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Antenatal clinic HIV data found to underestimate actual prevalence declines: evidence from Zambia

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Summary

OBJECTIVE To determine to what extent antenatal clinic (ANC)-based estimates reflect HIV prevalence trends among men and women in a high prevalence urban population.

METHODS Examination of data from serial population-based HIV surveys in 1995 (n = 2115), 1999 (n = 1962) and 2003 (n = 2692), and ANC-based surveillance in 1994 (n = 450), 1998 (n = 810) and 2002 (n = 786) in the same site in Lusaka, Zambia. The population-based surveys recorded refusal rates between 6% and 10% during the three rounds.

RESULTS Among ANC attendees, prevalence declined by 20% (25.0% to 19.9%; P = 0.101) in the age group 15–24 years and was stable overall. In the general population, the prevalence declined by 49% (P < 0.001) and by 32% (P < 0.001) in age group 15–24 and 15–49, respectively. Among women only, HIV prevalence declined by 44% (22.5% to 12.5%; P < 0.001) and by 27% (29.6% to 21.7%; P < 0.001) in age group 15–24 and 15–49 years, respectively. In addition, prevalence substantially declined in higher educated women aged 15–24 years (20.7% to 8.5%, P < 0.001).

CONCLUSION ANC-based estimates substantially underestimated declines in HIV prevalence in the general population. This seemed to be partially explained by a combination of marked differentials in prevalence change by educational attainment and changes in fertility-related behaviours among young women. These results have important implications for the interpretation of ANC-based HIV estimates and underscore the importance of population-based surveys.

keywords HIV prevalence, antenatal, population survey, trends and Zambia

Introduction

HIV prevalence among pregnant women attending antenatal clinics (ANCs) remains the principal data source of infection trends in sub-Saharan Africa (Kwesigabo *et al.* 2000; Ghys *et al.* 2006). This type of surveillance has been revised to meet changing needs yet maintaining the original objective (WHO/UNAIDS 2003). Despite this usefulness, ANC-based HIV prevalence estimates should be interpreted with caution due to multiple potential inherent selection biases (Mills *et al.* 2005). Methods of adjustment for some of the factors have improved the estimation (Boisson *et al.* 1996; Zaba *et al.* 2000; Fabiani *et al.* 2003, 2006). Despite these drawbacks regarding point estimates, it has often been argued that ANC sentinel surveillance data collected over time will reasonably well capture infection trends of men and women in the general population (Kwesigabo *et al.* 2000). This argument is based on the assumption that the inherent biases remain constant over time, hence will not influence patterns drastically (Mills *et al.* 2005). In the 1990s, very few communities had serial data to check this hypothesis and most validations of ANC-based HIV prevalence used single time-points or relatively short periods of time raising validity and accuracy concerns about the reliability of such extrapolations (Fylkesnes *et al.* 2001; Ghys *et al.* 2006).

In Zambia, the HIV epidemic has been monitored using both ANC-based and population-based data. The ANC-based surveillance system was established with few sites in 1990, and in 1994 a total of 27 sites were selected representing both rural and urban populations in all the provinces (Fylkesnes *et al.* 1998, 2001). Since then, three other rounds of surveillance have been conducted in 1998, 2002 and 2004 (Sandøy *et al.* 2006; Central Board of Health 2005). In one of the surveillance sites in Lusaka urban, serial cross-sectional surveys on HIV prevalence and risk factors were also conducted in 1995, 1999 and 2003 among randomly selected men and women (Fylkesnes *et al.* 2001; Michelo *et al.* 2006). Presently, this is the only site that has both ANC-based and population-based HIV

prevalence estimates from the same population consistently. We investigated how well antenatal-based HIV prevalence estimates capture trends in this general population between 1994 and 2003.

