

Loss to follow-up among female sex workers in Zambia: findings from a five-year HIV-incidence cohort

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1	Loss to follow-up among female sex workers in Zambia: findings from a 5-year HIV
2	incidence cohort
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1 ABSTRACT

HIV incidence studies are used to identify at-risk populations for HIV prevention trials and interventions, but loss to follow-up (LTFU) can bias results if participants who remain differ from those who drop out. We investigated the incidence and factors associated with LTFU among Zambian female sex workers (FSWs) in an HIV incidence cohort from 2012 to 2017. Enrolled participants returned at month 1, month 3 and quarterly thereafter. FSWs were considered LTFU if they missed six consecutive months, or if their last visit was six months before the study end date. Of 420 FSWs, 139 (33%) were LTFU at a rate of 15.7 per 100 person-years. In multivariable analysis, LTFU was greater for FSWs who: never used alcohol, began sex work above the age of consent, and had a lower volume of new clients. Our study appeared to retain FSWs in most need of HIV prevention services offered at follow-up. Keywords: Incidence studies; Risk behaviour; Prevention trials; Alcohol INTRODUCTION

With a prevalence of 11.5%, Zambia is a country with a generalised HIV epidemic 1 (UNAIDS, 2018). Female sex workers (FSWs) are at an even greater risk of acquiring 2 HIV, with a prevalence ranging from 46% to 73% in Zambia (Family Health 3 International & Tropical Disease Research Centre, 2015). High numbers of sexual 4 partners, inconsistent condom use, limited access to health care, stigma, and poverty 5 put Zambian FSWs at elevated risk of HIV infection (Family Health International & 6 Tropical Disease Research Centre, 2015; Scorgie et al., 2012; USAID, 2013; Prüss-7 8 Ustün et al., 2013).

9 Prospective cohort studies are the benchmark for measuring HIV incidence as they

allow chronological measurement of risk factors and HIV infection (Mann, 2003).

11 Studies of this nature require special attention because the drop out or loss to follow-

12 up (LTFU) of participants may introduce selection bias. HIV incidence rates

13 calculated from cohort studies with large rates of LTFU may thus be imprecise.

14 Understanding the characteristics of participants who drop out of studies is crucial for

15 generalisability of results. For FSWs in HIV incidence cohorts, measuring factors

associated with LTFU could help determine if FSWs who drop out are more

vulnerable to HIV than those who remain. High risk cohorts are needed for HIV

18 prevention studies, whose participants must be willing to be followed over time

19 (Fishbein & Coutinho, 1997; Mills et al., 2006).

20 Many studies on LTFU among FSWs focus on HIV-positive sex workers enrolled in

21 antiretroviral treatment programmes (Eakle et al., 2017; Mountain et al., 2014). There

is limited data on the incidence and predictors of LTFU among HIV-negative FSWs.

23 Previous longitudinal studies among HIV-negative FSWs in Kenya and China found

that FSWs who display increased HIV risk behaviour were less likely to be lost to

follow-up (Graham et al., 2013; Su et al., 2016). To our knowledge, there have been

no studies published on LTFU among Zambian FSWs. Measuring the incidence of

27 LTFU for Zambian FSWs and its associated factors could have important implications

for retaining this hard-to-reach population. Retaining FSWs in sex worker-friendly

29 services is important because routine health services typically marginalise FSWs

30 (Lafort et al., 2017). Using data from a five-year HIV incidence cohort of FSWs at the

31 Zambia-Emory HIV Research Project (ZEHRP), we analysed the incidence and

32 factors associated with LTFU.

33 METHODS

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1 Study Setting

- 2 The study took place in Zambia's capital city of Lusaka and the Copperbelt Province
- 3 city of Ndola, which are the two most populated and urbanised provinces in the
- 4 country. The HIV prevalences in Lusaka (15.7%) and the Copperbelt (13.8%) are well
- 5 above the national average (11.5%) (ZAMPHIA Collaborating Institutions, 2019).
- 6 FSWs in Zambia face difficulties accessing HIV testing services due to stigmatisation
- 7 from healthcare workers (Chanda, Perez-Brumer, et al., 2017). Recent efforts to
- 8 provide HIV testing services to FSWs in Zambia have employed self-tests that are yet
- 9 to be rolled out nationally (Chanda, Ortblad, et al., 2017).

