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Factors associated with HIV infection among transgender women in Cambodia: Results from a national integrated biological and behavioral survey

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3 1 **Factors associated with HIV infection among transgender women in Cambodia: Results from a**
4 **national integrated biological and behavioral survey**
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3 30 **ABSTRACT**

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5 31 **Objective:** To examine factors associated with HIV infection among transgender women in
6
7 32 Cambodia.

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9 33 **Design:** Cross-sectional study

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11 34 **Settings:** Thirteen HIV high-burden city and provinces

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13 35 **Participants:** This study included 1,375 sexually active transgender women with a mean age of
14
15 36 participants was 25.9 years (SD= 7.1) recruited by using respondent driven sampling for struc-
16
17 37 tured questionnaire interviews and rapid finger-prick HIV testing.

18
19 38 **Primary outcome measure:** HIV infection detected by using Determine™ antibody test.

20
21 39 **Results:** The overall HIV prevalence among this population was 5.9%. In multivariate logistic re-
22
23 40 gression, participants living in urban areas were twice as likely to be HIV infected as those living
24
25 41 in rural areas. Participants with primary education were 1.7 times as likely to be infected com-
26
27 42 pared to those with high school education (AOR=1.7, 95% CI= 1.0-2.9). HIV infection increased
28
29 43 with age; compared to those aged 18-24, the odds of being HIV infected were twice among
30
31 44 transgender women aged 25-34 and 2.8 times higher among those aged ≥ 35 . Self-injection of
32
33 45 gender affirming hormones was associated with a fourfold increase in the odds of HIV infection.
34
35 46 A history of genital sores over the previous 12 months increased the odds of HIV infection by
36
37 47 threefold. Transgender women with stronger feminine identity dressing up as a woman all the
38
39 48 time were twice as likely to be HIV infected compared to those who did not dress up as a wom-
40
41 49 an all the time. Having never used online services developed for transgender women was also
42
43 50 associated with higher odds of being HIV infected.

44
45 51 **Conclusions :** Transgender women in Cambodia are at high risk of HIV. To achieve the goal of
46
47 52 eliminating HIV in the country, effective combination prevention strategies focusing on the
48
49 53 above risk factors among transgender women are urgently needed.

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51 54
52 55 **Keywords:** Transgender women, HIV infection, Risk factors, Prevention, Integrated Biological
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54 56 and Behavioral Survey, Cambodia

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3 59 **Strengths and limitations of this study**
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- 7 61 ▪ This is the second Integrated Biological and Behavioral Survey (IBBS) that examined fac-
8 tors associated with HIV infection among transgender women—one of the most vulner-
9 62 able populations in Cambodia.
10 63
11 64 ▪ This study covered 13 study sites (seven greater than the coverage of the first IBBS),
12 thus providing more valid and comprehensive results.
13 65
14 66 ▪ This study provides useful data for identifying risk factors for HIV infection and ways in
15 which services and policies can be tailored to reduce HIV infection among this key popu-
16 67 lation.
17 68
18 69 ▪ Limitations of the study included the representativeness of the study sample, the poten-
19 tial bias in participant recruitment, the validity of self-reported measures, and the cross-
20 70 sectional nature of the data.
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75 INTRODUCTION

76 Globally, transgender women are at high risk for HIV infection, and little is known about the
77 burden of HIV infection and its related factors in this population. A 2013 systematic review
78 demonstrated a global HIV prevalence among transgender women of 19%, with 18% in low-
79 and middle-income countries and 22% in high-income countries.¹ The probability of being in-
80 fected with HIV among transgender women was approximately 49 times higher than among the
81 general adult population.¹ Epidemiologically, transgender women share some HIV risk factors
82 with other populations, such as ulcerative sexually transmitted infections (STIs),^{2,3} multiple sub-
83 stance use, victimization, intimate partner violence,⁴ unprotected sex,⁵ multiple sex partners,⁶
84 and injecting drug use.⁵⁻⁷

85 However, transgender women encounter additional and unique biological, social, cul-
86 tural, legal, and economic issues, which increase their vulnerability to HIV. Transgender women
87 are marginalized legally and economically through discriminative legislation and practice, which
88 accelerates their vulnerability to HIV infection by confining their job options to sex work. Sex
89 work in turn exacerbates their stigmatization and alienation,⁸ and further increases their HIV
90 risk.^{9,10} Stigmatization and discrimination further increase the risk of HIV infection by prevent-
91 ing transgender women from seeking essential health information and services.¹¹ Depression
92 and poor self-esteem, the major psychosocial consequences of stigma, further weaken the bar-
93 gaining power for safer sex in sexual relationships, resulting in low condom use.^{5, 12-15}
94 Transgender women who have sex with men often engage in receptive anal intercourse, which
95 increases their risk of HIV acquisition, compared to insertive sexual partners.² Syndemics of illic-
96 it drug use, abuse by family members, and mental health often occur together;¹⁶ and are often
97 associated with less likelihood of transgender women to use condoms during sex with commer-
98 cial partners.¹⁷ In addition, the confluence of arbitrary arrests,^{18,19} violence,^{16,20} economic pres-
99 sures,²¹ and other social problems further aggravate mental health risks among transgender
100 women, causing them to engage in risky sexual behaviors.^{6,17}

101 In Southeast Asia, systematic reviews of studies in Indonesia, Thailand, and Vietnam
102 demonstrated that transgender women were particularly at high HIV risk, with estimated
103 prevalence of 26.1%, 12.2%, and 6.7%, respectively.¹ In Cambodia, up to 2012, transgender

