis and Getis–Ord Gi Analysis of TDR

There were 183 county-level administrative districts in 21 municipal administrative regions in Sichuan. The overall, NNRTI, NRTI, and PI drug resistance rates in each county are presented in Figures 2A–D. In the spatial autocorrelation analysis of the TDR incidence rate, the overall and PI results were significant, indicating that their spatial autocorrelation was clustered (Figure 3A and B). The NRTI results were not significant and showed a random distribution without spatial clustering (Figure 3C). In addition, the NNRTI results suggested that there was a 90% possibility of a clustered distribution (Figure 3D).

Local Moran's I analysis revealed that the total TDR prevalence in five regions (two in the Chengdu Plain, one in the Panxi area, one in southern Sichuan, and one in northeastern Sichuan) was high-high (H-H), whereas three areas (two in western Sichuan and one in northeastern Sichuan) were low-low (L-L) (Figure 4A). For the TDR incidence of PI, three H-H clusters (one in the Panxi area, one in the adjacent areas to the Chengdu Plain and Northeast Sichuan, and one in Northeast Sichuan) and one L-L cluster (Panxi area) were detected (Figure 4B). The NRTI results revealed four H-H cluster areas (two in the Chengdu Plain, one in southern Sichuan, and one in the Panxi area) and one L-L cluster area (northeastern Sichuan) (Figure 4C). Apart from the Chengdu Plain and northeastern Sichuan, which had no H-H cluster areas of NNRTI, other areas had similar results to the total TDR rate (Figure 4D).

The local Getis Ord Gi* index depends on the differences between the observations and expectations of spatial data. spots and cold spots with different confidence intervals (CIs) were identified and marked. The hot spots indicate significant clusters of TDR in the region, whereas the cold spots indicate the opposite. Specifically, the hot spots of total TDR were mainly concentrated in the west, southwest, south, and northeast, whereas the cold spots were primarily concentrated in the north and east. The hot spot areas of PI were mainly concentrated in the southwest and northeast

Table 4 Detection Rate of Drug-Resistant Mutation Sites of HIV Subtype in Sichuan Province From July 2022 to June 2023

Major Mutation Sites	Mutation Frequency (n=13619)	CRF01_AE (n=2955)	CRF07_BC (n=7986)	CRF08_BC (n=1259)	CRF85_BC (n=602)	CRF105_108 (n=57)	CRF55_01B (n=165)	B (n=189)	Others (n=109)	URF (n=297)	X^2	*P*
PI												
M46	0.37	0.71	0.20	0.71	0.33	0.00	0.61	0.00	0.92	0.00	22.949	0.003
Q58	1.42	0.07	2.29	0.00	0.83	0.00	0.00	0.53	0.00	0.67	108.802	<0.001
NRTI												
S68	3.73	12.79	1.21	0.64	0.50	0.00	4.85	2.65	3.67	1.68	874.409	<0.001
M184	0.58	0.68	0.48	0.64	0.00	0.00	4.24	2.12	0.00	0.67	52.687	<0.001
T215	0.16	0.30	0.11	0.00	0.17	0.00	0.61	0.53	0.00	0.34	11.406	0.18
D67	0.22	0.30	0.18	0.08	0.33	0.00	1.21	0.00	0.00	0.67	14.113	0.079
K70	0.21	0.30	0.14	0.16	0.17	0.00	2.42	0.53	0.00	0.34	42.957	<0.001
K65	0.16	0.17	0.14	0.08	0.17	0.00	0.61	1.06	0.00	0.34	13.096	0.109
K219	0.25	0.58	0.14	0.08	0.83	0.00	0.00	0.00	0.00	0.00	28.267	<0.001
NNRTI												
K103	4.32	4.40	4.21	3.26	4.98	0.00	6.06	14.81	5.50	2.36	61.694	<0.001
V179	8.38	4.50	7.01	10.48	9.97	87.72	93.33	5.82	10.09	10.10	2108.561	<0.001
E138	1.99	0.85	1.24	8.90	2.33	3.51	7.88	1.59	0.00	1.01	384.953	<0.001
V106	1.42	2.30	0.83	1.91	1.00	0.00	0.00	7.41	2.75	4.04	107.15	<0.001
G190	0.32	0.61	0.21	0.40	0.17	0.00	0.00	1.06	0.00	0.34	15.442	0.051
K101	0.35	0.20	0.26	1.19	0.66	0.00	0.00	0.00	0.00	0.34	33.108	<0.001
Y181	0.39	1.22	0.15	0.00	0.66	0.00	0.61	0.00	4.59	0.00	111.21	<0.001
A98	0.18	0.17	0.16	0.16	0.00	0.00	0.00	0.00	0.00	0.00	2.338	0.969
Y188	0.19	0.27	0.21	0.08	0.00	0.00	0.00	0.00	0.00	0.00	4.726	0.786
V108	0.23	0.41	0.13	0.16	0.83	0.00	0.61	0.00	1.83	0.00	31.366	<0.001
H221	0.15	0.47	0.03	0.08	0.17	0.00	0.00	0.53	0.92	0.00	37.084	<0.001
P225	0.39	0.37	0.36	0.08	0.83	0.00	1.82	0.00	3.67	0.00	47.378	<0.001

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Table 5 TDR Factors of Newly Diagnosed PLWH in Sichuan Province From July 2022 to June 2023

Variables	TDR Number	%	The Rate of TDR%	OR	*P*	AOR	*P*
Demographic characteristics							
Sex							
Male	1057	71.13	10.61	1			
Female	429	28.87	11.73	1.12 (0.99~1.26)	0.062		
Age at diagnosis (Years)							
15-29	158	10.63	11.53	1		1	
30-49	390	26.24	11.68	1.02 (0.83~1.24)	0.883	0.96 (0.79~1.18)	0.705
50-69	686	46.16	10.12	0.86 (0.72~1.04)	0.117	0.86 (0.71~1.04)	0.119
≥70	225	15.14	11.17	0.97 (0.78~1.20)	0.745	0.96 (0.77~1.20)	0.721
<15	27	1.82	23.28	2.33 (1.47~3.69)	<0.001	1.64 (1.01~2.66)	0.046
Occupation							
farmer	938	63.12	11.17	1			
Housekeeping and job-waiting	230	15.48	11.10	0.99 (0.85~1.16)	0.923		
others	318	21.40	10.09	0.89 (0.78~1.02)	0.094		
Marital status							
Unmarried	307	20.66	10.95	1			
Married	751	50.54	10.90	0.99 (0.87~1.15)	0.946		
Divorced/widowed	428	28.80	10.90	0.99 (0.85~1.16)	0.951		
Ethnicity							
Han	1296	87.21	10.48	1			
Yi	173	11.64	16.20	1.65 (1.39~1.96)	<0.001		
Others	17	1.14	8.95	0.84 (0.51~1.39)	0.493		
Educational level							
No Formal Education	217	14.60	12.61	1			
Primary school	669	45.02	10.51	0.81 (0.69~0.96)	0.013		
Middle school	375	25.24	11.01	0.86 (0.72~1.03)	0.092		
High school/technical secondary school	104	7.00	10.40	0.80 (0.63~1.03)	0.085		
College and above	121	8.14	10.74	0.83 (0.66~1.06)	0.131		
Epidemic characteristics							
Risk of HIV infection							
Heterosexual	1350	90.85	10.90	1			
Homosexual	105	7.07	9.65	0.87 (0.71~1.08)	0.203		
Others	5	0.34	14.29	2.20 (1.48~3.29)	<0.001		
Clinicalcharacteristics							
Disease progression							
HIV infection	1001	67.36	11.13	1			
AIDS	485	32.64	10.49	0.94 (0.84~1.05)	0.26		

(*Continued*)

Table 5 (Continued).