10 Study Design

This analysis was part of a prospective cohort study to determine HIV incidence 11 among Zambian FSWs. Recruitment began in September 2012 and ended in March 12 2015. The end of the study follow up was the 15th of September 2017. Follow-up 13 lasted from the date of enrolment until LTFU or study end- whichever came first. The 14 maximum study duration for each participant was 60 months (five years). FSWs who 15 16 attended at least the baseline visit and completed all enrolment procedures were eligible for analysis. Individuals who were duplicate-enrolled in this study were 17 excluded from our analysis (Figure 1). 18

19 Study Population

- 20 In the prospective HIV incidence cohort, we defined sex workers as women who offer
- sex in exchange for money. Enrolment criteria included being HIV-negative,
- 22 unmarried, between the ages of 18 and 45, available for five-year follow-up and
- 23 willing to provide locator information.

24 Study Procedures

Community health workers and peer sex workers at ZEHRP recruited FSWs from
commercial sex work hotspots in Lusaka and Ndola, i.e. bars, restaurants, boarding
houses, lodges/hotels, and streets. Participants provided informed consent in
English, Nyanja or Bemba. At enrolment, FSWs completed a baseline questionnaire
administered face-to-face by nurses trained in risk reduction counselling. Information
was collected on sociodemographics, sexual history, risk factors for HIV acquisition,
and gynaecological and reproductive health status. Enrolled participants were invited

to attend a follow-up visit one month after enrolment, two months later, and quarterly
thereafter. Manual and electronic fingerprinting were used to verify the identity of
study participants at each visit as described in detail elsewhere (Wall et al., 2015). All
participants were reimbursed for travel to and from the study site, and were provided
food and beverages during study visits.

6 Ethical Approval

The University of Zambia Research Ethics Committee (Lusaka, Zambia) and the
Emory University Institutional Review Board (Atlanta, USA) approved the study
protocol.

10 Sexual and Reproductive Health Services

At each study visit, FSWs received HIV risk reduction and family planning 11 12 counselling, and testing for HIV (rapid antibody tests for screening and antigen test for confirmation of positive test results). Women who tested HIV-positive were 13 14 referred for assessment and treatment to a government antiretroviral therapy clinic in accordance with Zambian HIV testing guidelines. FSWs were also screened for 15 syphilis using rapid plasma reagin serology and for trichomonas vaginalis using 16 microscopy of vaginal swabs. Syphilis and trichomonas vaginalis were treated at the 17 study site. Participants were offered condoms and the choice of two long-acting 18 reversible contraceptives: an intrauterine device or a hormonal implant. 19

20 Outcome

The event of interest for the main study was HIV-incidence, but the event of interest for this analysis was LTFU. Participants were considered LTFU if they were absent for more than six months or their last study visit occurred greater than six months before the study end date. This six-month cut-off is, according to evidence from a multinational study, the least likely to lead to misclassification bias in defining LTFU (Chi et al., 2011).

27 Independent Variables

28 Potential factors associated with LTFU were a combination of time-fixed and time-

varying covariates. Time-fixed covariates comprised variables collected at baseline

- 30 and included year of entry into the study, city of residence, age at enrolment, age at
- 31 initiation into sex work, level of education, number of children, lifetime number of Page **5** of **22**

- 1 sexual partners and alcohol use. For our analysis, we combined the years 2012 and
- 2 2013 because only six FSWs (1.5% of the study sample) were enrolled in 2012.
- 3 Time-dependent variables considered for this analysis were syphilis and
- 4 trichomoniasis test results, number of non-repeat/new clients seen in the previous
- 5 month and consistent (100%)/inconsistent (less than 100%) condom use with clients
- 6 during vaginal, anal and oral sex in the previous month.

7 Statistical Analysis

8 LTFU rate

- 9 The time each participant spent in the study between enrolment and final visit was
- calculated in person-years (PY). The Kaplan-Meier method was used to estimate the
- 11 rate of LTFU in our cohort. The incidence rate of LTFU was calculated as the number
- of LTFU cases divided by the total number of PY. Time-fixed covariates were used to
- 13 generate stratified Kaplan-Meier failure curves. The log-rank test for equality of
- 14 survival functions was computed to determine statistically significant (p<0.05)
- 15 differences between curves.