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3 104 women were traditionally included in HIV prevention programs targeting men who have sex
4 with men (MSM). As a result, their unique needs were not reflected in the programs' goals.²²
5 105
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7 106 There was also a lack of transgender women-specific data to quantify the number reached by
8 outreach programs and services. In 2012, the first Integrated Biological and Behavioral Survey
9 107
10 108 (IBBS) that specifically included transgender women in Cambodia found a HIV prevalence of
11 4.2% among this population,²² which was six times higher than the 0.7% among the general
12 109
13 110 adult population aged 15-45,²³ and about twice higher than the 2.1% among MSM.²⁴ This 2012
14 study also found that older age (>35 years), residence in Siem Reap province (a major tourist
15 111
16 112 site), having sex during or after drug use, not using condoms during the last anal sex, and low
17 self-esteem were associated with HIV infection among transgender women.²²
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22 114 Recognizing the differences in HIV vulnerability and prevention needs between
23 transgender women and MSM, in 2013 the National Center for HIV/AIDS, Dermatology and STD
24 115
25 (NCHADS) separated these two populations in its revised Standard Operating Procedure (SOP)
26 116
27 known as Boosted Continuum of Prevention, Care and Treatment (B-CoPCT).^{22 25} Transgender
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29 women have since been considered a distinct at-risk group in HIV surveillance and programs.
30 118
31 119 The National B-CoPCT approach aims to increase the uptake of HIV testing and counseling (HTC)
32 and other related services, ensure the quality of outreach, and strengthen service delivery to
33 120
34 meet specific needs of most-at-risk key populations, including transgender women, in-order to
35 121
36 achieve Cambodia's 3.0 goal (i.e. zero new HIV infection, zero discrimination, and zero AIDS-
37 122
38 related deaths) by 2020.
39 123

40
41 124 Three years following the first IBBS in 2012,²² the TGIBBS-2016 was conducted between
42 December 2015 and February 2016 to identify factors associated with HIV infection among
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44 transgender women in Cambodia and ways in which services and policies can be tailored for
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46 this key population.
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50 129 **METHODS**

51 130 **Study sites**

52 131 Data collection was conducted in the capital city of Phnom Penh and 12 provinces including
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54 132 Battambang, Banteay Meanchey, Kampong Cham, Kandal, Koh Kong, Kampong Chhnang, Kam-
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133 pong Speu, Prey Veng, Preah Sihanouk, Siem Reap, Svay Rieng, and Tbong Khmum. These sites
 134 were purposively selected, for they were within the 23 HIV high-burden operational districts
 135 identified by NCHADS.^{26 27}

136

137 **Sample size and sampling procedures**

138 The sample size estimate was determined for the purpose of surveillance to track the change in
 139 the epidemic over time. The minimum sample size required for this study was 1,380, with suffi-
 140 cient power to detect significant differences between the following null and alternative hy-
 141 potheses. The hypothesis test was that of one-sample proportion in comparison to the null hy-
 142 pothesis.

143 The null hypothesis (H_0): HIV prevalence among transgender women was 4.2%. The al-
 144 ternative hypothesis (H_a): HIV prevalence among transgender women will decrease by 1.2%.
 145 The sample size was calculated using the following formula and assumptions:

$$n = \frac{p_0 q_0 \left(z_{1-\alpha} + z_{1-\beta} \sqrt{p_1 q_1 / p_0 q_0} \right)^2}{(p_1 - p_0)^2}$$

146

- 147 ▪ P_0 = estimated proportion in H_0 (In the most recent survey, HIV prevalence among TG
 148 women was 4.2% in 2012.)
- 149 ▪ P_1 = estimated proportion in H_a (The expected HIV prevalence among TG women in the
 150 current study is about 3.0 %)
- 151 ▪ $Z_{(1-\alpha)}$ = significance level at 5% in response to one-sided test (Z score = 1.645)
- 152 ▪ $Z_{(1-\beta)}$ = power level of 80% (Z score = 0.83)
- 153 ▪ Refusal rate of 10%
- 154 ▪ Design effect is assumed to be one.

155

156 This sample size was stratified by the study sites. Roughly half of the estimated transgender
 157 women in all but five sites were successfully recruited. In five study sites where the estimated
 158 number of transgender women was smaller than 100, all transgender women were recruited.
 159 The total estimated sample size was 1,368.

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3 160 Individuals were included in the study if they: (1) were biologically male at birth and
4
5 161 self-identified as a woman or third gender, (2) were Khmer-speaking, (3) were at least 18 years
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7 162 old at the time of screening, (4) reported having sex with at least one man in the past 12
8
9 163 months, and (5) were able and willing to provide an informed consent.

10 164 Among the 13 study sites, participant recruitment was conducted in 20 locations (six lo-
11
12 165 cations in Phnom Penh and 14 locations in the other provinces). The number of the selected
13
14 166 locations was determined based on the proportion of the required sample size in each study
15
16 167 site. Participants were recruited through a Respondent Driven Sampling (RDS) method. First,
17
18 168 four “seeds” (two seeds aged 18 to 24 and the other two seeds aged 25 or older) who were well
19
20 169 connected to other transgender women in each location were selected by outreach workers
21
22 170 working for the implementing partners (local NGOs) based in the selected locations. These
23
24 171 seeds had to meet the above-mentioned eligibility criteria for participation and have an
25
26 172 established and large social network comprising about 10 or more other transgender women in
27
28 173 their given location. Eligibility to participate as a seed was determined by the leader of data
29
30 174 collection team using a paper-based eligibility form.

31 175 Second, each seed was given a personal identity number (PIN) and enrolled in the study.
32
33 176 Third, each seed was given three coupons and asked to refer three additional transgender
34
35 177 women. US\$2 was given to each seed for a successful referral. Each seed was expected to ex-
36
37 178 tend to three to six “recruitment waves” in each site. If the initial seeds did not recruit
38
39 179 participants, or if enrollment was halted because all recruitment chains had “dried up” (i.e.
40
41 180 stopped recruiting), additional seeds would be selected. In total, 80 seeds were selected by the
42
43 181 outreach workers, and a total of 1,375 transgender women were enrolled in the study. Referred
44
45 182 participants were initially screened by the data collectors for eligibility.

