Measles and Rubella Seroprevalence Among HIV- Infected And Uninfected Zambian Youth

Catherine G. Sutcliffe, PhD, ScM[1], **Kelly Searle, ScM**[1], **Hellen K. Matakala, Dip Biomed**[2], **Michelle Greenman, BA**[1], **Kaitlin Rainwater-Lovett, PhD, MPH**[3], **Philip E. Thuma, MD**[2], and **William J. Moss, MD, MPH**[1]

[1]Department of Epidemiology, Bloomberg School of Public Health, Johns Hopkins University, Baltimore, MD, USA

[2]Macha Research Trust, Macha Hospital, Choma, Zambia

[3]Department of Pediatrics-Infectious Diseases, Johns Hopkins School of Medicine, Johns Hopkins University, Baltimore, MD, USA

Abstract

Background—Measles and congenital rubella syndrome remain significant causes of morbidity and mortality despite available vaccines. HIV-infected youth may be at increased risk of measles because of greater waning immunity following vaccination. At a population level, they constitute a potentially large pool of susceptibles to measles and rubella. More data among HIV-infected youth in sub-Saharan Africa are needed to guide vaccination policy and control strategies.

Methods—This cross-sectional study was nested within two ongoing studies of malaria and HIV in Zambia. Dried blood spot cards from youth (5–15 years) in these studies from 2009–2013 were tested for IgG antibodies to measles and rubella viruses. HIV-uninfected youth, HIV-infected treatment-naïve youth, and HIV-infected youth receiving antiretroviral therapy (ART) were compared.

Results—617 HIV-uninfected, 144 HIV-infected treatment-naïve, and 128 HIV-infected youth receiving ART were included in the study. The proportion seropositive for measles virus was significantly higher among HIV-uninfected youth (92.5%) compared to HIV-infected treatment-naïve youth (74.1%) and HIV-infected youth receiving ART (71.9%). No differences by age were observed. The proportion seropositive for rubella virus was significantly higher among HIV-uninfected youth (54.7%) compared with HIV-infected treatment-naïve youth (41.7%) and HIV-infected youth receiving ART (49.6%), with increases observed by age for all groups.

Conclusions—Measles seroprevalence was lower among HIV-infected than uninfected youth, consistent with waning immunity following measles vaccination. HIV-infected youth would likely benefit from revaccination. Half of all youth in rural Zambia were susceptible to rubella and may need targeting for catch-up rubella campaigns when measles-rubella vaccine is introduced.

Introduction

Measles and congenital rubella syndrome remain significant causes of morbidity and mortality among children, particularly in sub-Saharan Africa.[1] While safe and effective vaccines are available, high levels of vaccine coverage are required to interrupt measles virus transmission and few sub-Saharan African countries have introduced rubella vaccine into their routine immunization schedule as of 2015.[2] Plans for rubella vaccine introduction in 49 countries by the end of 2020 are underway.[3]

Large outbreaks of measles have occurred recently,[4–6] and in sub-Saharan Africa they increasingly involve older children and adolescents.[7] Many factors contribute to these outbreaks, including HIV infection. HIV-infected children, the majority of whom live in sub-Saharan Africa,[8] can have poorer primary measles vaccine responses and a higher likelihood of waning immunity than HIV-uninfected children in the absence of treatment,[9–12] and therefore can remain susceptible to measles despite vaccination. Until recently, the impact of HIV infection on the buildup of measles susceptibles was limited by the high mortality rate of HIV-infected infants.[13,14] However, with increased access to life-prolonging therapies,[15] more HIV-infected children survive into adolescence and adulthood, thus creating pockets of susceptible individuals that may sustain measles virus transmission and jeopardize elimination efforts.[14]

While antiretroviral treatment (ART) reverses some of the immunologic damage due to HIV infection,[16,17] immunity to vaccine-preventable diseases does not appear to be restored,[18,19] as immune reconstitution in children primarily occurs with naïve $CD4^+$ T cells[20–22] and abnormalities in B cell function persist.[23,24] Consequently, these children may remain susceptible to measles and need revaccination to restore protective immunity. Recently, the World Health Organization's Strategic Advisory Group of Experts (SAGE) recommended measles revaccination for HIV-infected children receiving highly active antiretroviral therapy following immune reconstitution.[25]

Limited data are available on immunity to vaccine-preventable diseases among older HIV-infected children and adolescents with and without ART,[26–29] particularly in sub-Saharan Africa.[30] These data will be needed to guide vaccine policy on revaccination against measles and catch-up rubella vaccination. Older children are not traditionally targeted by immunization programs but are an increasingly important age group, particularly as countries consider wider age ranges for supplemental immunization activities with measles and measles-rubella containing vaccines. For rubella, failure to target older girls in susceptible age groups could lead to an increase in congenital rubella cases.[31]

The objectives of this study were to estimate and compare the prevalence of immunity to measles and rubella viruses among HIV-infected treatment-naïve youth, HIV-infected treatment-experienced youth, and HIV-uninfected youth in rural Zambia.

Material and Methods

Study Setting and Population

The study was conducted in the catchment area of Macha Hospital in Choma District, Southern Province, Zambia. This area is populated by traditional villagers living in scattered homesteads, characteristic of much of rural sub-Saharan Africa. The prevalence of HIV infection in Choma District was estimated to be 15.7% in 2010.[32] Zambia had a large measles outbreak in 2010–2011 but has since reported few cases after a nationwide supplementary measles immunization campaign in 2012 targeting children from 9 months to 14 years of age. Reported vaccine coverage in Southern Province was 86% for measles and 69% were estimated to be fully vaccinated (BCG, measles, and three doses each of DPT-HepB-Hib and polio vaccine) in 2013.[33] Zambia plans to introduce rubella vaccine in 2016.