16 Cox model

- 17 A univariate extended Cox model with time-varying covariates was used to estimate
- the hazard ratios (HR) and 95% confidence intervals (CI) of potential factors
- associated with LTFU. Variables found to be significantly associated with LTFU
- 20 (p<0.25) in univariate analysis were tested in multivariable analysis through a forward
- selection procedure (Bursac et al., 2008). Adjusted hazard ratios (AHR) were
- computed for variables with a p-value <0.05.

23 Sensitivity analysis

- The majority (85%) of LTFU in our study occurred during the first three years (Figure
- 25 2). We therefore performed a sensitivity analysis with a right truncated cohort (cut-off
- point= visit month 36) to determine if our results would differ between the full and
- 27 truncated model.
- All statistical analyses were performed using Stata version 14.2 (StataCorp, College
 Station, Texas, USA).

30 **RESULTS**

(Figure 1). Almost half of all recruitments (46.7%) took place in the year 2014 (Table 2 1). The majority of study participants (64.8%) were from Ndola and the rest (35.2%) 3 were Lusaka based (Table 1). The mean age at enrolment was 25 years with a 4 standard deviation (SD) of 6.3 years. Most FSWs were educated up to primary level 5 (56.9%) and had an average of 1.6 children (SD: 1.3, range: 0-7). The majority of 6 participants (65.5%) at baseline reported inconsistent condom use with their clients 7 8 (Table 1). 9 As shown in Table 1, 281 (67%) participants completed the study and had a median

In total, 420 of FSWs enrolled between 2012 and 2015 were eligible for this study

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follow-up time of 33.7 months (IQR: 30.6-42.9). For the 139 (33%) FSWs who were
LTFU, the median follow-up time was 27.6 months (IQR: 15.5-36.5). The overall
person-years of observation contributed by all 420 participants was 883.5 (Table 1).
During this period, LTFU occurred at a rate of 15.7 per 100 PY (95% CI: 13.3218.58). The Kaplan-Meier failure curve in Figure 2 illustrates the rate of LTFU across
all five years of the study.

There was not much variation in the LTFU rate between FSWs by age, number of
children and relationship status (Table 1). However, FSWs who had the fewest
number of lifetime sexual partners had a higher LTFU rate than those who reported
more lifetime partners (p<0.05) (Table 1). FSWs who lived in Lusaka had a higher
LTFU rate than FSWs who lived in Ndola (p<0.05). We also found that FSWs
enrolled later in the study had higher LTFU rates than those enrolled earlier (p<0.05)
(Table1).

In multivariate analysis (Table 2), LTFU was significantly associated with later year of 23 enrolment, no alcohol use, older age at initiation into sex work, and lower monthly 24 volume of new clients. In comparison to FSWs enrolled in 2012/2013, FSWs enrolled 25 in 2014 (AHR: 2.24, 95% CI: 1.12-4.47) and 2015 (AHR: 2.97, 95% CI: 1.44-6.14) 26 had a higher likelihood of being LTFU. FSWs who reported never using alcohol were 27 45% more likely to be LTFU than FSWs who reported ever using alcohol (AHR: 1.45, 28 95% CI: 1.04-2.12). FSWs who began sex work at 16 and above were two-times 29 likelier to be LTFU (AHR: 2.02, 95% CI: 1.25-3.28) than FSWs who began sex work 30 at 15 and below. FSWs who received 1-4 (AHR: 0.61, 95% CI: 0.39-0.95), 5 to 9 31 (AHR: 0.51, 95% CI: 0.29-0.88) or >10 (AHR: 0.46, 95% CI: 0.26-0.82) new clients in 32

33 the previous month were less likely to be LTFU than FSWs who received no new Page 7 of 22 1 clients in the preceding month. The results of our sensitivity analysis mirrored those

2 of our original model, thus we maintained all study visits for greater statistical power.