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48 184 **Data collection training and procedures**

49 185 Data were collected by three teams; each team comprised of one field supervisor, five inter-
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51 186 viewers, one lab technician, and one counselor. Lab technicians and counselors were from the
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53 187 Municipal or Provincial AIDS and STI Program of the study sites. These data collection teams
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3 188 were trained for three days on study protocol, research ethics, interview techniques, and data
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5 189 collection procedures provided by the principle investigators and research coordinators.
6

7 190 To determine HIV prevalence, all participants received onsite rapid finger-prick testing.
8
9 191 Pre-test counseling was provided by qualified, trained counselors working for volunteer confi-
10
11 192 dential and counseling testing (VCCT) centers. Participants could receive their HIV-test result
12
13 193 verbally after the questionnaire interview. Blood samples were obtained from each participant
14
15 194 by trained lab technicians through finger-prick and tested for HIV using Determine™ test, in
16
17 195 keeping with the national protocol.²⁸ Post-test counseling was provided for each participant
18
19 196 regardless of their HIV test result by the same counselor who conducted the pre-test counseling
20
21 197 in accordance with NCHADS HIV testing guidelines.²⁸ Participants who were HIV reactive and
22
23 198 did not know their HIV status were referred by the counselor for confirmatory testing at the
24
25 199 nearest VCCT center. The quality of HIV tests was monitored using quality control samples. Dis-
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27 200 cordant results between the screening and confirmatory tests were investigated to elucidate
28
29 201 potential causes and minimize potential erroneous results. In the case of human error, individ-
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31 202 uals performing screening tests were retrained before resuming their involvement in the study.

32 203 After HIV testing, the participants were interviewed by the trained interviewers using a
33
34 204 computer-assisted survey instrument (CASI). The questionnaire was set up in Qualtrics, a web-
35
36 205 based application, and run on an Android tablet. HIV testing and interviews were conducted in
37
38 206 the selected 20 locations including drop-in centers, private houses, and private offices of im-
39
40 207 plementing partners, depending on the participants' convenience.
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43 209 **Questionnaire development and measures**

44 210 The questionnaire was developed using standardized and validated tools adapted from TGIBBS-
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46 211 2012,²² FHI 360's guideline for Behavioral Surveillance Survey (BSS),²⁹ and NCHADS's Boosted-
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48 212 CoPCT²⁵ to measure key variables related to the objectives of the study. The questionnaire was
49
50 213 initially drafted in English and translated into Khmer, the national language of Cambodia. Con-
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52 214 sultative meetings to improve the draft questionnaire were held with representatives of
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54 215 transgender women, communities, and NGOs working with transgender women, as well as re-
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3 216 searchers and practitioners working on HIV and AIDS in Cambodia. The questionnaire was pre-
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5 217 tested with 20 transgender women in Phnom Penh and finalized based on findings.
6

7 218 The questionnaire collected information on socio-economic characteristics, transgender
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9 219 identity and related experiences, sexual behaviors and condom use with different types of sex-
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11 220 ual partners, HIV/STI screening and care seeking behaviors, substance use, and exposure to HIV
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13 221 programs. For details of the questionnaire, please see Supplementary File 1.
14

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16 223 **Data analyses**

17 224 HIV prevalence was calculated by dividing the total number of participants with HIV reactive
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19 225 test results with the total number of participants. To examine the associated factors of HIV in-
20
21 226 fection, we conducted both bivariate and multivariate analyses. In the bivariate analyses, we
22
23 227 compared all characteristics and behavioral variables among participants with a reactive result
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25 228 to those among participants with a non-reactive test result. Chi-square test or Fisher's exact
26
27 229 test (for an expected cell value ≤ 5) was used for categorical variables and Student's *t*-test was
28
29 230 used for continuous variables. To facilitate the model, some continuous variables, such as age
30
31 231 were transformed to categorical variables. A multivariate logistic regression model was con-
32
33 232 structed to examine independent factors associated with HIV infection. Variables with a signifi-
34
35 233 cance level of $p < 0.05$ in the bivariate analyses were simultaneously included in the model.
36
37 234 Backward elimination method was used to eliminate variables with the highest p -value one-by-
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39 235 one from the model. STATA Version 12.0 for Windows (Stata Corp, TX, USA) was used to con-
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41 236 duct the data analyses.
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44 238 **ETHICAL STATEMENT**

45 239 Participation in this study was voluntary, and a written informed consent was obtained from
46
47 240 each study participant after a detailed description of the study objective and procedures was
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49 241 explained to them. Participants were informed that they could stop responding to questions
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51 242 and discontinue their participation at any time. Interviews were conducted at a private place,
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53 243 and confidentiality was enhanced by assigning a PIN to each participant. No personal identifiers
54
55 244 were contained in the questionnaires or dataset. The study protocol was approved by FHI 360's
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245 Protection of Human Subjects Committee (PHSC No. 713897) and the National Ethics Commit-
246 tee for Health Research (NECHR) of the Ministry of Health, Cambodia (No. 420 NECHR).

247

248 RESULTS

249 HIV prevalence

250 Of 1,375 participants tested, 81 (5.9%) had an HIV reactive test; of whom, 42 (52%) were not
251 aware of their HIV status prior to the study. Out of the 39 cases who already knew their status,
252 37 (94.9%) were on treatment, and two of them reported dropping the treatment.

253

254 Socio-demographic characteristics

255 As shown in Table 1, the majority of the participants (83.4%) were recruited from urban com-
256 munities; 53.0% were younger than 25 years old; and 97.2% were never married. More than
257 two-thirds of the participants (68.6%) had completed high school, and 9.1% had higher educa-
258 tion. The most common main occupations reported by the participants were hair dress-
259 ers/beauticians (35.1%), laborers/farmers (17.5%), and entertainment workers (14.8%). More
260 than one-third of the participants (38.6%) reported their average monthly income in the past
261 six months of US\$ 100-199, while 16.5% reported it to be more than US\$ 300. Regarding gender
262 identity, 42.2% identified themselves as female, while 57.2% identified themselves as third
263 gender. Almost half (48.0%) of the participants reported dressing up as a woman all the time;
264 45% ever used hormone/non-hormone substance; and 14.0% ever injected hormone.