Methods

Antenatal-based surveillance design

The data were collected from serial antenatal-based HIV surveys conducted in Chelstone in 1994 (n = 450), 1998 (n = 810) and 2002 (n = 786). The first epidemiological HIV sentinel surveillance among ANC attendees in Chelstone was conducted in 1990 as a pilot followed by another in 1993. However, the sample sizes in these surveys were very low. In 1994, the core antenatal-based HIV surveillance for the whole country was established, repeated in 1998 and in 2002. The detailed methods and major findings of the earlier national surveys have been reported elsewhere (Fylkesnes *et al.* 1998, 2001; Sandøy *et al.* 2006). Pregnant women who attended the ANC for the first time in the pregnancy were enrolled consecutively. Data were collected in a period not exceeding 4 months. Like in all the other major urban surveillance centres, the target number for Chelstone was 500 in 1994, but this was adjusted to 800 participants thereafter because of higher proportions of antenatal women in these sites compared with other and less urbanized areas. Serum from blood samples drawn for syphilis screening was tested unlinked and anonymously using Capillus HIV-1/HIV-2 rapid test (Cambridge Biotechnology, Galway, Ireland) at the ANCs. Randomly selected negative samples (5% in 1994 and 1998, 10% in 2002) and all positive samples were re-tested at the national laboratory using Wellcozyme HIV Recombinant HIV-1 (Murex, Johannesburg, South Africa). A third test, Bionor HIV-1 & 2 (Bionor As, Skien, Norway), was employed on the samples with discordant results of tests one and two, and this third result was considered final.

Population-based surveillance design

The first population-based HIV survey in Zambia was conducted in 1995 in Chelstone and Kapiri Mposhi and two follow-up surveys were later conducted. The detailed methods and major findings of these population-based studies have been reported elsewhere (Fylkesnes *et al.* 1997, 1998, 2001; Michelo *et al.* 2006). The data used in this study were collected from the surveys conducted in Chelstone in 1995 (n = 2115), 1999 (n = 1986) and 2003 (n = 2589) using random cluster sample design described below. The Zambian census population mapping system was used to establish the sampling frame, which consisted of 24 Standard Enumeration areas (SEAs) with 2786 households. Using 'probability proportional to size', 10 SEAs were selected for this study. In the sampled clusters, a personal structured interview was carried out with all eligible household members aged ≥15 years in order to collect information on education, socio-demographic characteristics and risk behaviours. The second part of the interview involved HIV testing using saliva. In 1995, all saliva samples were tested using Gacelisa HIV 1 & 2 (Welcome Diagnostics, Dartford, Kent, UK) and initially 450 randomly selected samples were tested using Bionor HIV-1 & 2 (Bionor AS) magnetic particle assay following modifications for saliva. Agreement between the two test kits was 99.8% (Fylkesnes & Kasumba 1998). Bionor 1 & 2 was used to test samples in the 1999 and 2003 follow-up surveys.

Validation strategy and statistical analyses

The ANC surveillance was conducted from August to November of 1994, 1998 and 2002. Each population survey was conducted within a year of the antenatal surveillance, that is, from October to December in 1995, October 1998 to May 1999 and from February to May in 2003. For comparability purposes, these years have been denoted as period 1, 2 and 3 to represent 1994, 1998 and 2002 for the ANC-based reports, whereas it is 1995, 1999 and 2003 in the population data, respectively. The health post where the ANC services are provided serves the same catchment area from which the sample for the population survey was drawn. Although in theory women in this area have a choice whether to go to another clinic for antenatal services or not, in practice this does not happen unless one is referred there (and vice versa) for a particular reason. ANC data from the 1990 pilot, 1993 and 2004 surveys were excluded from the analyses due to lack of appropriate population data for validation at the time. Analyses (stratified by age and sex) were performed using INTERCOOLED STATA version 8 (Texas, USA). The Mantel–Haenszel chi-square test (1 degree of freedom and with continuity correction) adjusted for cluster effect was used to test the linear trend of population-based HIV prevalence patterns over the periods. In order to check the effect of age structure, the estimates were standardized using the 2000 census urban female reference population. However, Clayton and Hills' score test for trend was employed for calculating linear trend in the ANC-based data. The median age of sexual debut was calculated using survival analysis and a log rank test for equality of survival curves was used to compare the median ages. To understand the possible differences and or similarities between ANC- and population-based respondents, further analyses were made in terms

of educational attainment, sexual debut and fertility patterns. The response to the question 'have you ever given birth?' was used to approximate fertility. The ANC-based estimates were then compared with the population-based estimates for similarity in all these aspects.