Variables	TDR Number	%	The Rate of TDR%	OR	*P*	AOR	*P*
Sexual Health History							
No	1108	74.56	11.02	1			
Yes	98	6.59	11.11	1.01 (0.81~1.26)	0.932		
Unknown	280	18.84	10.45	0.94 (0.82~1.08)	0.401		
Laboratory results							
CD4+ T cell count, cell/mm3 (pretreatment)							
<200	443	29.81	10.29	1			
200-499	703	47.31	10.96	1.07 (0.95~1.22)	0.272		
≥500	187	12.58	12.43	1.24 (1.03~1.48)	0.022		
Unknown	153	10.30	10.98	1.08 (0.89~1.31)	0.468		
HIV subtype							
CRF01_AE	292	19.65	9.88	1		1	
CRF07_BC	808	54.37	10.12	1.03 (0.89~1.18)	0.715	0.96 (0.83~1.10)	0.542
CRF08_BC	198	13.32	15.73	1.70 (1.40~2.07)	<0.001	1.65 (1.36~2.01)	<0.001
CRF85_BC	67	4.51	11.13	1.14 (0.86~1.51)	0.354	1.13 (0.85~1.50)	0.389
CRF105_108	4	0.27	7.02	0.69 (0.25~1.92)	0.474	0.64 (0.23~1.79)	0.39
CRF55_01B	31	2.09	18.79	2.11 (1.40~3.18)	<0.001	2.06 (1.37~3.12)	0.001
B	37	2.49	19.58	2.22 (1.52~3.24)	<0.001	2.23 (1.53~3.26)	<0.001
Others	19	1.28	17.43	1.93 (1.16~3.20)	0.012	1.90 (1.14~3.16)	0.014
URF	30	2.02	10.10	1.03 (0.69~1.52)	0.904	0.99 (0.67~1.48)	0.962
HIV Antiviral Treatment Outcomes							
The failure rate of HIV ART viralsuppression in residential area							
<5%	359	24.16	9.26	1		1	
5–14%	951	64.00	10.97	1.13 (1.00~1.28)	0.061	1.16 (1.02~1.33)	0.026
≥15%	176	11.84	16.45	1.98 (1.63~2.40)	<0.001	1.84 (1.49~2.27)	<0.001

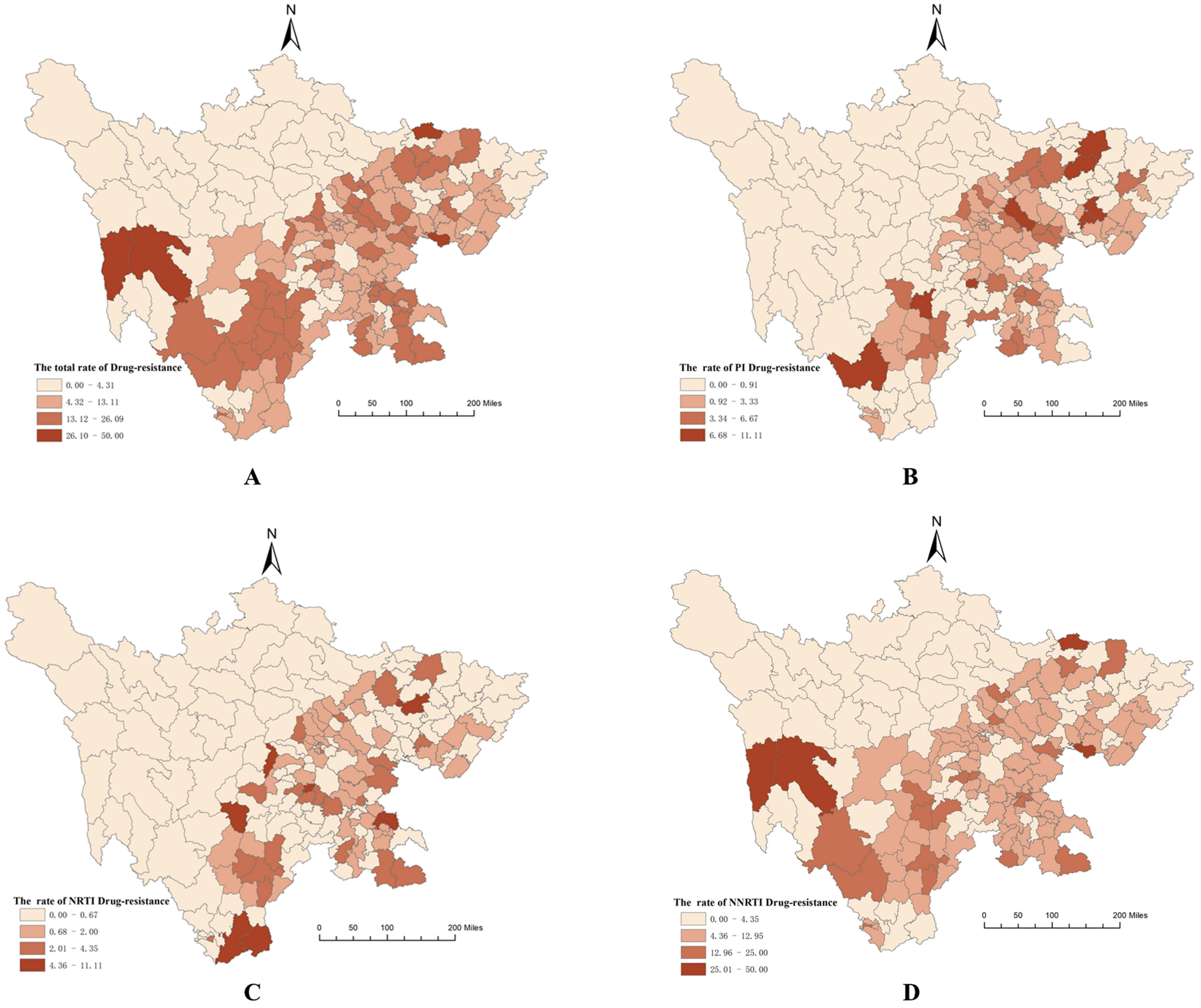

Figure 2 (**A**–**D**) The TDR incidence of newly diagnosed PLWH in Sichuan Province from July 2022 to June 2023.

regions, and NRTIs were mainly concentrated in the central and southwest regions, both of which had no cold spots. The hot spots of NNRTI were broadly similar to the total TDR, except for the central and northeast regions (Figure 5A–D).

HIV-1 Molecular Network of TDR and Transmission Characteristics

The HIV-1 molecular network contained 13,619 sequences. At a genetic distance threshold of 0.01, 7696 sequences (56.5%) were incorporated into the network, whereas among the 1486 drug-resistant PLWH, 784 (52.76%) were included (Figure 6A). There was no statistically significant difference in inclusion rates between drug-resistant patients and overall patients. Subsequently, among the 784 drug-resistant PLWH, the molecular network was constructed again via a rigorous screening process involving interconnected drug resistance patterns. There were 605 PLWH (77.17%) in the network, which comprised 181 distinct molecular clusters. The largest cluster contained 52 individuals, mainly from CD and DY, accounting for 61.54% (32/52) and 34.62% (18/52), respectively. There were 7 clusters of more than 10 individuals, mainly from the CD, DY, GA, LSZ, and other regions. Furthermore, 174 clusters with 2–9 individuals were identified, which were distributed mainly in the CD, LZ, LSZ, ZG, YB, and other regions (Figure 6B). Heterosexual transmission was the primary mode in the network, accounting for 90.08% (545/605), followed by homosexual transmission, accounting for 8.10% (49/605). In terms of drug types, 466 (77.02%) patients were resistant to NNRTIs, 138