Patients and Samples

Samples for testing antibodies to measles and rubella viruses were selected from participants within two ongoing studies conducted in Macha from 2009–2013. Three groups of youth 5–15 years of age were selected for comparison: 1) HIV-uninfected youth; 2) HIV-infected treatment-naïve youth; and 3) HIV-infected youth receiving ART. Measles vaccination status of the study children was not known.

All studies were approved by the Institutional Review Board at the Johns Hopkins Bloomberg School of Public Health, a local research ethics committee and the Government of Zambia Ministry of Health.

Source of HIV-uninfected youth—In 2007, community-based serial cross-sectional surveys were initiated around Macha Hospital to determine changes in the prevalence of malaria parasitemia.[34,35] Satellite images were used to construct a sampling frame and approximately 24 households were randomly selected to be enrolled in each cross-sectional survey conducted every other month. All individuals residing within a household were eligible to participate. Permission was obtained from the head of household and written informed consent was obtained from the individual or parent. For each participant, a questionnaire was administered to collect information on demographics, health-seeking behavior and use of medications, and a blood sample was collected by finger prick on filter paper (Whatman, Protein Saver card 903, Piscateway, New Jersey), dried overnight and stored individually with desiccant in a sealed plastic bag at −20°C.

For this study, samples were randomly selected from participants 5–15 years of age who enrolled from 2009–2013, had at least three filter paper spots available, and did not report use of ART. The target sample size was four HIV-uninfected youth per HIV-infected youth. HIV testing was not conducted as part of the study and all participants not reporting ART were presumed HIV-uninfected given the low prevalence of HIV infection expected in a population-based sample of youth.

Source of HIV-infected youth—In 2007, a cohort study was initiated and all HIV-infected children younger than 16 years attending the Macha HIV clinic were eligible for enrollment.[36,37] After obtaining written informed consent from a parent or guardian and

assent from children 8–15 years of age, children were seen at study visits every three months, at which time a questionnaire was administered, the child was examined and a blood sample was collected. Beginning in 2009, blood spots were collected on filter paper (Whatman, Protein Saver card 903) at enrollment and after ART initiation. The blood spot cards were dried overnight and stored individually with desiccant in a sealed plastic bag at −50°C.

For the group of HIV-infected treatment-naïve youth, all available samples were selected from the 163 treatment-naïve youth 5–15 years of age at enrollment from 2009–2013. For the group of HIV-infected youth receiving ART, the first available sample was selected from the 186 youth 5–15 years of age who had been receiving ART for 9 to 24 months from 2009–2013. Thirty-five youth contributed samples to both groups.

To evaluate the impact of ART on measles and rubella immunity, pairs of samples were obtained for children contributing to both the treatment-naïve and ART groups. Additional samples were selected for HIV-infected youth receiving ART who had a pre-ART sample available at study enrollment prior to 5 years of age.

Laboratory Methods

Serum eluted from dried blood spots (DBS) was tested by enzyme immunoassay (EIA) for IgG antibodies to measles and rubella virus (Enzygnost, Siemens, Germany) at the Clinical Research Laboratory at Macha Research Trust. Two dried blood spots per participant were used. Samples from each of the three groups were randomly selected for testing on each plate. Serum was eluted from DBS using 250 μL of elution buffer according to protocol 6 outlined by Mercader et al.[38] 50 μL of eluted sample was transferred to labeled 96-well plates and sealed. Samples not tested immediately were stored at 4°C overnight for testing the following day. The EIA was performed according to the manufacturer's protocol. The optical density (OD) for each sample was read at 450 nM using a BioTek ELx800 microplate reader. Corrected OD differences (cOD) were calculated using the OD differences (antigen-control well), the mean of the positive and negative references and the nominal value as specified by the manufacturer.

Volume adjustment—DBS from a known measles and rubella positive individual were eluted and tested for IgG antibodies to measles and rubella virus using serum elution volumes ranging from 5μL to 50μL. Linear regression was used to determine the effect of volume on OD value (estimated mean increase of 0.03 in OD value per 1μL increase in volume for the antigen well). The results were the same for measles and rubella tests. The regression coefficient was used to calculate adjusted OD values for the antigen well.

Statistical Analysis

Ten percent of measles samples and 3% of rubella samples had less than the required 50 μL of sample and adjusted cOD values were calculated (results based on unadjusted cOD values are presented in Supplemental Digital Content 1). For rubella, samples with cOD values ≥ 2.5 (12% of samples) were assigned a value of 2.5. For all samples with cOD values >0.1, measles (mIU/mL) and rubella antibody concentrations (IU/mL) were calculated using the

alpha method, as specified by the manufacturer. cOD values between 0.1 and 0.2 were considered equivocal and cOD values >0.2 were considered positive for antibodies to measles and rubella viruses. A second definition was also used in which cOD values 0.1 were considered positive.

Measles and rubella seroprevalence was compared between the three groups of HIV-infected and uninfected youth using chi-square tests and log-binomial regression to adjust for age. Antibody concentrations were compared using Wilcoxon rank sum tests. Risk factors for susceptibility (cOD 0.2) were assessed separately for HIV-infected treatment-naïve youth and youth receiving ART using log-binomial regression.

Results

Between 2009 and 2013, 617 HIV-uninfected, 144 HIV-infected treatment-naïve, and 128 HIV-infected youth receiving ART were enrolled and had samples available for testing. The characteristics of each group are described in Table 1.