Ethics

The National AIDS Research Committee approved the protocol for the ANC-based surveillance system in 1990 and all HIV testing was done unlinked and anonymously as part of routine standard antenatal care in Zambia (Fylkesnes *et al.* 1998). The testing algorithm complied with the WHO/UNAIDS guidelines for conducting such surveys. The population-based survey protocols received clearance from the National AIDS Research Council and the University of Zambia Research Ethics Committee. The ethical details for both data sources are reported already elsewhere (Michelo *et al.* 2006; Sandøy *et al.* 2006).

Results

In the population-based survey, 60% of the respondents were females. The overall mean age was 26.2 years. The mean age among ANC attendees was 24.1 years. Furthermore, ANC attendees had significantly lower educational levels ($P < 0.001$) than respondents in the population-based survey (mean school years: 7.2 *vs*. 8.6 in period 1, 7.2 *vs*. 9.2 in period 2 and 7.6 *vs*. 10.1 in period 3). In the population-based surveys the overall response rate was 89%. Population-based antenatal attendance (during the last pregnancy) in the three surveys was 97%, 98% and 93% in 1995, 1999 and 2003, respectively.

Age-specific HIV infection trends

Table 1 illustrates both ANC and population-based age-specific HIV prevalence trends. Population-based HIV prevalence of men and women in age group 15–49 years between 1995 and 2003 declined by 32% (26.5%, 23.7% to 18.0%, $P < 0.001$). Among women aged 15–49, prevalence declined by 27% (29.6%, 27.1% to 21.7%, $P < 0.001$). In the same age group, ANC-based prevalence over the period remained stable (24.2%, 25.9% to 24.3%, $P = 0.821$). In age group 15–24 years, the population-based prevalence declined from 16.5%, 13.9% to 8.5% ($P < 0.001$) among men and women, representing a 49% decline and in young women only, prevalence declined by 44% (22.5%, 18.3% to 12.5%, $P < 0.001$). ANC-based data in this age group showed marginal decline of 20% (25.0%, 22.8% to 19.9%, $P = 0.101$). However, after adjusting for fertility risk, the ANC-based estimates matched the estimates among young women aged 15–24 years in the general population. In this age group, fertility-adjusted prevalence declined by 42% (25.1%, 19.1% to 14.6%, $P = 0.001$) resembling the 44% decline observed in the general population of young women of the same age.

In the general population, prevalence declined largely in age group 15–29 years, whereas among ANC attendees, prevalence significantly declined only in the group aged 15–19 years. Furthermore, although ANC-based age-specific point estimates matched population-based estimates of young women aged 15–24 in time 1 (25% *vs*. 22.5%) and time 2 (22.8% *vs*. 18.3%), the ANC-based point estimate was significantly higher than population-based estimate in time 3 (19.9%, 95% CI 16.2–23.6.8 *vs*.12.5%, 95% CI 9.2–15.8). When considering men and women together, ANC-based estimates still significantly over-estimated population prevalence in the groups aged 15–24 years throughout the survey rounds; time 1 (25%, 95% CI 19.8–30.2 *vs*. 16.5%, 95% CI 13.3–19.8), time 2 (22.8%, 95% CI 18.9–26.6 *vs*. 13.9%, 95% CI 12.3–15.5) and time 3 (19.9%, 95% CI 16.2–23.6 *vs*. 8.5%, 95% CI 6.5–10.6).

HIV and educational attainment

In general, population-based estimates showed a universal shift towards declining prevalence of HIV infection in groups with higher education over the period, as illustrated in Table 2. Among higher educated young men and women aged 15–24 years, prevalence declined from 16.1%, 11.7% to 5.3% ($P < 0.001$). In the same age group, prevalence declined from 20.7%, 16.1% to 8.5% ($P < 0.001$) among women only with higher education. Similarly, in higher educated groups of men and women aged 25–49 years, prevalence declined from 43.2% to 26.4% ($P < 0.001$) and from 45.6% to 29.0% ($P < 0.001$) in women only. Overall (men and women), prevalence declined from 32.0% to 14.8% ($P < 0.001$) and from 34.1% to 17.5% ($P < 0.001$) in women only. In sharp contrast, among ANC attendees, prevalence remained stable over the study in all age groups and irrespective of educational attainment. The overall level of educational attainment among ANC attendees did not significantly change over the periods (mean school years: 7.2, 7.2 and 7.6, $P = 0.219$; median school years 7.0 in all periods) despite an increase in the proportion of attendees with ≥11 school years from 10%, 13% to 21% in period 1, 2 and 3, respectively. Among young women aged 15–24 years, the pooled proportion of pregnant women with up to 7 years of school remained >60%, whereas those with ≥11 school years was <22% in all the surveys. The distribution was the same even in age group 15–19 years.

