**Abstract**

Background: Understanding the local molecular epidemiology of HIV-1 subtypes may be important for the development of HIV vaccines. Here, we describe the HIV-1 subtype distribution in a cohort of MSM in Bangkok, followed from April 2006–September 2009. Typing was performed using MHAbe, an RT-PCR-based assay using subtype-specific probes targeting 8 gene regions.

Methods: Blood samples from 278 seroprevalent and 99 seroincident cases were analyzed. Among seroincident cases, MHA was performed on the first available blood-specimen after seroconversion. Samples were "non-typeable" when hybridization occurred at >4 loci. In apparent cases of dual infection, cloning and targeted genomic sequencing was performed to verify MHA results. Among seroincident cases, non-typeable and apparent recombinant cases were verified by direct sequencing of MHA amplification products.

Results: 16 apparent dual infections, sequence verification confirmed the presence of two subtypes in 5 single infection in 2, and highly diverse subtypes in 9. Sequencing confirmed a recombinant genome in 9/10 seroincident recombinants. In 6 non-typeable cases, MHA and sequencing results were entirely concordant. After confirmation, the overall distribution of HIV-1 subtypes was: CRF01_AE, 81.6%; CRF01_AE/B recombinants, 11.0%; B, 4.9%; dual CRF01_AE and B, 1.5%; B/C/CRF01_AE recombinants, 0.6%; and B/C recombinants, 0.3%. The subtype distribution did not vary significantly by year or by serostatus (incident vs. prevalent).

Conclusions: In this cohort of HIV-1 infected Thai MSM, CRF01_AE was the most common and B was least common subtype. All remaining infections were caused by complex recombinant forms. The predominance of CRF01_AE is similar to that currently observed in HIV-1 infected Thai heterosexuals and IDU. The presence of complex recombinant forms is probably due to rapid sexual partner turnover among MSM.

**Sequence-based verification of HIV-1 subtype among apparent recombinant strains (seroincident cases)**

**Sequence-based verification of HIV-1 subtype in “non-typeable” strains identified by MHAbe (seroincident cases)**

**HIV-1 subtype distribution among MSM 2006-2009**

- The subtype distribution did not vary significantly by year or by serostatus.

**Conclusions**

- MHAbe is an alternative molecular approach that can be used to accurately characterize highly complex epidemics of HIV-1 infections.
- HIV-1 CRF01_AE and subtype B were the most and least commonly identified subtypes, respectively, in our cohort of HIV-1 infected MSM in Thailand.
- The predominance of CRF01_AE in our MSM cohort is similar to that previously observed in Thai heterosexuals and IDU.
- Presence of dual infections and emergence of complex HIV-1 recombinant forms are probably due to rapid sexual partner turnover among MSM.
- The presence of multiple complex recombinant forms may pose new challenges for future HIV vaccine development.

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