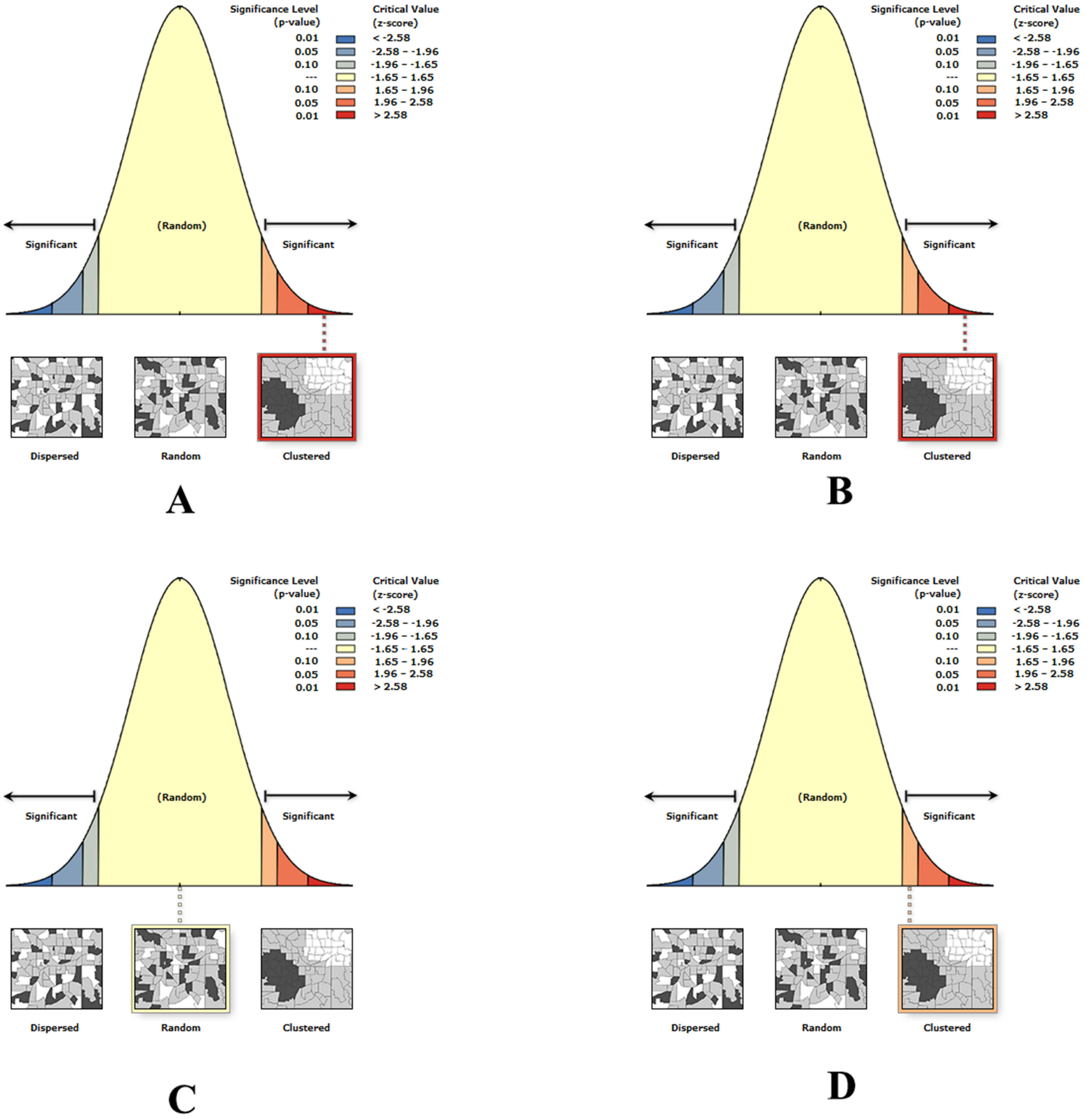

Figure 3 (**A–D**) Spatial autocorrelation analysis for the TDR incidence of newly diagnosed PLWH in Sichuan Province from July 2022 to June 2023.

(22.81%) were resistant to PIs, and 37 (6.12%) were resistant to NRTIs. Across the network, 140 NNRTI-resistant clusters were identified, with the largest cluster containing 24 individuals. For NRTI resistance, 12 clusters were found, the largest with 6 individuals. For PI resistance, 30 clusters were observed, the largest comprising 52 individuals (Figure 6C). For drug resistance mutations, the K103N, E138A, V179D, and Y181C clusters formed 39.14%, 25.36%, 10.14%, and 2.90% of the NNRTI clusters, respectively. The M184V cluster accounts for 41.67% of the NRTI clusters, whereas the Q58E cluster constitutes 63.33% of the PI clusters (Figure 6D).

Regional correlation analysis revealed a high proportion of TDR transmission between regions, reaching 54.12%. Cities such as LS (93.22%), MS (85.06%), DZ (82.76%), GY (80%), CD (67.16%), MY (66.98%), NC (66.67%), NJ (63.89%), and ZY (63.08%) presented strong interregional TDR, whereas GA (96.01%), LSZ (82.69%), ZG (78.95%),

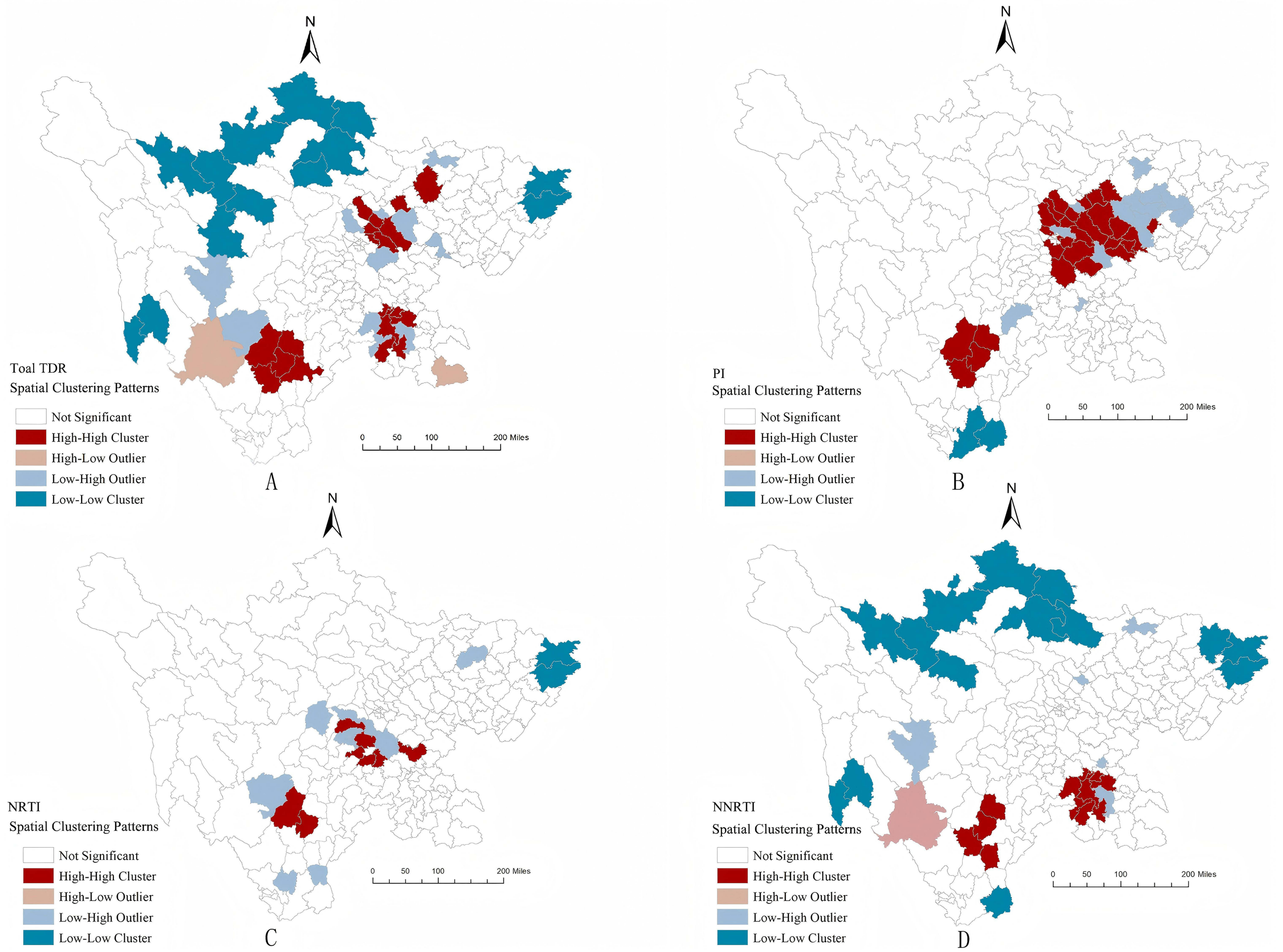

Figure 4 (**A**–**D**) Cluster by local Moran's I analysis for the TDR incidence of newly diagnosed PLWH in Sichuan Province from July 2022–June 2023.

SN (75.61%), and YA (75%) presented strong intraregional TDR. LZ, PZH, YB, and DY had similar proportions of intraregional and interregional TDR. The spatial distance transmission analysis revealed that CD and DY had the highest correlation, radiating strongly southward. Remarkably, CD had strong TDR with MS, ZY, and YB, whereas DY had strong TDR with MY, LS, LZ, MS, and YB. In the southwestern region of Sichuan, the LS, in addition to intraregional connections, had low-intensity connections (below 5) with multiple regions but formed a strong connection with the CD. Similarly, the DZ also had connections with multiple regions, forming a strong connection with the MY (Figure 7 and 8).