3 DISCUSSION

Over five years, a third (33%) of our study participants were LTFU at a rate of 15.7 4 per 100 PY. This is lower than the LTFU rates observed in HIV-negative FSWs 5 cohorts in Kenya and China, which were 23.4 to 26.7 per 100 PY respectively 6 7 (Graham et al., 2013; Su et al., 2016). Despite our relatively low LTFU rate, it is vital 8 that retention efforts be improved because participants who drop out may suffer poor health outcomes. Retention in our cohort was associated with earlier year of 9 enrolment and increased HIV risk factors (alcohol use, younger age at entry into sex 10 work, and more monthly number of new clients). 11

12 Our study appeared to retain FSWs at higher HIV risk. Retained women may have been motivated by their need for HIV prevention services offered routinely at follow-13 14 up. It is possible that participants who dropped out of the cohort no longer practised sex work, thus the services offered by the study were less pertinent to them. Previous 15 16 studies in Africa show that FSWs quit sex work because they had established steady relationships (Graham et al., 2013; Tekola, 2005). If applicable, this implies that 17 FSWs who dropped out of our study were at reduced HIV risk by virtue of having 18 19 fewer multiple concurrent partnerships. It is unclear why FSWs enrolled later in the study were more likely to be lost to follow-up than those enrolled earlier; other 20 longitudinal FSWs cohorts have observed this effect (Graham et al., 2013; Su et al., 21 22 2016).

Primary behavioural indicators for HIV acquisition among FSWs include number of 23 sexual partners and condom use with these partners (Odek et al., 2008; Platt et al., 24 2013). New clients may pose a risk to the sexual health of FSWs because their HIV 25 status is more likely to be unknown to the sex workers (Baral et al., 2014). Condom 26 27 use was not significantly associated with LTFU in our analysis. We did however note that FSWs who reported more new clients per month were less likely to be lost to 28 29 follow-up than those who reported none. Our findings suggest that FSWs who completed the study engaged in more behaviours considered to increase HIV risk 30 than FSWs who dropped out. Retained FSWs were therefore well suited for HIV risk 31 reduction counselling provided routinely at follow-up. 32

The Zambian National Registration Act defines a minor as any person under the age 1 of 16. In our cohort, a quarter of sex workers entered the profession as minors. 2 Underage FSWs in Southern Africa have poorer access to health facilities than their 3 adult counterparts (Busza et al., 2016; Inguane et al., 2015). In Mexico, underage 4 entry into sex work was associated with HIV risk factors such as fewer condom 5 negotiation attempts, drug use, and physical and sexual abuse (Goldenberg et al., 6 2012; Servin et al., 2015). We found that beginning sex work as a minor had a 7 protective effect against LTFU. This result strengthens our supposition that FSWs 8 who remained in the study were motivated by a greater need of HIV prevention 9 services than FSWs who were lost to follow-up. 10

11 The literature in Zambia and around the globe shows that alcohol use is associated with an increase in HIV infection (Coldiron et al., 2008; Rehm et al., 2012, 2017; 12 World Health Organization et al., 2018). Sex work often occurs in drinking venues 13 and in conjunction with alcohol consumption (Fritz et al., 2002; Li et al., 2010; Roth et 14 15 al., 2017). The consumption of alcohol among FSWs has been correlated with condomless sex and, STI and HIV acquisition (M. F. Chersich et al., 2007; Matthew F 16 Chersich et al., 2014; Semple et al., 2016; Wang et al., 2010). We found that FSWs 17 who ever used alcohol were more likely to be retained in our study than FSWs who 18 never used alcohol. It is possible that FSWs who consumed alcohol associated this 19 behaviour with a greater risk of HIV acquisition. This would further reinforce that 20 FSWs retained in our cohort perceived a greater benefit of the HIV prevention 21 services offered at every visit. 22

This study has its limitations. Our findings lacked information on structural factors that 23 24 could have been associated with LTFU, such as previous incarceration and migration for the purpose of sex work. Demographic and behavioural variables were self-25 26 reported by FSWs during face-to-face interviews, which could have introduced social desirability and/or recall bias. Biases of this nature were minimised by trained 27 counsellors who administered all questionnaires to study participants. Counsellors 28 received training in voluntary HIV counselling and testing, during which 29 negative/stigmatising attitudes towards participants engaging in high risk behaviours 30 were discouraged. 31

Despite its limitations, our study is—to the best of our knowledge—the first in Zambia
 to examine the incidence and predictors of LTFU among FSWs. These findings add
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to the limited evidence base in sub-Saharan Africa on factors associated with LTFU
among FSWs, who are a high-risk group for HIV infection. We recommend that future
studies build on our quantitative approach by interviewing FSWs who remain, and—if
they can be reached—FSWs who are lost to follow-up. Such studies would provide
vital information on the motivations for FSWs remaining in long-term studies and their
reasons for dropping out.