Measles

Seroprevalence—The proportion seropositive (cOD>0.2) for measles virus was significantly higher among HIV-uninfected youth (92.5%) compared to HIV-infected treatment-naïve youth (74.1%; age-adjusted prevalence ratio [PR]: 0.80, 95% CI: 0.73, 0.89) and HIV-infected youth receiving ART (71.9%; age-adjusted PR: 0.78, 95% CI: 0.70, 0.87) (Table 2). The proportion of youth with equivocal results was significantly higher for the two groups of HIV-infected youth (treatment-naïve=16.1%; receiving ART=15.6%) compared to HIV-uninfected youth (4.3%), regardless of age. As a result, the difference in the proportion seropositive between the groups was diminished when a lower cutoff for positivity (cOD 0.1) was considered (age-adjusted PR for HIV-infected treatment-naïve youth: 0.93, 95% CI: 0.88, 0.99; age-adjusted PR for HIV-infected youth receiving ART: 0.91, 95% CI: 0.85, 0.97; compared to HIV-uninfected youth).

Within HIV-infected groups, no significant differences by age were observed (Figure 1). Within all age categories, the proportion seropositive (OD >0.2) was significantly lower among HIV-infected youth, whether treatment-naïve or receiving ART.

Impact of ART—Among HIV-infected youth receiving ART, 54 had a corresponding pre-ART sample at study enrollment available for testing (18 were obtained prior to 5 years of age). The median age at study enrollment was 6.3 years (IQR: 4.7–9.3, range: 2.3–14.7) and the median time between pre- and post-ART samples was 1.3 years (IQR: 1.0–1.8). Thirty-two youth (two seronegative and 30 seropositive) maintained their original serostatus after initiating ART. Among the nine seronegative youth at study enrollment, 5 (56%) were seropositive and 2 (22%) had equivocal results after initiating ART. Among the 39 seropositive youth at study enrollment, 3 (8%) were seronegative and 6 (15%) had equivocal results. Among the 6 youth with equivocal results at study enrollment, all 6 (100%) were seropositive after initiating ART.

Risk factors for susceptibility among HIV-infected youth—No demographic, clinical, or immunologic characteristics were significantly associated with susceptibility in either group (Supplemental Digital Content 2).

Rubella

Seroprevalence—The proportion seropositive (cOD>0.2) for rubella virus was significantly higher among HIV-uninfected youth (54.7%) compared to HIV-infected treatment-naïve youth (41.7%) and HIV-infected youth receiving ART (49.6%) (Table 2), although the differences were smaller than for measles. After adjusting for age, the proportion seropositive for rubella remained marginally higher among HIV-uninfected youth compared to HIV-infected treatment-naïve youth (PR: 0.84, 95% CI: 0.69, 1.01) but no difference was observed for HIV-infected youth receiving ART (PR: 0.98, 95% CI: 0.84, 1.15). The proportion of youth with equivocal results was significantly higher for the two groups of HIV-infected youth (treatment-naïve=11.8%; receiving ART=7.9%) compared to HIV-uninfected youth (1.1%). As a result, there were no significant differences in the proportion seropositive between the groups when a lower cutoff for positivity (cOD 0.1) was considered (age-adjusted PR for HIV-infected treatment-naïve youth: 0.96, 95% CI: 0.83, 1.11; age-adjusted PR for HIV-infected youth receiving ART: 1.07, 95% CI: 0.93, 1.22; compared to HIV-uninfected youth).

Within HIV-infected groups, the proportion seropositive increased with age (Figure 1), although the trend was only significant for HIV-uninfected youth. Within age categories, the proportion seropositive (cOD>0.2) tended to be lower among HIV-infected treatment-naïve youth but not among HIV-infected youth receiving ART.

Impact of ART—Among HIV-infected youth receiving ART, 53 had a corresponding pre-ART sample at study enrollment available for testing. Twenty-eight youth (16 seronegative, 1 equivocal, and 11 seropositive) maintained their original serostatus after initiating ART. Among the 30 seronegative youth at study enrollment, 10 (33%) were seropositive and 4 (13%) had equivocal results after initiating ART. Among the 15 seropositive youth, 4 (27%) were seronegative after initiating ART. Among the 8 youth with equivocal results, 6 (75%) were seronegative and 1 (13%) was seropositive after initiating ART.

Risk factors for susceptibility among HIV-infected youth—Younger age was significantly associated with rubella susceptibility for both HIV-infected treatment-naïve youth (PR: 0.92; 95% CI: 0.87, 0.98) and HIV-infected youth receiving ART (PR: 0.91; 95% CI: 0.85, 0.98) (Supplemental Digital Content 2).

Discussion

In this cross-sectional study, HIV-infected youth in rural Zambia were less likely to be seropositive for measles and rubella viruses than HIV-uninfected youth. Based on these results, approximately 25% and 50% of HIV-infected youth in this area were susceptible to measles and rubella viruses, respectively. The more substantial differences in measles seroprevalence likely reflect higher rates of primary or secondary measles vaccine failure,

whereas the smaller differences in rubella seroprevalence may reflect waning immunity following wild-type virus infection.