Discussion

The study conducted a large-scale cross-sectional drug resistance surveillance among all newly diagnosed PLWH from July 2022 to June 2023. The results revealed a moderate TDR prevalence of 10.91%, significantly higher than the 4.9% reported in the 2022 Chinese CDC sampling survey. Combined with the 2014 and 2019 Sichuan drug resistance surveillance data, a distinct upward trend was observed.[20,28] Regionally, except for BZ (2.2%) and AB (0%), other regions exhibited moderate to high TDR prevalence, with LS (16.45%) reaching a high level. NNRTIs were the main contributor to drug resistance (8.08%), with EFV and NVP having TDR prevalence rates of 5.54% and 5.73% respectively. Key resistance mutations included V179 and K103. Further analysis revealed significant differences in the detection rates of drug resistance mutation sites among different HIV-1 subtypes. Multifactorial logistic regression analysis revealed that individuals under 15 years old, infected with CRF08_BC, CRF55_01B, or subtype B viruses, and those in areas with HIV antiviral treatment viral suppression rates ≥5% in the past three years were at a significantly elevated risk of TDR. Spatial autocorrelation analysis revealed clustered distributions of TDR and PIs, identifying

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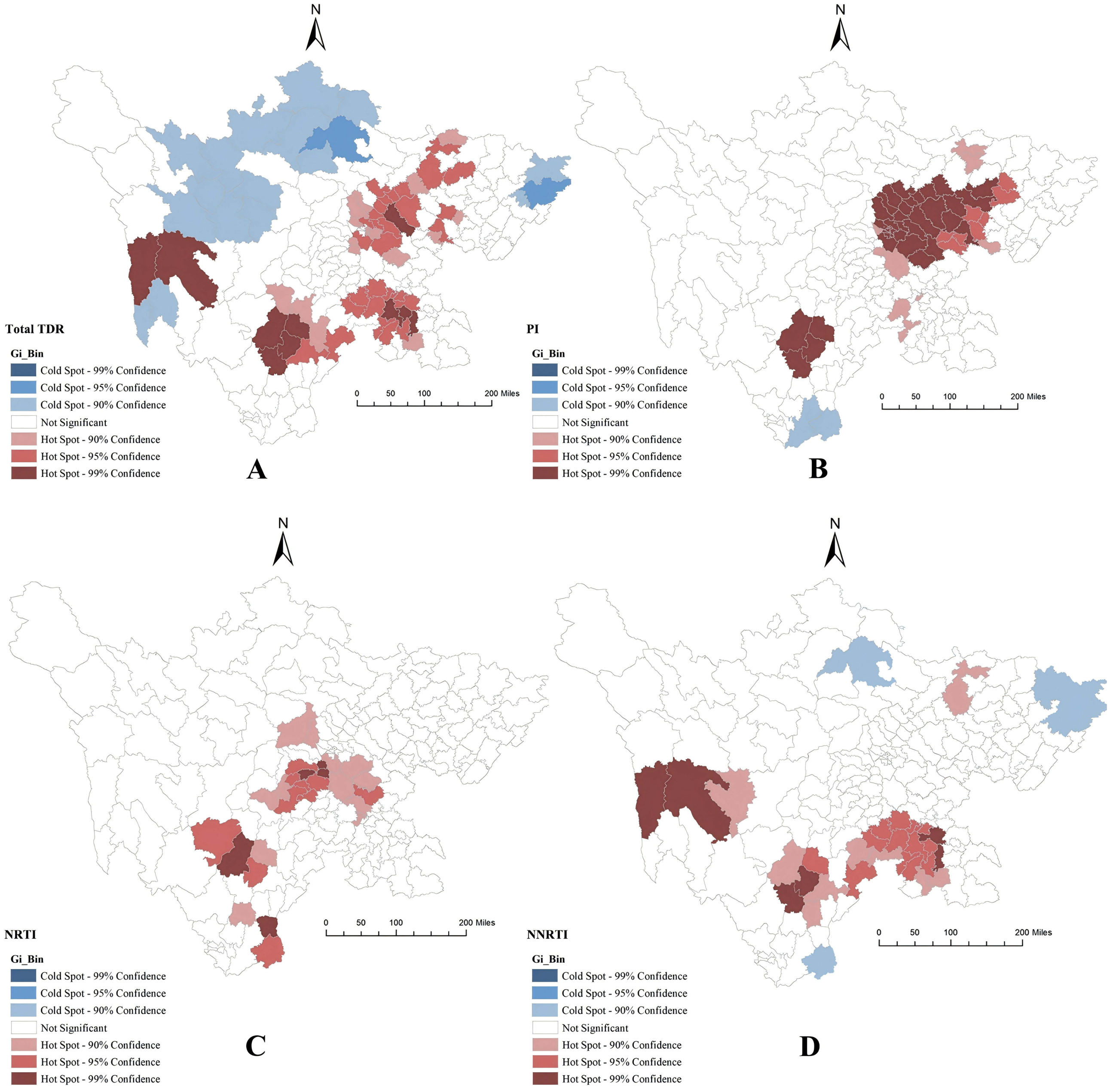

Figure 5 (**A–D**) Getis Ord Gi* analysis for the TDR incidence of newly diagnosed PLWH in Sichuan Province from July 2022 to June 2023.

geographic hotspots and high-high clusters. Molecular network analysis identified transmission clusters (52.76%) and interregional versus intraregional dynamics. Collectively, these findings emphasize the escalating threat of TDR, necessitating continuous TDR surveillance in Sichuan to monitor the prevalence of TDR in real-time, curb resistance transmission, and implement pretreatment drug resistance testing for newly diagnosed PLWH when necessary to guide clinical treatment.

In the analysis of drug types, Sichuan's NNRTI drug resistance rate rose steadily from 0% in 2014 to 8.08% in 2023, while NRTI's increased from 0% to 1.34% and PI's climbed from 2.13%to 2.22% over the same period.[8,20,28] The prevalence of NNRTI was moderate. Specifically, LZ, SN and LSZ exceeded 10%, reaching 10.85%, 10.37%, and 13.18%, respectively. These findings underscore the necessity for further investigations to identify the specific causes of the elevated NNRTI resistance rates in these regions. If the rates remain above 10%, the 2016 WHO ART guidelines recommend considering supplementary first-line regimens for PLWH initiating ART with NNRTI TDR ≥10%.[29] From

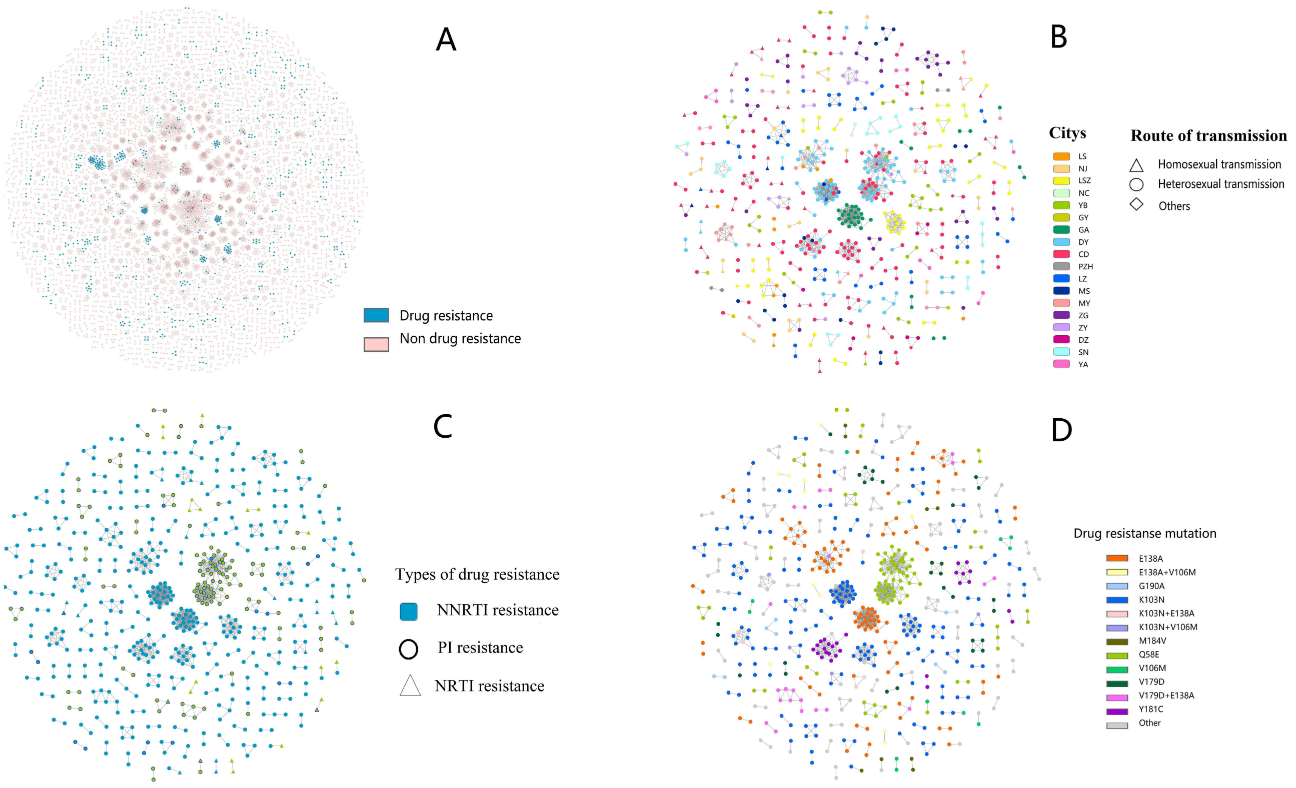

Figure 6 (**A**–**D**) HIV-1 molecular network for the TDR incidence of newly diagnosed PLWH in Sichuan Province from July 2022 to June 2023.