Table 1 Age-specific adjusted prevalence differential trends of HIV infection by sex in Chelstone, Lusaka: 1995–2003

		Age-specific prevalence [% (95% CI*), *n*]						
Survey type	Time	15–19	20–24	25–29	30–39	40–49	15–24	15–49
ANC sentinel surveillance	1994	21.7% (13.7–29.7) *n* = 106	27.2% (20.2–34.1) *n* = 162	27.8% (18.9–37.2) *n* = 90	20.0% (11.3–28.7) *n* = 85	*n* = 7	25.0% (19.8–30.2) *n* = 268	24.2% (20.2–28.2) *n* = 450
	1998	15.1% (10.1 20.1) *n* = 199	28.4% (23.0–33.7) *n* = 275	31.9% (24.8–39.1) *n* = 166	29.4% (21.6–37.2) *n* = 136	*n* = 10	22.8% (18.9–26.6) *n* = 474	25.9% (22.9–28.0) *n* = 786
	2002	9.2% (4.5–13.8) *n* = 153	25.5% (20.5–30.5) *n* = 294	28.7% (22.2–35.3) *n* = 188	33.1% (25.4–40.9) *n* = 145	*n* = 6	19.9% (16.2–23.6) *n* = 447	24.3% (21.3–27.3) *n* = 786
	Trend, *P*-value: 1994–2002	**0.008**	0.611	0.972	**0.042**	–	0.101	0.913
Population-based surveillance (females)	1995	12.3% (7.4–17.1) *n* = 391	35.4% (26.7–43.9) *n* = 311	48.7% (42.6–54.9) *n* = 199	34.5% (28.0–41.0) *n* = 310	26.0% (13.8–38.2) *n* = 100	22.5% (19.2–25.8) *n* = 702	29.6% (26.8–32.4) *n* = 1311
	1999	9.5% (5.5–13.6) *n* = 336	27.9% (24.3–31.4) *n* = 305	41.2% (35.6–46.8) *n* = 221	39.3% (34.9–43.8) *n* = 244	22.3% (15.9–28.7) *n* = 112	18.3% (16.3 20.2) *n* = 641	27.1% (25.2–28.8) *n* = 1218
	2003	7.7% (4.7–10.6) *n* = 431	17.6% (13.3–21.9) *n* = 409	31.1% (26.1–36.1) *n* = 286	39.6% (33.9–45.2) *n* = 273	22.9% (17.0–28.9) *n* = 148	12.5% (9.2–15.8) *n* = 840	21.7% (19.4–24.1) *n* = 1547
	Trend, *P*-value: 1995–2003	**0.026**	**<0.001**	**<0.001**	0.202	0.614	**<0.001**	**<0.001**
Population-based surveillance (males and females)	1995	9.1% (5.3–12.9) *n* = 657	26.7% (19.6–33.8) *n* = 479	43.1% (37.9–48.4) *n* = 313	36.3% (31.1–41.5) *n* = 460	33.9% (25.8–42.2) *n* = 206	16.5% (13.3 19.8) *n* = 1136	26.5% (23.9–28.9) *n* = 2115
	1999	8.2% (5.2–11.2) *n* = 560	20.1% (17.6–23.6) *n* = 513	34.8% (29.7–40.2) *n* = 330	39.3% (34.9–43.6) *n* = 365	29.9% (24.7–35.1) *n* = 194	13.9% (12.3–15.5) *n* = 1073	23.7% (22.3–25.2) *n* = 1961
	2003	5.4% (3.4–7.3) *n* = 745	11.8% (9.0–14.6) *n* = 718	24.7% (21.1–28.2) *n* = 446	37.9% (31.9–43.8) *n* = 454	26.5% (17.2–35.9) *n* = 226	8.5% (6.5–10.6) *n* = 1463	18.0% (15.3 20.8) *n* = 2589
	Trend, *P*-value: 1995–2003	**0.007**	**<0.001**	**<0.001**	0.626	0.091	**<0.001**	**<0.001**