Increasing numbers of HIV-infected youth could contribute to lower levels of population immunity in areas with a high HIV prevalence.[14] Similar to other studies,[26,39] HIV-infected youth in this study were less likely to be seropositive for measles virus than HIV-uninfected youth, despite presumed vaccination during childhood, possible revaccination during the supplemental immunization activities, and immune reconstitution with ART. Lower measles vaccine coverage among HIV-infected youth is an alternative explanation, although this is unlikely given that these children were receiving intensive care. In addition, lower antibody concentrations were found in this group, leading to a higher proportion of youth in the equivocal range compared to uninfected youth. It is not possible to determine if this was due to poorer response to primary vaccination or waning immunity over time. It is also unclear whether these children would develop clinical measles or rubella upon re-exposure as they may have an anamnestic response or cellular immunity that would confer some protection.[18] These results support existing studies suggesting that ART does not restore immunity to measles virus and that HIV-infected children and adolescents could benefit from revaccination,[18,19,40] as recently recommended by the World Health Organization.[25] This group could easily be targeted for revaccination through routine care, as they are frequently seen at healthcare facilities, or through inclusion of youth in supplemental immunization activities.

All HIV-infected youth in this study initiated ART after the age at which they would have received measles vaccination. With early infant diagnosis more widely available, infants are now initiating ART before their first dose of measles vaccine. Although data are limited, immune responses among these infants appear to be better and more similar to those observed among healthy HIV-uninfected infants.[41,42] Early initiation of ART preserves normal development and maintenance of memory B cells,[42] and these improvements appear to be sustained into adolescence.[26] As more children initiate ART in infancy, their immune function and responses to vaccines will need to be documented to guide policies.

In this study, approximately half of all adolescents, regardless of HIV or ART status were susceptible to rubella virus. As rubella vaccines have not yet been introduced in Zambia, immunity in this study was acquired through exposure to wild-type virus. As expected, the proportion seropositive increased with age, but was lower than previous reports in the region.[43–45] This may be due to differences in urban and rural populations, with lower population densities and risk of exposure in rural areas. Introduction of measles-rubella vaccines may increase the age of infection,[31] therefore a large number of susceptible youth is a cause for concern. This group may need to be targeted for catch-up immunization campaigns when introducing measles-rubella vaccines into the national immunization program.

This study had several limitations. First, youth in the control group were not tested for HIV and were presumed uninfected. While we expect the HIV prevalence among youth in the general population in this area to be low, it is possible that some youth were infected with HIV and that the measles and rubella seroprevalence in HIV-infected youth was

underestimated. Second, information on vaccination status was not known. Antibodies to measles virus were presumed to be primarily vaccine induced but some children may have been exposed to wild-type virus. In addition, supplemental immunization activities occurred before and during the study period, therefore antibodies levels may be the result of multiple vaccinations. As described above, lower measles vaccine coverage among HIV-infected youth could also explain the differences in seroprevalence. Regardless of the mechanism, these children would likely benefit from revaccination. Third, all HIV-infected youth survived to enter the study and initiate ART at older ages. Their experiences may be different from children with more rapid disease progression and children initiating ART in infancy. Fourth, measles antibody levels were measured by EIA rather than the gold standard plaque reduction neutralization assay. While these two assays have good agreement, EIAs are less sensitive at lower antibody levels,[46] which may have led us to underestimate measles seroprevalence among all groups. Fifth, DBS were used that had been stored for up to four years. Several studies have documented the concordance of results for antibody levels measured from DBS and serum,[47–49] even after being stored at $-20°C$ for up to two years.[50,51] It is possible that longer term storage resulted in some sample degradation, which may also have led to underestimation of measles and rubella seroprevalence among all groups. Lastly, the number of HIV-infected youth in each treatment group was small, which limited our ability to evaluate risk factors for susceptibility.

Global efforts have succeeded in decreasing morbidity and mortality related to measles and rubella viruses. Large outbreaks in the past few years, however, have demonstrated that high levels of population immunity are required and pockets of susceptible individuals can jeopardize control efforts. HIV-infected youth represent an important group that needs to be considered in these control efforts. These results support recommendations to revaccinate HIV-infected children against measles after initiation of ART and for inclusion of youth in supplemental immunization activities and catch-up campaigns for both measles and rubella.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

References

1. WHO. Global measles and rubella. Strategic Plan 2012–2020. Geneva, Switzerland: World Health Organization; 2012.

2. Regional Measles-Rubella Technical Advisory Group. MR introduction and rubella epidemiology in AFRO; Paper presented at: 2015 Africa Technical Advisory Group (TAG) Meeting; June 2–3, 2015; Nairobi, Kenya.

3. GAVI. Rubella Vaccine Support. 2012 [Accessed February 20, 2015] http://www.gavialliance.org/support/nvs/rubella/.

4. WHO. [Accessed February 20, 2015] Measles outbreaks: Regions of the Americas, Europe and Africa. 2011. http://www.who.int/csr/don/2011_10_07/en/index.html

5. WHO Western Pacific Region. [Accessed February 20, 2015] Measles-Rubella Bulletin. 2014. http://www.wpro.who.int/immunization/documents/mrbulletinvol8issue12.pdf

6. Zipprich J, Winter K, Hacker J, Xia D, Watt J, Harriman K. Measles Outbreak — California, December 2014–February 2015. Morbidity and Mortality Weekly Report (MMWR). 2015; 64(6): 153–154. [PubMed: 25695321]

7. Goodson JL, Masresha BG, Wannemuehler K, Uzicanin A, Cochi S. Changing epidemiology of measles in Africa. J Infect Dis. 2011; 204(Suppl 1):S205–S214. [PubMed: 21666163]

8. UNAIDS. The Gap Report. Geneva, Switzerland: Joint United Nations Programme on HIV/AIDS; 2014.

9. Moss WJ, Scott S, Mugala N, et al. Immunogenicity of standard-titer measles vaccine in HIV-1-infected and uninfected Zambian children: an observational study. J Infect Dis. 2007; 196(3):347–355. [PubMed: 17597448]