2014 to 2020, among 30 countries reporting to the WHO, 21 exhibited NVP or EFV TDR prevalence rates exceeding 10% in PLWH commencing first-line ART.[30] In this study, the rates for EFV and NVP were 5.54% and 5.73%, respectively, aligning with earlier surveys conducted in China in 2015 and 2018.[19,31] However, in regions such as PZH, LZ, and LSZ, the resistance rates to both drugs surpassed 7%, with LSZ exceeding 10%, higher than previous surveys in Sichuan and LS.[20,32,33] The primary resistance mutations identified in this study were V179, K103, E138, and V106. Studies from Zimbabwe, Kenya, and Argentina have identified K103 as the most common NNRTI resistance mutation associated with first-line antiviral drugs.[34–36] Given the high prevalence of NNRTI resistance, a reevaluation of national HIV treatment strategies and regimens is warranted. In 2023, China released the "National Free HIV Antiretroviral Therapy Drug Treatment Manual" (Fifth Edition),[37] incorporating RPV into the first-line treatment regimen.[38] In this study, the RPV resistance rate was 4.08%, with rates exceeding 5% in LZ, DY, SN, GA, and YA. However, less than 1% of PLWH are currently using RPV, suggesting that resistance may primarily stem from cross-resistance among PLWH receiving ART,[22] potentially contributing to the spread of RPV resistance. Therefore, RPV should be incorporated into routine drug resistance surveillance.

Furthermore, This study revealed significant differences in the detection rates of drug resistance mutations among different subtypes, such as S68 (12.79%) in CRF01_AE, Q58 (2.29%) in CRF07_BC, M184 (4.24%), K103 (4.98%), V179 (93.33%) in CRF55_01B, and V179 (87.72%) in CRF105_108, which were significantly greater than those in the other subtypes. The most common drug resistance mutation at S68 was S68G, which accounted for a relatively high proportion of NRTI mutations in CRF01_AE. It has no effect on drug sensitivity, but when combined with the major drug resistance mutation K65R, it can increase the replication fitness of K65R mutant resistant viruses.[39] Q58E does not affect sensitivity to LPV/r (a second-line treatment drug used only in China), so it can also be used as a specific marker for the spread of CRF07_BC.The most common mutation in V179 was V179D/E, which alone did not cause drug resistance, but when it occurred simultaneously with other mutation sites, such as K103R and V106I, it can cause moderate resistance to EFV and NVP, as well as low-level resistance to RPV. Some studies have also shown that, except for V179F, which was included in the 2009 version of SDRMs, other mutation sites have been excluded due to their high prevalence in certain

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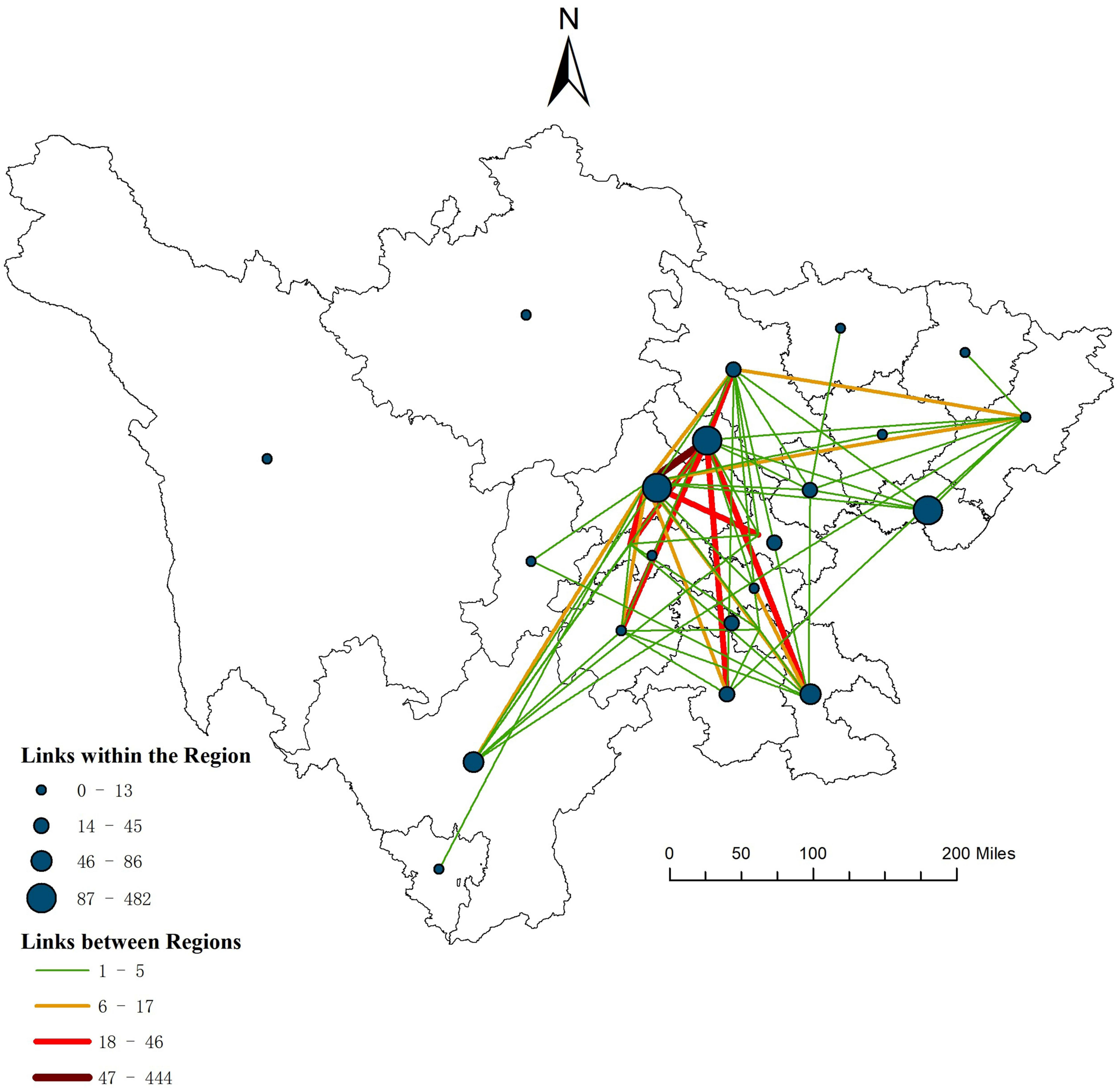
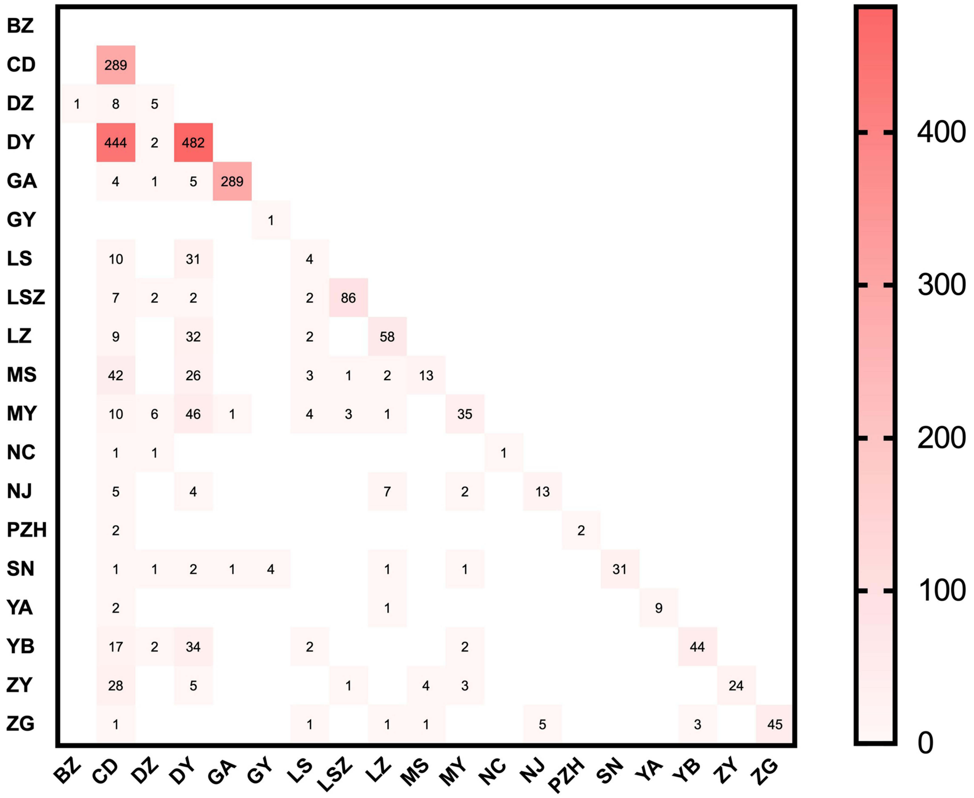


Figure 7 TDR incidence of newly diagnosed PLWH between and within regions in Sichuan.

viruses. Moreover, as a result of low genetic barrier of NNRTI, this site can be used as a specific marker for CRF55_01B.[40,41] In addition, this mutation site accounted for a relatively high proportion of CRF105_108, and whether it can be used as a specific marker for CRF105_108 requires further research. Moreover, the most prevalent drug resistance mutation observed in K103 was K103N, accounting for 93.20% (548/588). This mutation comprises more than 3% of the major subtypes in Sichuan and exceeds 14% in subtype B. K103N is highly resistant to EFV and NVP, both of which are free antiretroviral drugs in China.[42] According to studies by the WHO and China, K103N is the most prevalent mutation in countries reporting HIV drug resistance data.[42,43] Therefore, considering the variations in drug resistance mutation sites across diverse HIV-1 subtypes, incorporating molecular epidemiological findings into the development of targeted treatment strategies can significantly increase the efficacy of ART.