7 CONCLUSIONS

Taken together, our results demonstrate that LTFU among Zambian FSWs was 8 9 associated with no alcohol use, older age at entry into sex work, and lower volume of new clients. FSWs who were not lost to follow-up appeared to be in greater need of 10 the HIV prevention services offered during the study and may have perceived 11 12 themselves to be at higher HIV risk. This suggests that we retained at-risk individuals who are suitable candidates for future HIV prevention trials. However, more intensive 13 retention efforts -combining in-depth locator information, mapping, and adequate 14 backup measures such as next-of-kin information- will be required during these trials 15 16 to minimise LTFU and ensure the validity and generalisability of findings.

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			Lost to	o Follow-U	p	
		N (%)	n	ΡΥ	Rate per 100 PY	Log-rank
					(95% CI)	test
						(p-value)
All		420 (100)	139	883.5	15.7 (13.32-18.58)	
Year	of enrolment					
	2012/13*	78 (18.5)	18	244.6	7.4 (4.31-11.48)	0.0005
	2014	196 (46.7)	66	415.5	15.9 (12.46-20.26)	
	2015	146 (34.8)	55	223.4	24.6 (19.09-32.38)	
City						
	Ndola	272 (64.8)	81	650.2	12.5 (10.02-15.49)	0.0007
	Lusaka	148 (35.2)	58	233.3	24.9 (19.22-32.16)	
Age						
	18-20	125 (29.8)	36	287.8	12.5 (9.02-17.34)	0.2689
	21-25	142 (33.8)	49	294.2	16.7 (12.59-22.04)	
	26+	153 (36.4)	54	301.5	17.91 (13.72-23.38)	
Educa	ation					
	Primary or none	239 (56.9)	72	526.4	13.7 (10.86-17.23)	0.1027
	Secondary or higher	158 (42.9)	66	356.9	18.5 (14.53-23.54)	
Numb	per of children					
	0	93 (22.1)	29	193.9	15.0 (10.97-22.44)	0.8290
	1	135 (32.1)	47	275.2	17.1 (12.92-23.03)	
	2+	192 (45.8)	63	414.4	15.2 (11.64-19.32)	
Have	a non-paying partner					
	Yes	166 (39.5)	55	349.0	15.8 (12.10-20.52)	0.9946
	No	252 (60.0)	88	531.8	15.6 (12.59-19.35)	
Lifetir	me number of partners					
	<=50	132 (31.4)	48	207.5	23.1 (17.43-30.69)	0.0122
	51-200	111 (26.4)	34	281.8	12.1 (8.62-16.88)	

1 **Table 1:** Rate of LTFU among Zambian FSW by selected baseline characteristics (N=420)

	201-800	68 (16.2)	17	184.3	9.2 (5.73-14.84)	
	800+	107 (25.5)	39	206.0	19.0 (13.83-25.91)	
Age at	sex work initiation					
	<=15	101 (24.1)	21	237.94	8.8 (5.75-13.54)	0.0029
	16+	316 (75.2)	116	640.0	18.1 (15.11-21.74)	
Condo	m use with clients [#]					
	Inconsistent	275 (65.5)	71	380.9	18.6 (14.77-23.52)	-
	Consistent	77 (18.3)	50	338.9	14.8 (11.18-19.46)	
	er of new clients in us month [#]					
	None	19 (4.5)	32	140.4	21.8 (15.44-30.87)	-
	1-4	118 (28.1)	55	372.8	14.6 (11.19-18.98)	
	5-9	95 (22.6)	24	143.9	16.5 (11.06-24.62)	
	10+	128 (30.5)	24	128.0	18.4 (12.31-27.39)	
Ever u	se alcohol					
Yes		325 (77.4)	98	698.7	14.0 (11.51-17.10)	0.0190
No		91 (21.7)	39	177.6	22.0 (16.05-30.06)	
Syphili	s#					
Positiv	e	50 (11.9)	118	77.4	25.8 (16.67-40.05)	-
Negati	ve	370 (88.1)	18	806.1	14.8 (12.33-17.67)	
Tricho	monas <i>vaginalis</i> #					
Positiv	re	35 (8.3)	10	40.9	24.4 (13.15-45.42)	-
Negati	ve	371 (88.4)	126	819.7	15.4 (12.91-18.30)	
*2012 -	and 2012 compliand because	and C ECIM		1:- 2012		