265 HIV prevalence was significantly higher among participants living in urban communities
266 compared to that among participants living in rural communities (6.5% vs. 2.6%; $p= 0.02$). The
267 prevalence was also significantly different in age groups: 3.0% in participants in the age group
268 of 18-24, 8.2% in 25-34, 13.1% in 35-44, and 11.4% in 45 or older ($p< 0.001$). HIV prevalence
269 was also significantly higher among those who never attended school or dropped out of prima-
270 ry school when compared to those who at least attended high school or higher (10.4% vs. 8.0%;
271 $p= <0.001$). In addition, HIV prevalence was significantly higher among transgender women who
272 were NGO staff (mainly HIV focused NGOs) (20.6%) and unemployed (9.4%) ($p= 0.03$). Regard-
273 ing gender identity, HIV prevalence was significantly higher among transgender women who

274 reported dressing up in female-gendered clothing all the time (8.5% vs. 3.5%, $p= 0.001$), among
 275 those who ever used female hormone (8.1% vs. 4.1%, $p= 0.02$), and among those who ever in-
 276 jected hormone (9.1% vs. 5.2%, $p= 0.002$) compared to those who did not. Moreover, HIV prev-
 277 alence was significantly higher among transgender women who reported never using online
 278 services developed MSM or transgender women such as Facebook pages, websites, etc. (9.5%
 279 vs. 3.9%, $p< 0.001$).

280

281 **Table 1** Comparisons of socio-demographics, gender identity, and hormone use among

282 transgender women with HIV reactive and non-reactive test

Socio-demographics, gender identity, and hormone use	Total (<i>n</i> = 1375)	HIV test result		<i>P</i> -value*
		Reactive (<i>n</i> = 81)	Non-reactive (<i>n</i> = 1294)	
	<i>n</i> (%)	<i>n</i> (%)	<i>n</i> (%)	
Community type				
Urban	1146 (83.4)	75 (6.5)	1071 (93.5)	0.02
Rural	229 (16.6)	6 (2.6)	223 (97.4)	
Age in years				
18- 24	729 (53.0)	22 (3.0)	707 (97.0)	<0.001
25-34	503 (36.6)	41 (8.2)	462 (91.8)	
≥35	143 (10.4)	18 (12.6)	125 (87.4)	
Current marital status				
Married	7 (0.5)	0 (0.0)	7 (100)	0.47
Widowed/divorced/separated	18 (1.3)	1 (5.6)	17 (94.4)	
Never married	1334 (97.2)	78 (5.9)	1256 (94.1)	

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4	Refuse to answer	16 (1.2)	2 (15.4)	11 (84.6)	
5					
6	Years of formal education completed				
7					
8					
9	Primary (0-6 years)	307 (22.3)	32 (10.4)	275 (89.6)	<0.001
10					
11	High school or higher (>7 years)	1068 (77.7)	49 (4.6)	1019 (95.4)	
12					
13					
14	Main occupation				
15					
16	Unemployed	64 (4.7)	6 (9.4)	58 (90.6)	0.03
17					
18	Hair dresser/beautician	482 (35.1)	24 (5.0)	458 (95.0)	
19					
20	Office worker	50 (3.6)	3 (6.0)	47 (94.0)	
21					
22	Labor/farmer	241 (17.5)	15 (6.2)	226 (93.8)	
23					
24	Seller	149 (10.8)	10 (6.7)	139 (93.3)	
25					
26	Entertainment Worker	203 (14.8)	10 (4.9)	193 (95.1)	
27					
28	Student	108 (7.9)	1 (0.9)	107 (99.1)	
29					
30	NGO staff	34 (2.5)	7 (20.6)	27 (79.4)	
31					
32	Other	44 (3.2)	5 (11.4)	39 (88.6)	
33					
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39	Monthly income in the past 6 months (USD)				
40					
41	< 100	351 (25.6)	22 (6.3)	329 (93.7)	0.70
42					
43	100-199	530 (38.6)	29 (5.5)	501 (94.5)	
44					
45	200-299	266 (19.3)	19 (7.1)	247 (92.9)	
46					
47	≥300	226 (16.5)	11 (4.9)	215 (95.1)	
48					
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52	Gender identity (self-identified)				
53					
54					
55	Female	580 (42.2)	29 (5.0)	551 (95.0)	0.35
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60					

Third gender	786 (57.2)	52 (6.6)	734 (93.4)	
Uncertain	8 (0.6)	0 (0.0)	8 (100)	
Frequency of dressing up in female-gendered clothing				
All the time	660 (48.0)	56 (8.5)	604 (91.5)	0.001
Not all the time	715 (52.0)	25 (3.5)	689 (96.5)	
Ever use injected hormone				
No	1123 (81.7)	58 (5.2)	1065 (94.8)	0.02
Yes	252 (18.3)	23 (9.1)	229 (90.9)	
Ever self-injected hormones				
No	1,358 (98.8)	77 (5.7)	1281 (94.3)	0.002
Yes	17 (1.2)	4 (25.5)	13 (76.5)	
Ever shared needle during injecting hormone/beauty substance				
No	1355 (98.5)	78 (5.8)	550 (94.2)	0.33
Yes	20 (3.2)	2 (10.0)	18 (90.0)	
Ever used online services developed MSM/TG (e.g. Facebook, website)				
No	483 (35.1)	46 (9.5)	437 (90.5)	<0.001
Yes	892 (64.9)	35 (3.9)	857 (96.1)	

Abbreviations: HIV, human immunodeficiency virus; USD, United States dollar.

*Chi-square or Fisher's exact test was used as appropriate.

Sexual behaviors

Sexual behaviors among transgender women with reactive and non-reactive HIV test are shown in Table 2. An overwhelming majority of study participants (87.5%) reported only having had receptive anal sex in the past 12 months. Of the total, 86.0% reported having had anal sex with at least one man within the previous three months, with the median number of male sex part-

ners in the past three months of three (IQR 1-9). Of those who were sexually active with men in the past three months, 61.9% reported having used a condom at last sex with a man.

A total of 94.8% of participants who had anal sex in the past three months reported having at least one non-transactional male sex partner within the past three months; of which, 62.1% reported always using condoms with non-transactional male sex partners in the past three months. Of participants who had sex in the past three months, 41.8% reported having sex with at least one man in exchange for money or gift in the past three months; of whom 60.0% reported always using condoms with the partners.

Table 2 also shows that HIV prevalence was significantly higher among participants who reported having anal sex with a man in the past three months (6.5% vs. 2.1%, $p= 0.02$) and who reported having sex with at least one man in exchange for money or gift in the past three months (8.1% vs. 5.4%, $p= 0.04$) compared to that among participants who did not.