Notes: 1, *Confidence Intervals adjusted for clustering effect in the population-based estimates; 2, chi-square for linear trend (all significant *P*-values are highlighted in bold); 3, sample sizes were (a) ANC: 450, 786 and 786 in 1994, 1998 and 2002, respectively (b) Population-based: 2115, 1961 and 2589 in 1995, 1999 and 2003, respectively.

Table 2 Comparing trends of HIV prevalence by years of schooling between antenatal clinic (ANC) attendees and the general population in (1995–2003) Chelstone

Age	Population	Education (years)	Prevalence, % (*n*)			§ MH Chi-square for linear trend
			Time 1	Time 2	Time 3	Time 1 to 3
15–24	ANC attendees	0–7	27.2% (53/195)	22.6% (56/248)	20.8% (49/236)	0.118
		8–10	19.4% (7/36)	21.0% (26/124)	19.2% (23/120)	0.840
		≥11	20.0% (4/20)	23.7% (14/59)	18.5% (15/81)	0.634
	PBS (F)	0–7	21.5% (59/274)	18.4% (32/174)	20.2% (35/173)	0.664
		8–10	23.8% (66/277)	20.0% (50/250)	13.8% (33/240)	**0.000**
		≥11	20.7% (30/145)	16.1% (35/217)	8.5% (36/423)	**0.000***
	PBS (M + F)	0–7	16.8% (64/380)	17.5% (41/234)	15.9% (39/244)	0.495
		8–10	16.2% (77/476)	14.0% (59/421)	10.0% (44/440)	0.011
		≥11	16.1% (44/273)	11.7% (49/418)	5.3% (41/774)	**0.001***
25–49	ANC attendees	0–7	26.2% (28/107)	34.9% (51/146)	29.8% (45/151)	0.646
		8–10	14.3% (3/21)	27.4% (20/73)	32.2% (19/59)	0.166
		≥11	21.1% (4/19)	27.3% (9/33)	33.8% (25/74)	0.264
	PBS (F)	0–7	28.5% (63/221)	26.7% (47/176)	34.6% (55/159)	0.288
		8–10	41.2% (86/209)	42.4% (86/203)	36.5% (78/214)	0.389
		≥11	45.6% (77/169)	39.9% (79/198)	29.0% (96/331)	**0.001***
	PBS (M + F)	0–7	30.1% (84/279)	26.5% (56/211)	36.1% (74/205)	0.278
		8–10	38.8% (116/299)	38.2% (100/262)	34.9% (98/281)	0.213
		≥11	43.2% (168/389)	38.6% (160/415)	26.4% (168/637)	**0.000***
15–49	ANC attendees	0–7	26.8% (84/314)	27.2% (116/426)	24.3% (101/415)	0.423
		8–10	17.2% (10/58)	22.5% (46/204)	22.9% (44/192)	0.469
		≥11	22.0% (9/41)	26.6% (25/94)	26.1% (43/165)	0.707
	PBS (F)	0–7	24.7% (122/495)	22.6% (79/350)	27.1% (90/332)	0.657
		8–10	31.3% (152/486)	30.0% (136/453)	24.5% (111/454)	**0.009**
		≥11	34.1% (107/314)	27.5% (114/415)	17.5% (132/754)	**0.000***
	PBS (M + F)	0–7	22.5% (148/659)	21.8% (97/445)	25.2% (113/449)	0.501
		8–10	24.9% (193/775)	23.3% (159/683)	19.7% (142/721)	**0.033**
		≥11	32.0% (212/662)	25.1% (209/833)	14.8% (209/1411)	**0.000***

Notes: 1. Years: ANC, 1 = 1994, 2 = 1998, 3 = 2002; PBS: 1 = 1995, 2 = 1999, 3 = 2003. 2. Sample sizes: PBS: *n* = 2115 in 1995; *n* = 1961 in 1999; *n* = 2589 in 2003; ANC: *n* = 450 in 1994; *n* = 786 in 1998; *n* = 786 in 2002. 3. Chi-square test for trend (a) § MH denotes Mantel–Haenszel chi square for linear trend adjusted for cluster effect (1 degree of freedom and with continuity correction), in the population-based estimates. However, Clayton and Hills' score test for trend was used in the ANC data (b) statistically significant *P*-values are highlighted in bold (c) *signifies $P < 0.001$.

