

Objectives To estimate HIV-1 incidence and cofactors for HIV-1 incidence during pregnancy and postpartum. **Design** Retrospective study among women who were HIV seronegative during pregnancy. **Methods** Mothers accompanying their infants for routine 6-week immunizations at 6 maternal child health clinics in Nairobi and Western Kenya were tested for HIV-1 after completing a questionnaire that included assessment of sociodemographics, obstetric history and HIV-1 risk perception. **Results** Of 2,135 mothers who had tested HIV-1 seronegative antenatally, 2,035 (95.3%) accepted HIV-1 re-testing at 6 weeks postpartum. Of these, 53 (2.6%) were HIV-1 seropositive yielding an estimated HIV-1 incidence of 6.8 (95% CI: 5.1-8.8) per 100 woman-years). Mothers who seroconverted were more likely to be employed (45.3% vs 29.0%, $p=0.01$), married (96.2 vs 86.6%, $p=0.04$) and from a higher HIV-1 prevalence region (60.4% in Western Kenya vs 28.8% in Nairobi, $p<0.001$). Among married women, those in polygamous relationship were significantly more likely to seroconvert (19.6% vs 6.7%, $p<0.001$). In multivariate analysis, region and employment independently predicted seroconversion. **Conclusions** Repeat HIV-1 testing in early postpartum was highly acceptable and resulted in detection of substantial HIV-1 incidence during pregnancy and postpartum period. Within prevention of mother-to-child HIV-1 transmission programs strategic approaches to prevent maternal HIV-1 acquisition during pregnancy are urgently needed.