10. Arpadi SM, Markowitz LE, Baughman AL, et al. Measles antibody in vaccinated human immunodeficiency virus type 1-infected children. Pediatrics. 1996; 97(5):653–657. [PubMed: 8628602]

11. Helfand RF, Witte D, Fowlkes A, et al. Evaluation of the immune response to a 2-dose measles vaccination schedule administered at 6 and 9 months of age to HIV-infected and HIV-uninfected children in Malawi. J Infect Dis. 2008; 198(10):1457–1465. [PubMed: 18828743]

12. Thaithumyanon P, Punnahitananda S, Thisyakorn U, Praisuwanna P, Ruxrungtham K. Immune responses to measles immunization and the impacts on HIV-infected children. The Southeast Asian journal of tropical medicine and public health. 2000; 31(4):658–662. [PubMed: 11414407]

13. Newell ML, Coovadia H, Cortina-Borja M, Rollins N, Gaillard P, Dabis F. Mortality of infected and uninfected infants born to HIV-infected mothers in Africa: a pooled analysis. Lancet. 2004; 364(9441):1236–1243. [PubMed: 15464184]

14. Scott S, Mossong J, Moss WJ, Cutts FT, Cousens S. Predicted impact of the HIV-1 epidemic on measles in developing countries: results from a dynamic age-structured model. Int J Epidemiol. 2008; 37(2):356–367. [PubMed: 18234739]

15. WHO. Global update on the health sector response to HIV, 2014. Geneva, Switzerland: World Health Organization; 2014.

16. Sutcliffe CG, van Dijk JH, Bolton C, Persaud D, Moss WJ. Effectiveness of antiretroviral therapy among HIV-infected children in sub-Saharan Africa. Lancet Infect Dis. 2008; 8(8):477–489. [PubMed: 18652994]

17. Ciaranello AL, Chang Y, Margulis AV, et al. Effectiveness of pediatric antiretroviral therapy in resource-limited settings: a systematic review and meta-analysis. Clin Infect Dis. 2009; 49:1915–1927. [PubMed: 19916798]

18. Rainwater-Lovett K, Nkamba HC, Mubiana-Mbewe M, Bolton-Moore C, Moss WJ. Changes in measles serostatus among HIV-infected Zambian children initiating antiretroviral therapy before and after the 2010 measles outbreak and supplemental immunization activities. J Infect Dis. 2013; 208(11):1747–1755. [PubMed: 23911708]

19. Sutcliffe CG, Moss WJ. Do children infected with HIV receiving HAART need to be revaccinated? Lancet Infect Dis. 2010; 10(9):630–642. [PubMed: 20797645]

20. Gibb DM, Newberry A, Klein N, de Rossi A, Grosch-Woerner I, Babiker A. Immune repopulation after HAART in previously untreated HIV-1-infected children. Paediatric European Network for Treatment of AIDS (PENTA) Steering Committee. Lancet. 2000; 355(9212):1331–1332. [PubMed: 10776748]

21. Vigano A, Vella S, Saresella M, et al. Early immune reconstitution after potent antiretroviral therapy in HIV-infected children correlates with the increase in thymus volume. AIDS. 2000; 14(3):251–261. [PubMed: 10716501]

22. Rainwater-Lovett K, Nkamba H, Mubiana-Mbewe M, Moore CB, Margolick J, Moss WJ. Changes in cellular immune activation and memory T-cell subsets in HIV-infected Zambian children receiving HAART. J Acquir Immune Defic Syndr. 2014; 67(5):455–462. [PubMed: 25226208]

23. Moir S, Fauci AS. Pathogenic mechanisms of B-lymphocyte dysfunction in HIV disease. J Allergy Clin Immunol. 2008; 122(1):12–19. [PubMed: 18547629]

24. Moir S, Fauci AS. B cells in HIV infection and disease. Nat Rev Immunol. 2009; 9(4):235–245. [PubMed: 19319142]

25. Meeting of the Strategic Advisory Group of Experts on immunization, October 2015 - conclusions and recommendations. Wkly Epidemiol Rec. 2015; 90(50):681–699. [PubMed: 26685390]

26. Siberry GK, Patel K, Bellini WJ, et al. Immunity to Measles, Mumps, and Rubella in US Children With Perinatal HIV Infection or Perinatal HIV Exposure Without Infection. Clin Infect Dis. 2015; 61(6):988–995. [PubMed: 26060291]

27. Abzug MJ, Qin M, Levin MJ, et al. Immunogenicity, immunologic memory, and safety following measles revaccination in HIV-infected children receiving highly active antiretroviral therapy. J Infect Dis. 2012; 206(4):512–522. [PubMed: 22693229]

28. Sticchi L, Bruzzone B, Caligiuri P, et al. Seroprevalence and vaccination coverage of vaccine-preventable diseases in perinatally HIV-1-infected patients. Hum Vaccin Immunother. 2014; 11(1)

29. Morris LE, Posada R, Hickman CJ, et al. Susceptibility to Measles Among Perinatally HIV-Infected Adolescents and Young Adults. Journal of the Pediatric Infectious Diseases Society. 2015; 4(1):63–66. [PubMed: 25844166]

30. Newman LP, Njoroge A, Ben-Youssef L, et al. Measles seropositivity in HIV-infected Kenyan children on antiretroviral therapy. Pediatr Infect Dis J. 2014; 33(8):843–845. [PubMed: 24618938]

31. World Health Organization. Rubella vaccines: WHO position paper. Weekly Epidemiological Record. 2011; 29:301–316.

32. Zambia National HIV/AIDS/STI/TB Council. Online National AIDS Reporting System. District Profile. 2010 [Accessed July 7, 2015] http://www.zambianacmisonline.org/index.php.