Population-based sexual debut and ever given birth by educational attainment

In the general population of women aged 15–24 years, the mean number of years in school increased from 8.64 (95% CI 8.64–8.83) in 1995 and 9.21 (95% CI 9.04–9.38) in 1999 to 10.1 (95% CI 9.95–10.26) in 2003. In this group, older age at first sex was associated with higher educational attainment. In 2003, the median age for sexual debut among women aged 20–29 years and with >11 years of school was 19 years whereas it was 16.6 years in groups with 0–7 years of school ($P < 0.001$). Similarly, the proportion of young people aged 15–24 who said they ever had sex dropped between 1999 and 2003 (males: Adjusted Odds Ratio (AOR) 0.83 95% CI 0.62–1.10; females: AOR 0.68 95% CI 0.52–0.88). Following a similar pattern, higher educated young people showed significant postponement in ages at first birth. Consequently, among women with >11 school years, the proportion ever given birth in age group 15–24 years decreased from 33.1%, 22.2% to 19.2% ($P = 0.002$) whereas the decline was marginal in groups with 0–7 school years (Figure 1). In age group 15–19, the proportion of women ever given birth declined by 57% (16.8%, 9.4% to 7.9%; OR 0.43, 95% CI 0.26–0.68) and from 65.5%, 47.5% to 41.7% ($P < 0.000$) in age group 20–24 years (see Table 3). Figure 2 illustrates further the general decline in age at first birth among women <30 years of age in the general population.

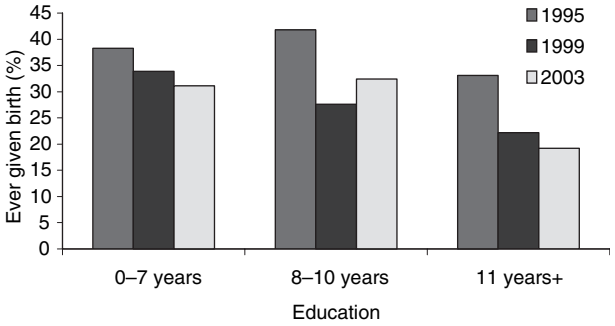
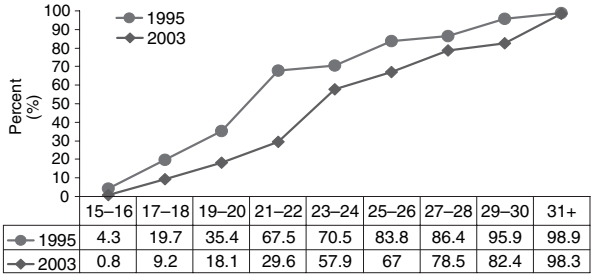


Figure 1 Proportion (per cent) of women 'ever given birth' by educational level in aged 15–24 years in Chelstone (urban), Zambia. *Notes*: 1, Sample sizes n = 886, 853 and 1018 in 1995, 1999 and 2003, respectively; 2, P-value for MH chi square for trend: 0–7 school years, P = 0.405, 8–10 school years, P = 0.289 and for ≥11 school years, P = 0.0001.



	15–16	17–18	19–20	21–22	23–24	25–26	27–28	29–30	31+
1995	4.3	19.7	35.4	67.5	70.5	83.8	86.4	95.9	98.9
2003	0.8	9.2	18.1	29.6	57.9	67	78.5	82.4	98.3

Figure 2 Increase in age at first birth (1995–2003) among HIV negative women ever given birth in Chelstone (Lusaka urban), Zambia. *Notes*: 1, Sample sizes in group aged 15–30 years, n = 1511, 1462 and 1994 in 1995, 1999 and 2003, respectively (overall all ages, n = 6665); 2, P-value for the increase in age at first birth was significant ($P < 0.05$) in all age groups <31 years of age.