303

Table 2 Comparisons of sexual behaviors among transgender women with reactive and non-reactive HIV test

Sexual behaviors	Total (<i>n</i> = 1375)	HIV test result		<i>P</i> -value*
		Reactive (<i>n</i> = 81)	Non-reactive (<i>n</i> = 1294)	
	<i>n</i> (%)	<i>n</i> (%)	<i>n</i> (%)	
Role in anal sex with a man (past 12 months)				
Incentive	29 (2.2)	1 (3.5)	28 (96.5)	0.73
Receptive	1145 (87.5)	72 (6.3)	1073 (93.7)	
Both	135 (10.3)	7 (5.2)	128 (94.8)	
Had anal sex with a man (past 3 months)				
No	192 (14.0)	4 (2.1)	188 (97.9)	0.02

1					
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3					
4	Yes	1183 (86.0)	77 (6.5)	1106 (93.5)	
5					
6	Number of male sexual partners (past 3 months)				
7					
8					
9	<2	512 (37.3)	24 (4.7)	488 (95.3)	0.14
10					
11	≥2	861 (62.7)	57 (6.6)	804 (93.4)	
12					
13					
14	Used condom last sex (past 3 months)				
15					
16	No	451 (38.1)	27 (6.0)	423 (94.0)	0.57
17					
18	Yes	732 (61.9)	50 (6.8)	682 (93.2)	
19					
20					
21	Had anal sex with a man not in exchange for money or gift (past 3 months)				
22					
23	No	61 (5.2)	5 (8.2)	56 (91.8)	0.37
24					
25	Yes	1122 (94.8)	72 (6.4)	1050 (93.6)	
26					
27					
28					
29	Number of male sexual partner, not in exchange for money or gift (past 3 months)				
30					
31	< 2	357 (31.9)	24 (6.7)	333 (93.3)	0.78
32					
33	≥2	763 (68.1)	48 (6.3)	715 (93.7)	
34					
35					
36					
37	Condom use with male sexual partner not in exchange for money or gift (past 3 months)				
38					
39	Not always	697 (62.1)	50 (7.2)	647 (92.8)	0.19
40					
41	Always	425 (37.9)	22 (5.2)	403 (94.8)	
42					
43					
44					
45	Had anal sex with a man in exchange for money (past 3 months)				
46					
47	No	688 (58.2)	37 (5.4)	651 (94.6)	0.04
48					
49	Yes	495 (41.8)	40 (8.1)	455 (91.9)	
50					
51					
52	Number male sexual partners in exchange for money/gifts (past 3 months)				
53					
54					
55	1	161 (32.5)	12 (7.5)	149 (92.6)	0.72
56					
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4 ≥ 2 334 (67.5) 28 (8.4) 306 (91.6)

5
6 Condom use with male sexual partner in exchange for money or gifts (past 3 months)

7
8
9 Not always 164 (40.0) 12 (7.3) 152 (92.7) 0.56

10
11 Always 246 (60.0) 22 (8.9) 224 (91.1)

12
13
14 306 *Abbreviation: HIV, human immunodeficiency virus.*

15 307 **Chi-square test or Fisher's exact test was used as appropriate.*

16 308
17
18 309 **Sexually transmitted infections (STIs)**

19 310 Overall, 14.0% of participants reported having had at least one STI symptom in the past 12
20 311 months. Anal and perianal symptoms on the anus were the most common symptoms (6.1%). As
21 312 shown in Table 3, HIV prevalence was significantly higher among participants who reported
22 313 having had an STI symptom compared to that among participants who did not have an STI
23 314 symptom in the past 12 months (11.4% vs. 5.0%; $p < 0.001$). HIV prevalence was also significant-
24 315 ly higher among participants who reported having had an ulceration or sore in the genital area
25 316 in the past 12 months compared to that among participants who did not have it (15.2% vs.
26 317 5.6%; $p < 0.001$).
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37 319 **Table 3** Comparisons of STI symptoms among transgender women with reactive and non-
38 320 reactive HIV test

STI symptoms	Total (n= 1375)	HIV test result		P-value*
		Reactive (n= 81)	Non-reactive (n= 1294)	
	n (%)	n (%)	n (%)	
Had experienced any STI symptom (past 12 months)				
No	1182 (86.0)	59 (5.0)	1123 (95.0)	<0.001
Yes	193 (14.0)	22 (11.4)	171 (88.6)	

Ulceration or sores in the genital area (past 12 months)

No	1329 (96.7)	74 (5.6)	1255(94.4)	<0.001
Yes	46 (3.4)	7 (15.2)	39 (84.8)	

Swelling in the genital area (past 12 months)

No	1359 (98.8)	80 (5.9)	1279 (94.1)	1.000
Yes	16 (1.2)	1 (6.3)	15 (93.7)	

Abnormal urethral discharge (past 12 months)

No	1339 (97.4)	77 (5.8)	1262(94.2)	0.16
Yes	36 (2.6)	4 (11.1)	32 (88.9)	

STI symptoms on anal area (past 12 months)

No	1291 (93.9)	73 (5.6)	1218 (94.4)	0.15
Yes	84 (6.1)	8 (9.52)	76 (90.48)	

STI symptom in the mouth or throat (past 12 months)

No	1336 (97.2)	76 (5.7)	1260 (94.3)	0.07
Yes	39 (2.8)	5 (12.82)	34 (87.18)	

321 *Abbreviations: HIV, human immunodeficiency virus; STI, sexually transmitted infections.*

322 **Chi-square or Fisher's exact test was used as appropriate.*

323

324 **Substance use**

325 As shown in Table 5, the majority (75.9%) of participants reported drinking at least one can of
 326 beer or a glass of wine in the past 3 months, while 10.1% reported using amphetamine-type
 327 stimulants (Yama, Crystal Ice, Ecstasy), and 0.9% reported using other drugs (marijuana, Heroin,
 328 etc.) in the previous 12 months. Of total, 1.5% reported having injected any illicit drugs in the
 329 past 3 months.

330 Table 4 also shows that HIV prevalence was significantly higher among participants who
 331 did not drink alcohol when compared to that among participants who reported drinking at least
 332 one can of beer or a glass of wine in the past three months (8.5% vs. 5.1%; $p= 0.02$). Moreover,
 333 HIV prevalence was significantly higher among participants who reported injecting any illicit
 334 drugs in the past 3 months compared that among participants who did not (15.0% vs. 5.8%, $p=$
 335 0.01).