This study found that individuals under 15 years old, those infected with CRF08_BC, CRF55_01B, or the B subtype, and residents in regions with an HIV ART viral suppression rate of 5% or higher in the past three years face a



	BZ	CD	DZ	DY	GA	GY	LS	LSZ	LZ	MS	MY	NC	NJ	PZH	SN	YA	YB	ZY	ZG
BZ																			
CD		289																	
DZ	1	8	5																
DY		444	2	482															
GA		4	1	5	289														
GY						1													
LS		10		31			4												
LSZ		7	2	2			2	86											
LZ		9		32			2		58										
MS		42		26			3	1	2	13									
MY		10	6	46	1		4	3	1		35								
NC		1	1									1							
NJ		5		4					7		2		13						
PZH		2												2					
SN		1	1	2	1	4			1		1				31				
YA		2							1							9			
YB		17	2	34			2				2						44		
ZY		28		5				1		4	3							24	
ZG		1					1		1	1			5				3		45

Figure 8 TDR matrix diagram of HIV transmission within and between regions for newly diagnosed PLWH in Sichuan.

significantly increased risk of TDR. In the study, most PLWH under 15 were from LS (86.8%, 99/114), which had the highest TDR prevalence. Despite LS's successful AIDS prevention and control project since 2017,[44] several issues remain: school students, the majority in this age group, may show poor medication adherence due to privacy concerns; pediatric patients struggle to access consistent medical services due to schooling; local medical resources and healthcare quality are limited; and overall treatment adherence in the region is poor, with guardians' adherence indirectly affecting their children's treatment. The high TDR prevalence among young PLWH may also stem from a large proportion of female PLWH infected heterosexually, mostly young and middle-aged adults,[45] and the region's high viral suppression failure rate,[46] which increases mother-to-child transmission risk. Even if mother-to-child transmission is blocked, daily blood contact may still transmit drug-resistant viruses to children. Therefore, it is vital to ensure healthcare facilities and trained healthcare workers in rural or resource-constrained settings like LS. In addition, point-of-care resistance testing has the potential to provide rapid results, enabling clinicians to make timely treatment decisions in high risk populations. This capability is particularly valuable, and we urgently need the development of this technology. This highlights the need for ongoing vigilance over young PLWH in specific regions to prevent a decline in their quality of life, limit treatment options, and exacerbate drug resistance.[47–49]

Compared with CRF01_AE, PLWH infected with CRF55_01B had 2.07 times higher TDR (OR: 2.07, 95% CI=1.37–3.13), aligning with Chen team's research.[8] This may stem from V179, a drug resistance mutation site in CRF55_01B, often linked with E138G and causing resistance to EFV and NVP.[50] CRF55_01B's prevalence has risen in Sichuan (from 0.38% in 2019 to 1.21% now),[20] so controlling its spread may curb drug resistance. Similarly, TDR prevalence among PLWH with CRF08_BC is 1.64 times higher than those with CRF01_AE, consistent with a 2019 Sichuan survey. The E138 mutation site (8.90%) detection rate was higher than in other subtypes, mainly conferring low-level RPV resistance, necessitating strengthened RPV resistance monitoring for CRF08_BC-infected PLWH. Additionally, the drug resistance rate among PLWH with the B subtype was 2.23 times higher than those with CRF01_AE (OR: 2.23, 95% CI = 1.52–3.25). Despite recombinant viruses' prevalence in Sichuan, some studies found a short-term B subtype outbreak in certain regions (5.79% with an 83.3% detection rate),[51] mainly among those aged 50 and above and heterosexually infected. The survey showed higher K103 (14.81%) and V106 (7.41%) mutation detection rates in the B subtype, indicating a need for further drug resistance surveillance.

The higher HIV viral suppression failure rate in regions where PLWH have lived over the past three years positively correlates with a higher TDR prevalence, highlighting the need to improve ART quality. The viral suppression failure rate among PLWH has stayed around 10% in recent years, mainly due to drug resistance.[52,53] Persistent high viral loads spread drug-resistant strains, increasing resistance prevalence.[54] Poor treatment quality and drug resistance are mutually causative. Thus, it's essential to standardize ART management in high TDR prevalence areas, especially by improving healthcare personnel training and patient adherence education. This will boost antiviral treatment success and curb drug-resistant strain generation. Additionally, drug resistance monitoring should be strengthened in areas with poor ART quality, with timely feedback to guide clinicians in regimen selection.

At a genetic distance threshold of 0.01, clustered drug resistance mutations were significant, encompassing 605 PLWH (77.17%, 605/784). NNRTI mutations were predominant (77.02%), followed by PI (22.81%) and NRTI (6.12%), indicating severe TDR mutation transmission among newly diagnosed PLWH. K103N (40.83%, 247/605) was the most frequent TDR mutation, which is consistent with the findings of the 2022 national survey and Kenyan coastal research.[8,55] Owing to the good viral fitness of K103N-containing strains, which can establish stable TDR genetics, this mutation likely has circulated and spread as new infections regionally.[54] A Q58E-dominated PI resistance cluster (52 individuals) emerged in CD and DY, primarily via heterosexual and MSM((men who have sex with men). Conversely, only five (2.76%) NRTI clusters with M184V mutations were found, far fewer than in ART PLWH, underscoring viruses with the M184V mutation have limited transmission potential due to reduced replicative fitness.[54] These findings suggest integrating drug resistance surveillance with molecular network analysis in Sichuan to pinpoint TDR clusters. The molecular clusters provide a valuable framework for prioritizing contact tracing efforts. Combining this information and epidemiological data can guide targeted contact tracing, enabling health authorities to focus their resources on those most at risk of further transmission. The molecular findings can also inform targeted counseling strategies. Healthcare professionals can better educate patients about their individual risks, adherence to treatment, and preventive measures by understanding the genetic characteristics of different clusters. Furthermore, researchers and clinicians can design ART regimens that are more effective against the prevalent strains in those areas. By molecular networks with epidemiological data, precise interventions for high-risk groups and targeted measures to curb TDR can be implemented.

ArcGIS-based spatial clustering and hotspot analysis of TDR prevalence showed high-high clusters and hotspots in adjacent counties, especially in the southwest, south, central, and northeast, indicating spatial correlation likely due to similar lifestyles, convenient transport, and economic ties. More medical resources should be directed to these areas, particularly key epidemic regions like LS, CD, ZG, YB, DY, and MY, for enhanced AIDS prevention and control, such as increasing the number of laboratories, adding laboratory facilities, and raising the frequency of viral load testing as well as follow-up visits. In contrast, cold spots may require a different surveillance strategy, namely a more cost-effective and less intensive surveillance model, such as periodic sampling and data analysis. Compared with cold spots, hot spots have a more urgent need to update first-line treatment regimens to achieve virological suppression.

Geographical analysis of TDR among PLWH revealed strong ties between CD, DY, and other cities, contrasting with a 2018 Sichuan survey on PLWH with viral suppression failure.[56] Key cross-regional TDR transmitters to DY were LS (31), YB(34), LZ(32), and MS(26), despite long distances, while only neighboring CD(444) and MY(46) showed high transmission intensity, indicating distant cities can also be closely linked. Within-city TDR linkages were prominent in GA(96.01%), LS(82.69%), ZG(78.95%), and SN(75.61%), suggesting regional transmission. Deeper-rooted societal determinants are likely the driving forces behind the spatial movement patterns of PLWH harboring TDR-associated HIV strains. Prior studies show increased population exposure and mobility raise disease transmission risks, with cities acting as outbreak incubators, and urbanization may strengthen intercity disease spread.[57]

There are still limitations in our research. Firstly, variations in sample preservation conditions and delivery times across regions in 2022 resulted in a detection success rate of only 79.49% (13,619/17,132), potentially introducing bias into the findings. Secondly, as a cross-sectional study, it could not accurately capture the trends of TDR in recent years across regions or properly evaluate the impact of ART on TDR in Sichuan. Thirdly, we used the reported location of

PLWH in the AIDS prevention and treatment information system as the geographic parameter, but due to population mobility, the actual regional TDR prevalence and geographic characteristics may differ. Lastly, this study did not collect behavioral risk data which may limit interpretations of transmission dynamics. And While current guidelines advocate for INSTI-based regimens, the absence of INSTI resistance testing in our study resulted in a critical gap in data on transmitted INSTI resistance.