33. Central Statistical Office (CSO) [Zambia] MoHMZ, and ICF International. Zambia Demographic and Health Survey 2013–14. Rockville, Maryland, USA: Central Statistical Office, Ministry of Health, and ICF International; 2014.

34. Moss WJ, Hamapumbu H, Kobayashi T, et al. Use of remote sensing to identify spatial risk factors for malaria in a region of declining transmission: a cross-sectional and longitudinal community survey. Malar J. 2011; 10:163. [PubMed: 21663661]

35. Sutcliffe CG, Kobayashi T, Hamapumbu H, et al. Changing individual-level risk factors for malaria with declining transmission in southern Zambia: a cross-sectional study. Malar J. 2011; 10:324. [PubMed: 22039751]

36. van Dijk JH, Sutcliffe CG, Munsanje B, Hamangaba F, Thuma PE, Moss WJ. Barriers to the care of HIV-infected children in rural Zambia: a cross-sectional analysis. BMC Infect Dis. 2009; 9:169. [PubMed: 19835604]

37. van Dijk JH, Sutcliffe CG, Munsanje B, et al. HIV-infected children in rural Zambia achieve good immunologic and virologic outcomes two years after initiating antiretroviral therapy. PLoS One. 2011; 6(4):e19006. [PubMed: 21552521]

38. Mercader S, Featherstone D, Bellini WJ. Comparison of available methods to elute serum from dried blood spot samples for measles serology. Journal of virological methods. 2006; 137(1):140–149. [PubMed: 16860401]

39. Tejiokem MC, Gouandjika I, Beniguel L, et al. HIV-infected children living in Central Africa have low persistence of antibodies to vaccines used in the Expanded Program on Immunization. PLoS One. 2007; 2(12):e1260. [PubMed: 18060056]

40. Rainwater-Lovett K, Moss WJ. The urgent need for recommendations on revaccination of HIV-infected children after successful antiretroviral therapy. Clin Infect Dis. 2010; 51(5):634–635. [PubMed: 20684683]

41. Simani OE, Izu A, Violari A, et al. Effect of HIV-1 exposure and antiretroviral treatment strategies in HIV-infected children on immunogenicity of vaccines during infancy. AIDS. 2014; 28(4):531–541. [PubMed: 24468996]

42. Pensieroso S, Cagigi A, Palma P, et al. Timing of HAART defines the integrity of memory B cells and the longevity of humoral responses in HIV-1 vertically-infected children. Proceedings of the National Academy of Sciences of the United States of America. 2009; 106(19):7939–7944. [PubMed: 19416836]

43. Manirakiza A, Kipela JM, Sosler S, Daba RM, Gouandjika-Vasilache I. Seroprevalence of measles and natural rubella antibodies among children in Bangui, Central African Republic. BMC Public Health. 2011; 11:327. [PubMed: 21586151]

44. Cutts FT, Abebe A, Messele T, et al. Sero-epidemiology of rubella in the urban population of Addis Ababa, Ethiopia. Epidemiol Infect. 2000; 124(3):467–479. [PubMed: 10982071]

45. Kombich JJ, Muchai PC, Tukei P, Borus PK. Rubella seroprevalence among primary and pre-primary school pupils at Moi's Bridge location, Uasin Gishu District, Kenya. BMC Public Health. 2009; 9:269. [PubMed: 19640288]

46. Ratnam S, Gadag V, West R, et al. Comparison of commercial enzyme immunoassay kits with plaque reduction neutralization test for detection of measles virus antibody. J Clin Microbiol. 1995; 33(4):811–815. [PubMed: 7790442]

47. Riddell MA, Byrnes GB, Leydon JA, Kelly HA. Dried venous blood samples for the detection and quantification of measles IgG using a commercial enzyme immunoassay. Bull World Health Organ. 2003; 81(10):701–707. [PubMed: 14758429]

48. Novello F, Ridolfi B, Fiore L, et al. Comparison of capillary blood versus venous blood samples in the assessment of immunity to measles. Journal of virological methods. 1996; 61(1–2):73–77. [PubMed: 8882939]

49. Condorelli F, Scalia G, Stivala A, et al. Detection of immunoglobulin G to measles virus, rubella virus, and mumps virus in serum samples and in microquantities of whole blood dried on filter paper. Journal of virological methods. 1994; 49(1):25–36. [PubMed: 7829589]

50. Colson KE, Potter A, Conde-Glez C, et al. Use of a commercial ELISA for the detection of measles-specific immunoglobulin G (IgG) in dried blood spots collected from children living in low-resource settings. J Med Virol. 2015; 87(9):1491–1499. [PubMed: 25988945]

51. Helfand RF, Keyserling HL, Williams I, et al. Comparative detection of measles and rubella IgM and IgG derived from filter paper blood and serum samples. J Med Virol. 2001; 65(4):751–757. [PubMed: 11745941]