336

337 **Table 4** Comparisons of substance use among transgender women with reactive and non-
 338 reactive HIV test

339

Substance use	Total (<i>n</i> = 1375)	HIV test result		<i>P</i> -value*
		Reactive (<i>n</i> = 81)	Non-reactive (<i>n</i> = 1294)	
	<i>n</i> (%)	<i>n</i> (%)	<i>n</i> (%)	
Drank at least one can of beer or glass of wine in the past 3 months				
No	331 (24.1)	28 (8.5)	303 (91.5)	0.02
Yes	1042 (75.9)	53 (5.1)	989 (94.9)	
Frequency of having more than 5 drinks in one day in the past 3 months				
Never more than five drinks	336 (24.4)	28 (8.3)	308 (91.7)	0.17
Less than once a month	771 (56.1)	40 (5.2)	731 (94.8)	
1-3 times a week	157 (11.4)	9 (5.7)	148 (94.3)	
4 or more times a week	111 (8.1)	4 (3.6)	107 (96.4)	
Used illicit drugs in the past 12 months				
Never	1224 (89.0)	72(5.9)	1152 (94.1)	0.74

Yes, ATS (yama, crystal ice, ecstasy)	139 (10.1)	8 (5.8)	131 (94.2)	
Other (marijuana, heroin, etc.)	12 (0.9)	1 (8.3)	11 (91.7)	
Injected any illicit drugs in the past 3 months				
No	1355 (98.6)	78 (5.8)	1277 (94.2)	0.01
Yes	20 (1.5)	3 (15.0)	17 (85.0)	
Had sex during/after using illicit drugs in the past 3 months				
No	1286 (93.5)	76 (5.9)	1210 (94.1)	1.00
Yes	89 (6.5)	5 (5.6)	84 (94.4)	

Abbreviation: ATS, amphetamine-type stimulant; HIV, human immunodeficiency virus.

*Chi-square or Fisher's exact test was used as appropriate.

Factors associated with HIV infection

Table 5 presents independent factors associated with HIV infection in multivariate logistic regression analyses. After adjustment for other covariates, participants living in urban areas were twice as likely to be HIV infected as those living in rural areas (AOR= 2.7, 95% CI= 1.1-6.5). Participants with primary education were 1.7 times as likely to be infected compared to those with high school education (AOR=1.7, 95% CI= 1.0-2.9). HIV infection increased with age; compared to those aged 18-24, the odds of being HIV infected were twice among transgender women aged 25-34 (AOR= 2.1, 95% CI= 1.2-3.6) and 2.8 times higher among those aged ≥ 35 (AOR= 2.8, 95% CI= 1.3-6.1). Self-injection of gender affirming hormones was associated with a four-fold increase in the odds of HIV infection (AOR= 4.4, 95% CI= 1.1-17.3). A history of genital sores over the previous 12 months increased the odds of HIV infection by three-fold (AOR= 3.0, 95% CI= 1.2-7.8). Transgender women with stronger feminine identity dressing up as a woman all the time were twice as likely to be HIV infected compared to those who did not dress up as a woman all the time (AOR= 2.1, 95% CI= 1.2-3.8). Having never used online services developed for transgender women in the past six months was also associated with higher odds of being HIV infected (AOR= 1.9, 95% CI= 1.2-3.2).

359

360 **Table 5** Factors associated with HIV infection in multivariate logistic regression model

Variables in the final model	AOR (95% CI)	P-value
Community type		
Rural	Reference	
Urban	2.7 (1.1-6.5)	0.03
Formal education attained		
Primary (0 – 6 years)	1.7 (1.1-2.9)	0.04
High school or higher (≥ 7)	Reference	
Age in years		
<25	Reference	
25-34	2.1 (1.2-3.6)	0.01
≥ 35	2.6 (1.3-5.4)	0.01
Frequency of express and/or dressing up as a woman		
Not all the time	Reference	
All the time	2.1 (1.2-3.8)	0.01
Ever self-injected hormone		
No	Reference	
Yes	4.4 (1.1-17.3)	0.03
Ulcerations or sores in the genital area in the past 12 months		
No	Reference	

Yes	3.0 (1.2-7.8)	0.02
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Used online services developed MSM/TG in the past 6 months (e.g. Facebook, website)

No	1.9 (1.2-3.2)	0.01
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Yes	Reference	
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361 *Abbreviations: AOR, adjusted odds ratio; CI, confidence interval; HIV, human immunodeficiency virus.*

362 **Variables in the table were the ones that remained statistically significant after several steps of model*
 363 *fitting.*

365 DISCUSSION

366 This study reports important findings related to factors associated with HIV infection among
 367 transgender women in Cambodia. First, the HIV prevalence among transgender women residing
 368 in urban communities was twice compared to that among transgender women living in rural
 369 communities. This finding is similar to that in a previous study that found higher HIV prevalence
 370 in transgender women living in urban areas of Siem Reap and Phnom Penh, compared to that
 371 among transgender women living in other provinces.²² Findings from our study suggest that
 372 these differences may not necessarily be attributable to common sexual risk practices. The
 373 mean number of sexual partners (8.4 vs. 7.9; p -value=0.70) and the mean number of male
 374 commercial sexual partners (0.7 vs. 0.7; p -value= 0.48) in the past three months among
 375 transgender women living in urban and rural communities were not significantly different. In
 376 addition, the rates of inconsistent condom use with male non-commercial sexual partners in
 377 the past three months were also similar in the two groups (58.0% vs. 62%; p = 0.32).