The findings from our 2023 TDR survey in Sichuan underscore the necessity to review and reconsider some current first-line ART regimens. Pre-treatment resistance testing allows for the selection of targeted antiretroviral therapy, which is crucial for achieving the goal of 95% viral suppression among treated individuals, within 95–95-95 target set by UNAIDS (UNAIDS, 2023) and China's Plan for Containing and Preventing AIDS (2024–2030). This approach aligns with WHO recommendations for genotype-guided therapy initiation Regions in Sichuan with higher TDR prevalence should formulate targeted strategies and allocate additional resources. Feasible interventions include expanding genotypic TDR surveillance pilot programs (enabling HIV genotypic resistance testing in medical institutions), promptly conducting resistance testing for all newly diagnosed PLWH, regularly performing viral load testing to facilitate rapid ART regimen adjustments. Additionally, enhancing the resistance interpretation capabilities of frontline healthcare workers and implementing tailored measures to improve patient adherence are crucial. Interregional joint prevention and control efforts should be strengthened, leveraging molecular network technologies and fostering enhanced communication and collaboration to curb the emergence of transmitted drug resistance.

Conclusion

The overall prevalence of transmitted HIV drug resistance in Sichuan exhibits an upward trend, reaching a moderate epidemic level, with certain cities even experiencing high-level prevalence. Resistance to NNRTIs was predominant. Through molecular epidemiology, spatial cluster analysis, subtype-specific mutation profiling, and interregional transmission network mapping, we identified elevated drug resistance among infections with CRF55_01B, CRF08_BC, and subtype B. Additionally, we pinpointed key mutations (K103N, V179D, M184V, Q58E) that are critical for understanding both the emergence of drug resistance and its transmission dynamics.Further analysis revealed an increasing rate of RPV resistance driven by cross-resistance. In regions with high HIV prevalence and strong transmission linkages, such as LS, DY, and CD, baseline drug resistance testing should be integrated into first-line ART decision-making. Municipal and county-level health authorities should enhance surveillance capacity through mobile sequencing platforms. Additionally, frontline healthcare workers require training in drug resistance analysis and molecular network techniques to implement precision interventions and adherence education for high-risk individuals, leveraging molecular network data combined with epidemiological investigations. Resistance data must also be submitted to the National CDC to inform treatment guideline updates.

Abbreviations

ARV, Antiretroviral; ART, antiretroviral therapy; WHO, World Health Organization; TDR, transmitted drug resistance; PLWH, people living with HIV; NNRTIs, nonnucleoside reverse transcriptase inhibitors; NRTIs, nucleoside reverse transcriptase inhibitors; PIs, protease inhibitors; EFV, efavirenz; NVP, nevirapine; AIDS, acquired immune deficiency syndrome; CDC, Center for Disease Control and Prevention; DOR, doravirine; ETR, etravirine; RPV, rilpivirine; ABC, abacavir; AZT, zidovudine; FTC, emtricitabine; 3TC, lamivudine; TDF, tenofovir; D4T, stavudine; DDI, dehydroxyinosine; ATV, azanavir; DRV, darunavir; LPV, lopinavir (LPV); FPV, fosanavir; IDV, indinavir; NFV, nelfinavir; SQV, sadequinavir; TPV, tipranavir; CIs, confidence intervals; WHO, World Health Organization; INSTI,integrase strand transfer inhibitor.

Data Sharing Statement

The datasets used and analysed during the current study are available from Dan Yuan on reasonable request.

Ethics Approval and Consent to Participate

The Ethics Committee of the Sichuan Provincial Center for Disease Control and Prevention approved both the informed consent process and the research (Project No: SCCDCIRB 2025-026).

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Disclosure

The authors report no conflicts of interest in this work.

References

1. Zhang T, Liao L, Shao Y, Feng Y, Ruan Y, Xing H. Relationship between drug resistance and death in HIV-infected patients receiving antiretroviral therapy - 7 PLADs, China, 2010-2019. *China CDC Wkly*. 2021;3(14):291–297. doi:10.46234/ccdcw2021.068
2. Volk JE, Nguyen DP, Hare CB, Marcus JL. HIV infection and drug resistance with unsupervised use of HIV pre-exposure prophylaxis. *AIDS Res Hum Retroviruses*. 2018;34(4):329–330. doi:10.1089/aid.2017.0285
3. Zhao Y, Zhang M, Shi CX. Mortality and virological failure among HIV-infected people who inject drugs on antiretroviral treatment in China: an observational cohort study. *Drug Alcohol Depend*. 2017;170:189–197. doi:10.1016/j.drugalcdep.2016.11.011
4. Yu F, Li Q, Wang L, et al. Drug resistance to HIV-1 integrase inhibitors among treatment-naive patients in Beijing, China. *Pharmgenomics Pers Med*. 2022;15:195–203. doi:10.2147/PGPM.S345797
5. Richardson ET, Grant PM, Zolopa AR. Evolution of HIV treatment guidelines in high- and low-income countries: converging recommendations. *Antiviral Res*. 2014;103:88–93. doi:10.1016/j.antiviral.2013.12.007
6. The strategy and methoodology for HIV drug resistance surveillance and detection, Renmin Weisheng Chubanshe,2010.
7. Shafer RW, Rhee S-Y, Pillay D. HIV-1 protease and reverse transcriptase mutations for drug resistance surveillance. *AIDS*. 2007;21(2):215–223. doi:10.1097/QAD.0b013e328011e691
8. Hi Chen, J Hao, J Hu, et al. Pretreatment HIV drug resistance and the molecular transmission network among HIV-positive individuals in China in 2022: multicenter observational study. *JMIR Public Health Surveill*. 2023;9:e50894.
9. Rhee SY, Kassaye SG, Barrow G, et al. HIV-1 transmitted drug resistance surveillance: shifting trends in study design and prevalence estimates. *J Int AIDS Soc*. 2020;23(9):e25611. doi:10.1002/jia2.25611
10. Kantzanou M, Karalexi MA, Papachristou H, et al. Transmitted drug resistance among HIV-1 drug-naïve patients in Greece. *Int J Infect Dis*. 2021;105:42–48. doi:10.1016/j.ijid.2021.02.043
11. Á É, Müller V, Mezei M, et al. Transmitted drug resistance in newly diagnosed and treatment-naïve HIV type 1-infected patients in Hungary. *J Glob Antimicrob Resist*. 2020;20:124–130. doi:10.1016/j.jgar.2019.07.014
12. Hattori J, Shiino T, Gatanaga H, et al. Characteristics of transmitted drug-resistant HIV-1 in recently infected treatment-naive patients in Japan. *J Acquir Immune Defic Syndr*. 2016;71(4):367–373. doi:10.1097/QAI.0000000000000861
13. Taffa N, Roscoe C, Sawadogo S, et al. Pretreatment HIV drug resistance among adults initiating ART in Namibia. *J Antimicrob Chemother*. 2018;73(11):3137–3142. doi:10.1093/jac/dky278
14. Kassaye SG, Grossman Z, Balamane M, et al. Transmitted HIV drug resistance is high and longstanding in metropolitan Washington, DC. *Clin Infect Dis*. 2016;63(6):836–843. doi:10.1093/cid/ciw382
15. Tchouwa GF, Eymard-Duvernay S, Cournil A, et al. Prevalence of pretreatment HIV drug resistance in Cameroon following a nationally representative WHO survey. *J Antimicrob Chemother*. 2018;73(9):2468–2474. doi:10.1093/jac/dky221
16. Wagner T, Zuckerman NS, Halperin T, et al. Epidemiology and transmitted HIV-1 drug resistance among treatment-naïve individuals in Israel, 2010-2018. *Viruses*. 2021;14(1):71. doi:10.3390/v14010071
17. Ismael N, Wilkinson E, Mahumane I, et al. Molecular epidemiology and trends in HIV-1 transmitted drug resistance in Mozambique 1999-2018. *Viruses*. 2022;14(9):1992. doi:10.3390/v14091992
18. Matías-Florentino M, Chaillon A, Ávila-ríos S, et al. Pretreatment HIV drug resistance spread within transmission clusters in Mexico City. *J Antimicrob Chemother*. 2020;75(3):656–667. doi:10.1093/jac/dkz502
19. Hao J, Zheng S, Gan M, et al. Changing proportions of HIV-1 subtypes and transmitted drug resistance among newly diagnosed HIV/AIDS individuals - China, 2015 and 2018. *China CDC Wkly*. 2021;3(53):1133–1138. doi:10.46234/ccdcw2021.251
20. Zhou C, Liang S, Li Y. Characterization of HIV-1 molecular epidemiology and transmitted drug-resistance in newly diagnosed HIV-infected patients in Sichuan, China. *BMC Infect Dis*. 2022;22(1):602. doi:10.1186/s12879-022-07576-z
21. Chinese center for disease control and prevention. Guideline for HIV-1 genotyping drug resistance testing and quality assurance. 2013.
22. HIV drug resistance database. Stanford University. Available from: https://hivdb.stanford.edu/. Accessed October 31, 2023.
23. WHO HIVDR QC Tool. Available from: https://sequenceqc-dev.bccfe.ca/who_qc. Accessed October 31, 2023.
24. Chinese Center for Disease Control and Prevention. *Technical Guide for HIV Transmission Networks Monitoring and Intervention(2021 Trial Version)*; 2021-11.
25. Wertheim JO, Kosakovsky Pond SL, Forgione LA, et al. Social and Genetic Networks of HIV-1 Transmission in New York City. *PLoS Pathog*. 2017;13(1):e1006000. doi:10.1371/journal.ppat.1006000
26. Moran PA. Notes on continuous stochastic phenomena. *Biometrika*. 1950;37(1–2):17–23. doi:10.1093/biomet/37.1-2.17
27. Estimating inequities in sanitation-related disease burden and estimating the potential impacts of pro-poor targeting. 2012.
28. Su Ling SL, Wei DongBing WD, Yang Hong YH, et al. Drug resistance threshold survey of HIV-1 in Sichuan in 2014. *Chin J Viral Dis*. 2016;6(3):194–197.
29. Consolidated guidelines on the use of antiretroviral drugs for treating and preventing HIV infection: recommendations for a public health approach, 2nd ed. World Health Organization. 2016. Available from: https://www.who.int/publications/i/item/9789241549684. Accessed October 31, 2023.