Discussion

We found diminishing representativeness of ANC-based data in capturing HIV prevalence and trends in the general population. First, the overall ANC-based HIV prevalence estimates remained stable, contrasting the population-based pattern where prevalence declined in both women and men. Second, both sources of data showed HIV declines in age group 15–24 years, but declines were substantially steeper in population-based data (which showed a decline of 49%) compared with ANC-based data (which showed a decline of 20%). These findings seem to be partially explained by a combination of marked differentials in prevalence change by educational attainment as well as marked changes in fertility-related behaviours such as the postponement of first birth among young women. This postponement was associated with educational attainment. We have already reported substantial HIV declines among higher educated groups in this population (Michelo *et al.* 2006). The findings are a challenge to the interpretation of results from HIV surveillance systems, and whether the results are generalizable to other areas in Zambia and countries in sub-Saharan Africa merits further study. A similar pattern of change in ANC-based HIV prevalence reported here has been observed in the majority of surveillance sites in Zambia, and the most likely scenario is that similar change mechanisms were seen in many of these populations (Sandøy *et al.* 2006). Historically, the only and dramatic parallel declines and not associated with postponement of birth, observed from both antenatal- and population-based data was in Kagera region during the 1990s (Asiimwe-Okiror *et al.* 1997; Kwesigabo *et al.* 1998, 2000, 2005). The declines in Kagera may have been a signal of continuing declines in the general population as the epidemic matured. At such a stage of the epidemic as currently observed, dynamics among antenatal attendees differ from the general population; hence, using ANC-based estimates alone could even lead to a possibility of serious

Table 3 Population-based declining trend in the proportion of women ever given birth in Chelstone (urban), Zambia: 1995–2003

	Proportion of women ever given birth in respective periods			
Age	1995*, % (n)	1999, % (n)	2003, % (n); OR† 95% CI	P-value (trend‡)
15–19	16.8 (382)	9.4 (330)	7.9 (355); 0.43 95% CI 0.26–0.68	<0.001
20–24	65.5 (310)	47.5 (299)	41.7 (384); 0.38 95% CI 0.28–0.51	<0.001
25–29	85.4 (198)	83.9 (218)	74.8 (282); 0.51 95% CI 0.32–0.82	0.003
30–39	95.2 (310)	95.0 (242)	94.5 (273); 0.38 95% CI 0.28–0.51	0.722

*Sample sizes: n = 1100, 1089 and 1294 in 1995, 1999 and 2003 respectively.
†OR denotes odds ratio for the proportions in 2003 using the proportions in 1995 as a reference category and the 95% CI have adjusted for cluster effect.
‡Denotes Mantel–Haenszel chi square for linear trend adjusted for cluster effect (1 degree of freedom and with continuity correction).

underestimation of what could be intervention-linked impacts.

The basis for our assessment of how well ANC-based data capture HIV trends in the general population was data from a population-based survey conducted in the area covered by the ANC. There is a potential of non-response to have biased the results. Among ANC attendees, non-response can be estimated largely by ANC coverage, and coverage had been stable and above 90% in all the surveys (estimated to be 98% in Lusaka) (Central Statistical Office (Zambia) *et al.* 2003). In addition, movements of nearby populations of women to and from the ANC catchment area were the other possible sources of bias. However, the influence exerted by nearby populations on ANC prevalence estimates in our study was minimal as movement between ANCs to and from this catchment area is not by choice but dictated by a need-driven functioning referral system. One such need is the availability of Prevention of Mother to Child Transmission (PMTCT) services. In Chelstone, PMTCT services were only firmly grounded in 2004 and are unlikely to have been a factor in the stability of the ANC-based prevalence over the period in question. In the population-based study, the most significant cause of non-participation was absence of men. Refusal to give saliva for HIV testing was low in all the three surveys. In view of this it does not seem likely that non-response bias was an important factor affecting the results. In addition, fertility-linked factors which could be the other possible sources of selection bias were critical explanatory factors in this study. Furthermore, we also exclude laboratory-associated differentials in test result to have been a factor (Fylkesnes & Kasumba 1998; Fylkesnes *et al.* 1998).