A
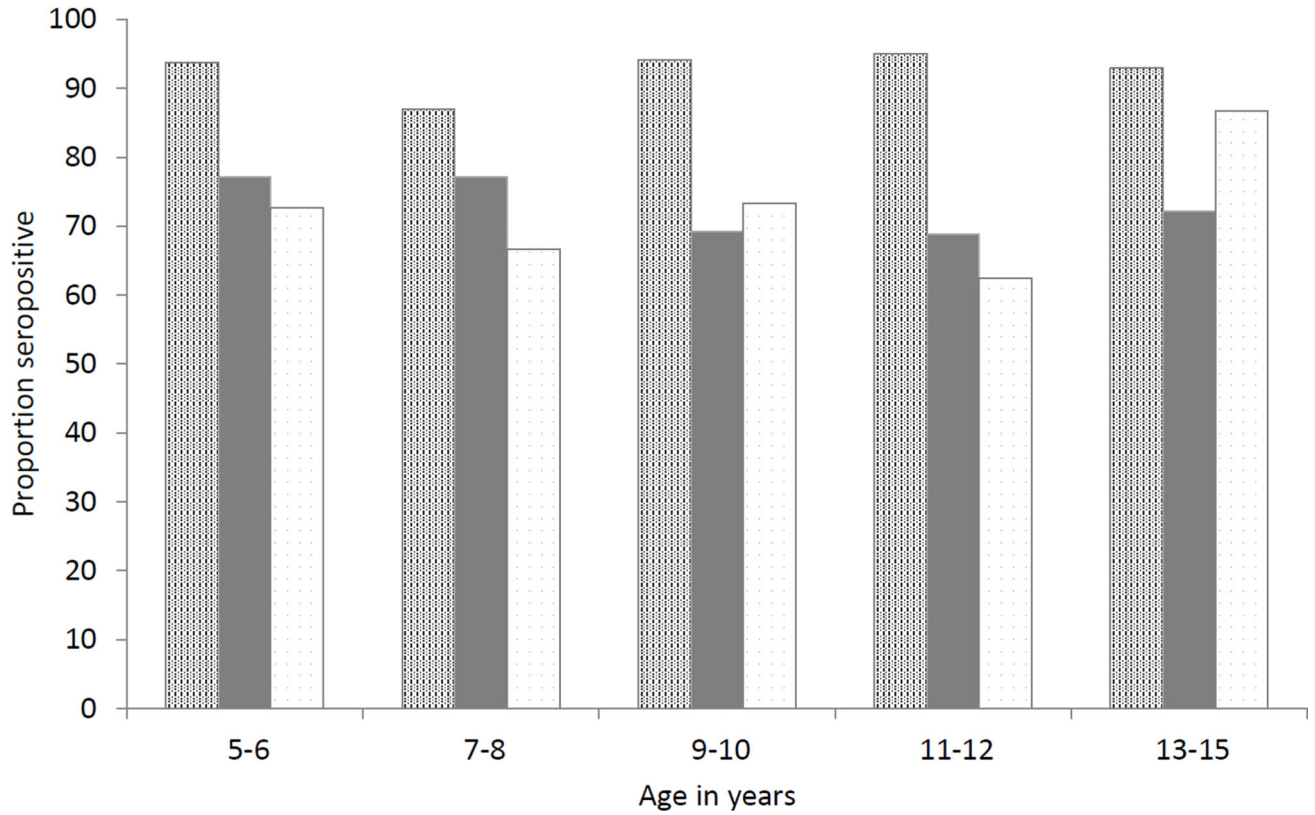
	5-6	7-8	9-10	11-12	13-15
HIV-uninfected	140	106	89	98	128
HIV-infected treatment naïve	48	35	26	16	18
HIV-infected receiving ART	51	30	16	16	15

B

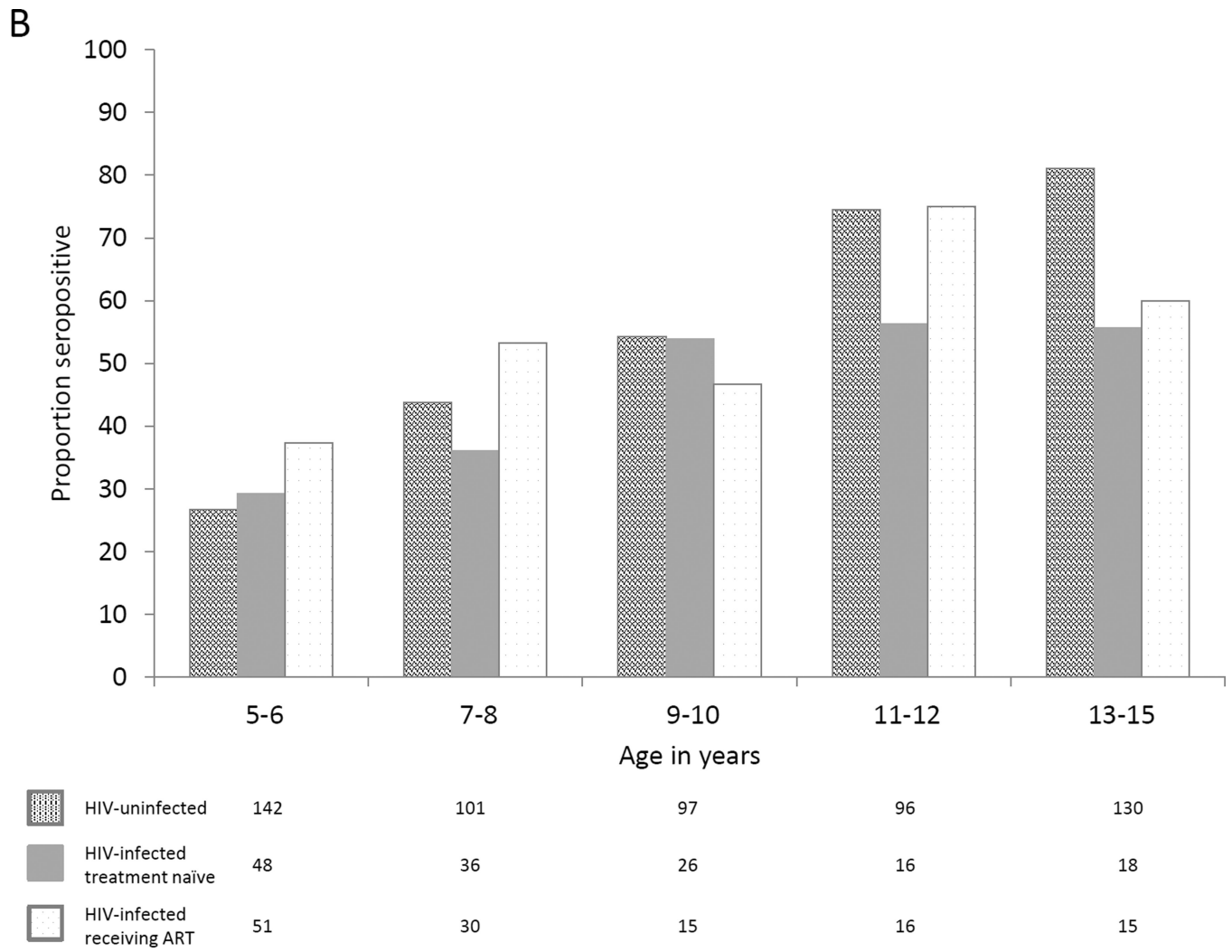|	5-6	7-8	9-10	11-12	13-15
HIV-uninfected	142	101	97	96	130
HIV-infected treatment naïve	48	36	26	16	18
HIV-infected receiving ART	51	30	15	16	15