1 *2012 and 2013 combined because only 6 FSW were enrolled in 2012

2 FSW: female sex worker; PY: person-years; #: time-dependent variable (for which log-rank test was

3 *not applicable); CI: confidence interval*

4 Ns don't

5

1 **Table 2:** Factors associated with loss to follow-up in univariate and multivariable Cox regression

2 models for Zambian FSW (N=420)

	Loss to Follow-up				
	Crude HR	95% CI	Adjusted HR	95% CI	
Year of enrolment					
2012/2013	1	-	1	-	
2014	2.08	1.21-3.58**	2.24	1.12-4.47*	
2015	2.93	1.66-5.17***	2.97	1.44-6.14**	
City of residence					
Ndola	1	-	-	-	
Lusaka	1.77	1.26-2.50**	-	-	
Age					
18-20	1		-	-	
21-25	1.32	0.84-1.84	-	-	
26+	1.40	0.92-2.14	-	-	
Education					
Primary or none	1	-	-	-	
Secondary or higher	1.32	0.94-1.84	-	-	
Number of Children					
0	1	-	-	-	
1	1.15	0.72-1.82	-	-	
2+	1.05	0.68-1.64	-	-	
Have a non-paying partner					
Yes	1	-	-	-	
No	1.00	0.71-1.41	-	-	
Lifetime sexual partners					
<=50	1	-	-	-	
51-200	0.59	0.38-0.92*	-	-	
201-800	0.46	0.26-0.80**	-	-	
800+	0.86	0.56-1.31	-	-	

Age at sex work initiation							
≤15	1	-	1	-			
16+	1.99	1.25-3.17**	2.02	1.21-3.37**			
Condom use with clients [#]							
Inconsistent	1	-	-	-			
Consistent	1.12	0.78-1.63	-	-			
Number of new clients in previous month [#]							
None	1	-	1	-			
1-4	0.57	0.37-0.88*	0.52	0.30-0.91*			
5-9	0.50	0.29-0.86*	0.50	0.26-0.95*			
10+	0.46	0.27-0.81**	0.47	0.25-0.91*			
Ever use alcohol							
Yes	1	-	1	-			
No	1.55	1.07-2.24*	1.60	1.04-2.47*			
Syphilis [#]							
Positive	1	-	-	-			
Negative	0.66	0.41-1.07	-	-			
Trichomonas <i>vaginalis</i> #							
Positive	1	-	-	-			
Negative	0.78	0.41-1.49	-	-			

1

2 FSW: female sex worker; HR: hazard ratio; CI: confidence interval; #: time-varying covariate

3 *p-value < 0.05, **p-value < 0.01, ***p-value < 0.001

4

5

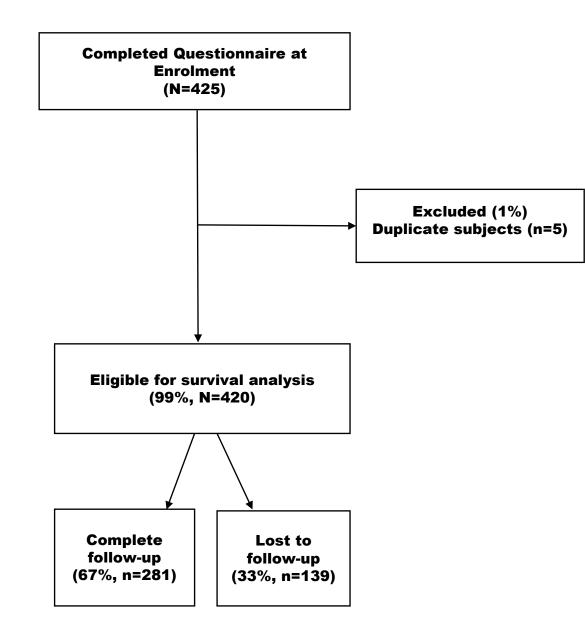


Figure 1: Flow chart showing Zambian FSW who were eligible for survival analysis, lost to follow-up, and completed the fiveyear prospective cohort study (N=420)