There is convincing evidence that HIV transmission among adults in sub-Saharan Africa is predominantly heterosexual (Schmid *et al.* 2004). Women tend to under-report socially undesirable sexual behaviours, such as young age at sexual debut (Upchurch *et al.* 2002; Central Statistical Office (Zambia) *et al.* 2003). However, elsewhere we have argued that 'it is reasonable to believe that asking a woman whether she has a ever given birth will give more reliable answers than whether she has ever had sex, as childbearing is associated with high respect in this society and is difficult to keep secret' (Sandøy *et al.* 2007). Therefore, the reduction in the proportion who had ever given birth and parallel postponement of age at first birth among higher educated young people in the general population suggests a convincing behaviour change. Postponement of childbearing might be due to a combination of abstinence, consistent use of condoms as well as utilization of other contraceptives, all of which have increased during the period (Central Statistical Office (Zambia) *et al.* 2003). These factors were of critical importance in explaining the reduced odds of HIV infection among higher educated groups in the general population of young females (Fylkesnes *et al.* 1998; Michelo *et al.* 2006).

However, the picture is dissimilar among ANC attendees. First, the overestimation of HIV prevalence seen in ANC data from age group 15–19 years might be related to the fact that this group engaged in unprotected sex, thereby increasing the likelihood of infection compared with their counterparts in the general population, some of whom are not sexually active (Konde-Lule *et al.* 1997; Fylkesnes *et al.* 1998; Gray *et al.* 1998; Pettifor *et al.* 2004). Second, the higher prevalence in age group 20–24 years also suggests that there is a steady increase in the number of newly infected women arising from younger age groups. Since male HIV prevalence in parallel ages in the general population is generally low, there was evidence suggesting that these young women most probably were involved in cross-generation relationships and had unprotected sex with infected older men (Gregson *et al.* 2002). This could be the major reason why young antenatal women have a higher likelihood of HIV infection (Strickler *et al.* 1995; Fylkesnes *et al.* 1998; Fabiani *et al.* 2003; Mills *et al.* 2005). We also noted that the observed parallel HIV declines associated with education in the general population was absent among ANC attendees. Higher educated young women in the general population were postponing pregnancy and the postponement was substantial in a relatively short period of time. This is likely to be due to a combination of an ongoing process of fertility change which is probably linked with the success of other preventive strategies against HIV transmission such as condom use, abstinence and a longer and more careful search of 'safer' marital partners. In contrast, young ANC women may have ignored messages about abstinence and protection against HIV infection, which was probably influenced by biological and social pressures including the mere desire for motherhood. This differentiating feature between ANC attendees and young women in the general population merits further study and monitoring, especially since the population distribution continues to be highly dynamic.

Therefore, the interpretation of ANC-based HIV prevalence estimates and their extrapolation to the general population remain difficult tasks (Strickler *et al.* 1995; Mills *et al.* 2005). This is largely because in general, ANC-generated prevalence is also vulnerable to several time-dependent sources of bias such as usage and coverage of ANC services, migration, deaths, convenience sampling of sites, population distribution by age, differential distribution by social classes, contraceptive use and

fertility differences with HIV negative women including behavioural differences. This limits generalizability (Mills *et al.* 2005). Adjusting for fertility risk using data from a reference general population within the region improves estimates from ANC attendees (Fabiani *et al.* 2003). In our study, we achieved similar findings by adjusting for fertility risk among ANC attendees using the same source general population as a reference population.

In summary, our results have some important implications for the interpretation of ANC-based HIV estimates. They underscore the importance of population-based surveys, and particularly surveys conducted in selected communities in order to improve the interpretation of HIV trends captured by ANC attendees. These surveys should measure biological, behavioural and socio-demographic information concomitantly to generate critical knowledge for improving our understanding of dynamics of population responses. This also calls for adjustments in the current ANC surveillance systems to allow for sampling adjustments, collection of additional important details such as testing and treatment history as well as indicators for behavioural patterns, in order as to assess the extent of inherent biases which are in turn useful in interpreting ANC-based HIV trends.

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