Figure 1.
Measles (A) and rubella (B) seroprevalence by age

Table 1

Characteristics of youth tested for IgG antibodies to measles and rubella viruses in rural southern Zambia

Characteristics at testing	HIV-uninfected youth	HIV-infected treatment-naïve youth	HIV-infected youth receiving ART
Number	617	144	128
Male: n (%)	288 (47)	65 (45)	68 (53)
Median age in years (IQR)[a]	9.6 (6.9–12.7)	8.3 (6.3–10.7)	7.9 (5.9–10.5)
5–6 years: n (%)	157 (25)	48 (33)	51 (40)
7–8 years: n (%)	118 (19)	36 (25)	30 (23)
9–10 years: n (%)	99 (16)	26 (18)	16 (12.5)
11–12: n (%)	108 (18)	16 (11)	16 (12.5)
13–15: n (%)	135 (22)	18 (13)	15 (12)
Median CD4$^+$ T-cell count (IQR)	---	546 (345–828)	494 (338–818)
Median years on ART (IQR)	---	---	0.96 (0.88–1.2)

ART: antiretroviral therapy; HIV: human immunodeficiency virus; IQR: interquartile range

[a]p<0.0001 for difference between groups

Table 2

Measles and rubella seroprevalence among HIV-infected and uninfected youth in rural southern Zambia

	HIV-uninfected youth	HIV-infected treatment-naïve youth	p-value[a]	HIV-infected youth receiving ART	p-value[b]
MEASLES					
Number	562	143		128	
Positive: >0.2 / Equivocal: 0.1–0.2 / Negative: <0.1					
% positive (95% CI)	92.5 (90.4, 94.8)	74.1 (67.0, 81.3)		71.9 (64.1, 79.7)	
% equivocal (95% CI)	4.3 (2.6, 5.9)	16.1 (10.1, 22.1)	<0.001	15.6 (9.3, 21.9)	<0.001
Median mIU/mL (IQR) among positives	2606 (1301–5069)	1548 (761–3288)	<0.001	1671 (754–3834)	0.01
Median mIU/mL (IQR) among equivocals	231 (199–302)	243 (189–275)	0.99	219 (200–271)	0.67
Positive: 0.1 / Negative: <0.1					
% positive (95% CI)	96.8 (95.4, 98.3)	90.2 (85.3, 95.1)	0.001	87.5 (81.8, 93.2)	<0.001
Median mIU/mL (IQR) among positives	2428 (1126–4890)	1166 (449–2461)	<0.001	1250 (451–3427)	<0.001
RUBELLA					
Number	567	144		127	
Positive: >0.2 / Equivocal: 0.1–0.2 / Negative: <0.1					
% positive (95% CI)	54.7 (50.6, 58.8)	41.7 (33.6, 49.7)		49.6 (40.9, 58.3)	
% equivocal (95% CI)	1.1 (0.2, 1.9)	11.8 (6.5, 17.1)	<0.001	7.9 (3.2, 12.6)	<0.001
Median IU/mL (IQR) among positives	240 (172–318)	99 (51–195)	<0.001	150 (78–207)	<0.001
Median IU/mL (IQR) among equivocals	5 (4–5)	5 (5–5)	0.44	4 (4–6)	0.66
Positive: 0.1 / Negative: <0.1					
% positive (95% CI)	55.7 (51.6, 59.8)	53.5 (45.3, 61.6)	0.63	57.5 (48.9, 66.1)	0.72
Median IU/mL (IQR) among positives	237 (160–318)	64 (8–161)	<0.001	121 (50–197)	<0.001

ART: antiretroviral therapy; CI: confidence interval; HIV: human immunodeficiency virus; IQR: interquartile range; IU/mL: international units per milliliter

[a] p-value comparing HIV-infected treatment-naïve children with HIV-uninfected children

[b] p-value comparing HIV-infected children receiving ART with HIV-uninfected children