378 Given the failure of sexual risk behaviors accounting for the difference in HIV preva-
 379 lence, these data suggest that a higher HIV prevalence among sexual partners of transgender
 380 women in urban communities may explain the higher probability of urban transgender women
 381 to get infected, although this may not be stated with certainty. This is particularly relevant giv-
 382 en that previous studies among all transgender women found a higher prevalence of HIV in ur-
 383 ban areas.²² Studies among MSM in Cambodia have also found a higher prevalence of HIV in
 384 urban areas.^{24 30} In addition, a recent national sentinel survey among women attending antena-
 385 tal care clinics found that HIV prevalence among women in urban areas was consistently high,³¹

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3 386 implying that HIV prevalence in urban areas among most populations, who potentially include
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5 387 transgender women and their partners, is high. It is also possible that transgender population in
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7 388 urban communities face unique or additional risks compared to other members of this popula-
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9 389 tion in rural areas,²² which could also contribute to the difference in HIV prevalence between
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11 390 urban and rural communities. Nevertheless, further research is needed with regard to this ob-
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13 391 servation.

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15 392 Second, HIV prevalence among older transgender women (≥ 25 years) was significantly
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17 393 higher, compared to that among their younger counterparts. This finding is similar to those of
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19 394 previous studies in Cambodia²² and Thailand.³ A possible explanation is that older transgender
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21 395 women had been exposed to risks for a longer period of time; the number of their sexual part-
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23 396 ners accumulated over time and thus increased cumulative probability of HIV acquisition.²²

24 397 Third, our study suggested that more overt feminine expression (e.g., those who wore
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26 398 female gendered clothes all the time) was associated with a higher HIV prevalence among
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28 399 transgender women. A study in the United States found that transgender women who disclosed
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30 400 their female gender identity possessed a higher HIV prevalence.³² In our study, these
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32 401 transgender women who dressed up as a woman all the time had higher numbers of both male
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34 402 non-commercial (mean= 8.1 vs. 5.1; $p= 0.004$) and male commercial sexual partners (mean= 3.7
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36 403 vs. 1.6; $p < 0.001$), compared to those who did not. Making their female status visible could ren-
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38 404 der more chances to meet with men. However, it could make them difficult to find a decent job
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40 405 due to stigmatization and discrimination, thus leading them to engage in sex work. This finding
41
42 406 is in line with existing literature showing that transgender women try to have sex with men –
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44 407 both casual and paid sex– to prove or validate their female gender identity.^{33–35} In these con-
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46 408 texts, “sex work provides both desired gender affirmation and economic stability, often with
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48 409 greater financial rewards for sex without a condom.”³³ The desire to affirm their gender identi-
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50 410 ty and attractiveness to men also incentivizes them to engage in casual sex with multiple part-
51
52 411 ners.¹⁵ This sexual behavior makes them more prone to HIV infection.

53 412 There is a tendency to self-inject gender-affirming hormones, often with shared needles,
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55 413 among transgender women in Cambodia.³⁶ This risky practice was associated with an increased
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57 414 risk of HIV infection, implying that self-injecting was performed through unsafe means, and that
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3 415 injecting by a trained health or other professional might mitigate the likelihood of HIV infection.
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5 416 Although hormone injection to augment femininity is becoming more common, it can potential-
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7 417 ly cause adverse health effects among transgender women.^{3 37} A Thai study also found that
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9 418 transgender women who injected hormone to make them more feminine had a higher HIV
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11 419 prevalence.³

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13 420 Fourth, transgender women who completed or dropped out primary school had a higher
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15 421 HIV prevalence than those with high-school education. This finding confirms results of the
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17 422 TGIBBS 2012 in Cambodia²² and other studies^{6 38} that associated low education with high HIV
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19 423 prevalence among transgender women. This has important implications for HIV programs to
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21 424 ensure that poorly educated transgender women are reached with education, information,
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23 425 communication, and skills related to HIV prevention and other health-related services.

24
25 426 Fifth, transgender women with self-reported STI symptoms (having ulcerations or sores
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27 427 in the genital area in the past 12 months) had a greater HIV prevalence. This is in line with exist-
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29 428 ing evidence showing that STIs promote HIV transmission via a variety of biological mecha-
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31 429 nisms.³⁹ Transgender women with high rates of STIs, particularly ulcerative genital diseases, are
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33 430 at high risk of HIV acquisition.^{2 3}

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35 431 Finally, transgender women who reported never using online services developed for
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37 432 transgender women or MSM had an increased risk of HIV infection. Low utilization of online
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39 433 services by key populations in Cambodia is iterated in other studies.⁴⁰ Non-users of online ser-
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41 434 vices might have riskier sexual behaviors. Our data suggest that transgender women who did
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43 435 not use online services had higher numbers of overall male sexual partners (10.2 vs. 6.3, $p=$
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45 436 0.002) and male commercial sexual partners (0.8 vs. 0.6, p -value= 0.001) in the past three
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47 437 months. Also, they had a higher rate of inconsistent condom use with male commercial sexual
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49 438 partners in the past three months (48.5% vs. 34.4%, $p=$ 0.004). Studies in other settings confirm
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51 439 that transgender women with multiple sexual partners are exposed to a riskier level of HIV in-
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53 440 fection.^{6 15} This finding suggests that access to community-based services tailored to
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55 441 transgender population can reduce their vulnerability to HIV as demonstrated in other set-
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57 442 tings.⁴¹ Sustaining and increasing the coverage of internet and peer-led community-based ser-
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59 443 vices for this population is particularly important, given that they are often unable to access
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3 444 health services due to stigmatization and discrimination,^{16 40} even in health care settings.¹⁸ As in
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5 445 other settings globally,⁹ only half of those who were infected with HIV were aware of their sta-
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7 446 tus, demonstrating the need to intensify HIV testing, including self-testing at the community
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9 447 level, which transgender women in Cambodia are willing to use.⁴² Our study also found that
10
11 448 once diagnosed, the coverage of ART was high in this population.
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13 449

14 450 **LIMITATIONS OF THE STUDY**

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16 451 Our study covered only 13 city and provinces with the most numbers of transgender women,
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18 452 leaving out the other venues with fewer participants, and thus may not be generalized to all
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20 453 transgender women nationally. Second, the initial seeds of participants were identified and re-
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22 454 cruited by outreach workers of community-based organizations, which could introduce bias to-
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24 455 wards transgender women under their programs, leading to a recruitment bias. This problem
25
26 456 could be exacerbated by outreach workers interviewing some participants who had received
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28 457 services from their NGO, which could induce the participants' responses. Third, this study em-
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30 458 ployed a self-reporting questionnaire on sensitive health and sexual behaviors, which may have
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32 459 been limited by social desirability bias. Fourth, albeit minimal, the monetary incentive given to
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34 460 the participants to recruit seeds may have affected their genuine motivation to partake in the
35
36 461 study, which could influence their responses. Finally, as this study was cross-sectional, it reports
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38 462 associations at a given time, and may not be construed to be reporting causal relationships.
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40 463