30. HIV drug resistance report 2021. World Health Organization. 2021. Available from: https://apps.who.int/iris/handle/10665/349340. Accessed October 31, 2023.
31. Chen H, Hu J, Song C, et al. Molecular transmission network of pretreatment drug resistance among human immunodeficiency virus-positive individuals and the impact of virological failure on those who received antiretroviral therapy in China. *Front Med*. 2022;9:965836. doi:10.3389/fmed.2022.965836
32. Liu L, Dong A, Liao L. Survey of pretreatment HIV drug resistance and genetic transmission network analysis among hiv patients in a high drug-use area of Southwest China. *Current HIV Research*. 2019;17:441–451. doi:10.2174/1570162X17666191128101426
33. Yuan D, Bin Y, Yiping L, et al. Prevalence and molecular epidemiology of transmitted drug resistance and genetic transmission networks among newly diagnosed people living with HIV/AIDS in a minority area, China. *Front Public Health*. 2021;2021:9.
34. Mungati M, Mhangara M, Gonese E, et al. Pre-treatment drug resistance among patients initiating antiretroviral therapy (ART) in Zimbabwe: 2008-2010. *BMC Res Notes*. 2016;9:302. doi:10.1186/s13104-016-2101-8
35. Bissio E, Barbás MG, Bouzas MB, et al. Pretreatment HIV-1 drug resistance in Argentina: results from a surveillance study performed according to WHO-proposed new methodology in 2014-15. *J Antimicrob Chemother*. 2017;72(2):504–510. doi:10.1093/jac/dkw445
36. Silverman RA, Beck IA, Kiptinness C, et al. Prevalence of pre-antiretroviral-treatment drug resistance by gender, age, and other factors in HIV-infected individuals initiating therapy in Kenya, 2013-2014. *J Infect Dis*. 2017;216(12):1569–1578. doi:10.1093/infdis/jix544
37. Chinese center for disease control and prevention. guojia mianfei aizibing kangbingdu zhiliao shouce(edition 5), Renmin Weisheng Chubanshe, 2023-6.
38. “National free HIV antiviral drug treatment manual (2023 edition)” officially released. national center for AIDS/STD control and prevention, China CDC. Available from: https://ncaids.chinacdc.cn/zxzx/zxdteff/202306/t20230616_266839.htm. Accessed October 31, 2023.
39. Svarovskaia ES, Feng JY, Margot NA. The A62V and S68G mutations in HIV-1 reverse transcriptase partially restore the replication defect associated with the K65R mutation [J]. *J Acquired Immune Deficiency Syndromes*. 2008;48(4):428–436. doi:10.1097/QAI.0b013e31817bbe93
40. Yu G, Li Y, Huang X. Genetic diversity and drug resistance of HIV-1 CRF55_01B in Guangdong, China[J]. *Curr HIV Res*. 2020;18(3):210–218. doi:10.2174/1570162X18666200415140652
41. Pang X, Tang K, He Q. HIV drug resistance and HIV transmission risk factors among newly diagnosed in- dividuals in Southwest China[J]. *BMC Infect Dis*. 2021;21:160. doi:10.1186/s12879-021-05854-w
42. Zuo L, Liu K, Liu H, et al. Trend of HIV-1 drug resistance in China: a systematic review and meta-analysis of data accumulated over 17 years (2001-2017). *EClinicalMedicine*. 2020;18:100238. doi:10.1016/j.eclinm.2019.100238
43. World Health Organization. (2019). HIV drug resistance report 2019 (WHO/CDS/HIV/19.21). Geneva: World Health Organization.
44. Jian LI. HAN Yu, LIU Yufen. HIV control and prevention in Liangshan prefecture: a literature review. *Chin J AIDS STD*. 2020;26(12):1276–1279.
45. Liu LH, Liu L, Zhang LL, et al. Analysis on premarital HIV screening results in Liangshan, Sichuan, 2014−2020. *Disease SurveillancE*. 2022;37(8):1073–1077.
46. Z Yan,L Yi-Ping,Z Chang, et al. Analysis of HIV infection and drug resistance in patients with antiviral therapy failure in SichuanProvince in 2016-2019. *Int J Lab Med*. 2022;43(8):973–978.
47. World Health Organization. *HIV Drug Resistance Report 2017*. Geneva: WHO; 2017.
48. Muri L, Gamell A, Ntamatungiro AJ. Development of HIV drug resistance and therapeutic failure in children and adolescents in rural Tanzania: an emerging public health concern. *AIDS*. 2017;31(1):61–70. doi:10.1097/QAD.0000000000001273
49. Y Li, Y Liang, L Yiping, et al. Comparative analysis of antiviral therapy and drug resistance in children and adults with HIV-1 infection in Butuo County from 2016 to 2019. *Int J Lab Med*. 2022;38(11).
50. Liu Y, Li H, Wang X, et al. Natural presence of V179E and rising prevalence of E138G in HIV-1 reverse transcriptase in CRF55_01B viruses. *Infect Genet Evol*. 2020;77:104098. doi:10.1016/j.meegid.2019.104098
51. Zhong X, Yuan D, Fan SF. Molecular network analysis of 308 newly diagnosed HIV infection and 210 ART failure patients from rural counties in Sichuan. *PLoS One*. 2024;19(2):e0298324. doi:10.1371/journal.pone.0298324
52. Dong K, Ye L, Leng Y. Yun Leng, et al.Prevalence of HIV-1 Drug Resistance among Patients with Antiretroviral Therapy Failure in Sichuan, China, 2010-2016. *Tohoku J Exp Med*. 2019;247:1–12. doi:10.1620/tjem.247.1
53. Y Dan, Y Li, GF Hong, et al. Influencing factors for therapeutic effect and drug resistance among HIV /aids patients receiving antiviral therapy in some areas of Sichuan,2014. *Mod Preventive Med*. 2016;43(13):2445–2451.
54. Hanping LI, Lin LI, Jingyun LI. Surveillance of HIV transmitted drug resistance. *China Trop Med*. 2024;24(1):6–11.
55. Hassan AS, Esbjörnsson J, Wahome E, et al. HIV-1 subtype diversity, transmission networks and transmitted drug resistance amongst acute and early infected MSM populations from coastal Kenya. *PLoS One*. 2018;13(12):e0206177. doi:10.1371/journal.pone.0206177
56. Zhou C., Kang R, Liang S. et al. Risk factors of drug resistance and the potential risk of HIV-1 transmission of patients with ART virological failure: a population-based study in Sichuan, China. *Infect Drug Resist*. 2021(14):5219–5233.
57. Alirol EP, Getaz LM, Stoll BM, et al. Urbanisation and infectious diseases in a globalised world. *Lancet Infect Dis*. 2011;11(2):131–141. doi:10.1016/S1473-3099(10)70223-1