over a decade, our understanding of its origin and dissemination patterns is limited. In this study, we employed a Bayesian phylogeographic approach to reconstruct the spatio-temporal dispersion pattern of this clade in Afghanistan and Iran for the first time. We performed a secondary data analysis on eligible HIV-1 CRF35_AD (gag and pol) sequences available in the Los Alamos HIV database (432 sequences available from Iran, 16 sequences available from Afghanistan, and a single CRF35_AD-like pol sequence available from USA). Sequences were excluded prior to analysis if they showed evidence of incorrect subtype assignment, frameshift, or drug resistance mutations, and/or stop codon positions. Subtype assignment was confirmed by maximum likelihood phylogenetic analysis. In order to reconstruct the spatio-temporal history of CRF35_AD, we used discrete Bayesian phylogeographic model in BEAST v1.8.1. Between-country viral dispersion rates were tested with Bayesian Stochastic Search Variable Selection method as implemented in SPREAD v1.0.7, and were considered as significant when Bayes factor values were >3. We checked the robustness of the key parameter estimates through a sensitivity analysis, using different priors and data subsets. The findings suggested that CRF35_AD sequences were genetically similar to parental sequences from Kenya and Uganda, and to a set of subtype A1 sequences available from Afghan refugees living in Pakistan. Our results also showed that across all phylogenies, Afghan and Iranian CRF35_AD sequences formed a monophyletic cluster (posterior clade credibility> 0.9). The divergence date of this cluster was estimated to be between 1990 and 1992. Within this cluster, a bidirectional dispersal of the virus was observed across Afghanistan and Iran. We could not clearly identify if Afghanistan or Iran first established or received this epidemic, as the root location of this cluster could not be robustly estimated. Three CRF35_AD sequences from Afghan refugees living in Pakistan nested among Afghan and Iranian CRF35_AD branches. However, the CRF35_AD-like sequence available from USA diverged independently from Kenyan subtype A1 sequences, suggesting that it may not be a true CRF35_AD lineage. The CRF35_AD viruses from Afghanistan, Iran, and Afghan refugees living in Pakistan seem to constitute a single epidemic, with multiple genetic exchanges among these populations. The date of onset for this epidemic (1990–1992) coincides with the rise of heroin production in Afghanistan (1970s). This highlights the potential role of drug trafficking in epidemic ignition in this region. Mass migration of Afghan refugees and illegal workers to Iran may be other possible contributors to among-country virus transmission.

A23 Large phylogenetic clusters highlight the HIV-1 epidemic in Canadian at risk populations

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Since 1998, Canadian provinces and the Public Health Agency of Canada collaborated to monitor the country's HIV epidemic to form the Canadian HIV Strain and Drug Resistance Surveillance Program (SDR). The program traditionally reported incidence rates and prevalence of subtypes and drug resistance among newly diagnosed and not yet treated individuals. However, modern methods in phylogenetics have not yet been implemented in SDR. Here, we attempt to further characterize the HIV-1epidemic in Canadian populations experiencing high diagnosis rates by using phylogenetic clustering to deduce transmission dynamics. HIV-1 pol sequencing and genotyping was conducted on specimens submitted to the SDR program from treatment-naive individuals newly diagnosed with HIV from 2004 to 2013. REGAv3 and COMET were used for subtyping and Sierra, the Stanford Algorithm web service, was used for drug resistance genotyping. Phylogenetic clusters were inferred using patristic distances generated from bootstrap resampled trees (FastTree2). Statistical analyses were done using R. After quality filtering, 1,009 specimens were successfully sequenced and genotyped. Of those, 907 (89.9 per cent) formed a cluster of two or more. Overall, fifty-six clusters were inferred: three clusters ($n > 100$), six clusters ($n = 29–74$), seven clusters ($n = 8–15$), thirteen clusters ($n = 3–7$), and twenty-seven clusters ($n = 2$). We investigated predictors of clustering and found that people who self-reported inject drugs significantly clustered ($P = 0.006$), independent of other epidemiological variables such as age, sex, geographical regions, and risk behaviors. Phylogenetic clustering is a valuable tool to enhance HIV surveillance and monitoring efforts. Timely identification and investigation of clusters can inform focused prevention interventions. Effective use of HIV drug resistance genotype data for public health action will require revising information flows of the current provincial surveillance system building upon recommended clinical laboratory testing practices.

A24 Role of phylogenetic analysis in epidemiological case definitions during an outbreak of HIV-1 in people who inject drugs in Ireland

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In 2015, an upsurge in acute HIV-1 subtype B infections was observed in Ireland among people who inject drugs, the majority of whom was homeless. The epidemiological investigation identified a significant association with injection of a new cathinone derivative, colloquially known as 'snow blow'. Concatenated HIV-1 polymerase and protease partial gene sequences (881 nucleotides; $n = 48$) were aligned with all subtype B patient sequences analysed at the NVRL from 2000 to 2015 ($n = 918$) and appropriate reference sequences using Bioedit v7.05. A neighbour-joining guide tree was constructed under a Kimura-2-Parameter model. Directed by this initial analysis, a maximum likelihood tree was constructed with a smaller number of more related Irish reference sequences ($n = 274$) under a HKY model of evolution and a gamma distribution. Statistical support was provided by bootstrapping with 1,000 replicates. Trees were constructed using PAUP* software version 4.0 beta10 and Mega version 7. The sequences from cases under investigation clustered within larger transmission networks of Irish people who inject drugs. More refined phylogenetic analyses confirmed that 79 per cent of the cases fell into four distinct clusters; cluster 1 ($n = 16$), cluster 2 ($n = 16$), cluster 3 ($n = 3$), and cluster 4 ($n = 3$), with high bootstrap support for each cluster (>75 per cent). There were ten outliers which branched outside the four clusters. The phylogenetic analysis largely supported the epidemiological investigation and the majority of epidemiologically linked cases were found to be contained within the same genetic clusters. In addition, this analysis identified two further possible cases and also eliminated three more 'cases'