41 464 **CONCLUSIONS**

42
43 465 This study demonstrates that HIV prevalence among transgender women in Cambodia remains
44
45 466 persistently high. With the prevalence rate at 5.9%, higher prevalence was observed among
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47 467 transgender women who resided in urban areas, of older age, with low education levels, with
48
49 468 previous STI symptoms, with a history of self-injected hormone, and those who had never used
50
51 469 online services developed for transgender women or MSM. Therefore, to eliminate HIV infec-
52
53 470 tion among transgender women, tailored interventions need to focus on these sub-populations
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55 471 and attendant risk factors.
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2
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4
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12
13 478 sibility of the authors and does not reflect the view of USAID or our respective institutions.
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16
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18
19 481 tools. SC, CN and SY analyzed the data, interpreted the results and wrote the manuscript. PC,
20
21 482 SC, CL, KP, ST and PM were responsible for training and data collection. JM, JD and GB support-
22
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24
25 484 script.
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27 485

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31

32 488

33 489 **Competing interests** None declared.
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35 490

37 491 **Patient consent** Obtained
38

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41 493 **Ethics approval** The National Ethics Committee for Health Research, Ministry of Health, Cam-
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43 494 bodia.
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47 496 **Consent for publication**

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49 497 Consent to publish was obtained from participants as stated in the informed consent form.
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52 499 **Data sharing statement** Data used for this study cannot be made available in the manuscript,
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54 500 the supplemental files, or a public repository due to the ethical restriction stated in the agree-
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3 501 ment with the National Ethics Committee for Health Research. However, they can be accessed
4
5 502 upon request from the Principal Investigator (Dr. Siyan Yi) at siyan@doctor.com.

6
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For peer review only

Participant ID Code:

Integrated Biological and Behavioral Survey of Transgender Women in Cambodia, 2016

Date of interview: day.....month.....year.....

Province/City:

- | | | |
|---------------------|---------------------|------------------|
| 1. Phnom Pehn | 6. Preah Sihanouk | 11. Kandal |
| 2. Kampong Cham | 7. Prey Veng | 12. Koh Kong |
| 3. Battambang | 8. Svay Rieng | 13. Thbong Khmum |
| 4. Siem Reap | 9. Kampong Spue | |
| 5. Banteay Meanchey | 10. Kampong Chhnang | |

Interview Location (City/NGOs):

Name of administrative districts (current residence):

Status of administrative districts:

- 1- Urban
- 2- Rural

SECTION 1: SOCIODEMOGRAPHIC CHARACTERISTICS			
No.	Questions and filters	Coding categories	Skip to
Now I would like to ask you some questions related to your personal information.			
Q001	How old are you?	Age in completed years:	
Q002	What is your current marital status? (only one response)	Married and living together 1 Married ,but not living together 2 Widowed, Divorced or separated 3 Not married, not living with any partner 4 Not married, living with sweetheart (female) 5 Not married but living with male lover 6 Other (Specify.....) 7	
Q003	For how long have you been living in the current city?Years <u>Record 0</u> if not living in this city <u>Record 1</u> if living in this city 1 year or less Refuse to answer 99	
Q004	In the past 6 months, on average, how much money did you make per month?	Amount of money (in US\$): Refuse to answer 99	

Questionnaire; Integrated Biological and Behavioral Survey among Transgender Population in Cambodia, 2015, Version 2.0, 17 September 2015 Page 1 of 11

	(exchange rate: 4000 riel per 1 US\$)		
Q005	How many years of formal education have you completed?	Number of years: <u>Record 0</u> if never attending school. Refuse to answer 99	
Q006	What is your current job (main source of income)? (only one response) <i>Note: If you are both studying and having a paid job, report your main source of income.</i>	Unemployed 0 Hair dresser/Beautician 1 Government officer 2 Laborer (factory, construction work) 3 Seller 4 Entertainment Worker (bar, karaoke, etc.) 5 Sex worker 6 Student 7 NGO staff 8 Private company staff 9 Farmer/fisherman 10 Artist 11 Other (Specify.....) 12	

SECTION 2. TRANSGENDER IDENTITY & EXPERIENCES			
Q007	What gender do you consider yourself?	Female 1 Male 2 Third gender 3 Uncertain 4 Refuse to answer 99	
Q008	How often do you express or dress yourself as a woman?	All the time 1 Often 2 Sometimes 3 Rarely 4 Refuse to answer 99	
Q009	How many friends in your social network are also transgender?	Number:	
Q010	Have you ever taken hormones or any substances for beauty purposes?	No 1 Yes 2 Refuse to answer 99	2→ Q014
Q011	What are the methods you have used to take hormones or any substances for beauty purposes? [multiple answer]	Pills 1 Injections 2 Skin patches 3 Other (Specify.....) 4 Refuse to answer 99	
Q012	If you have taken hormones or any substances for beauty purposes by injection, how	Never injected 0 Injected by myself 1 Injected by skilled personnel (beauty clinic) 2	

	have you received it?	Injected by non-skilled personnel 3 Other (Specify.....) 4	
Q013	If you have injected hormones or any substances for beauty purposes, have you shared needle?	Never injected 0 No 1 Yes 2 Refuse to answer 99	
Q014	Have you ever had any operation to change any parts of your body to become a woman?	No 1 Yes 2 Refuse to answer 99	

Section 3. SEXUAL PARTNERS AND SEXUAL HISTORY			
No.	Questions and filters	Coding categories	Skip to
	Now I would like to ask questions about your recent sexual relationship with various kinds of sexual partners.		
Q015	Have you ever had sex with any partner in your life time? [Including vaginal, or anal sex]	No 0 Yes 1 Refuse to answer 99	
Q016	How likely do you think you are infected with HIV?	Very Likely 1 Likely 2 Unlikely 3 Very Unlikely 4 Refuse to answer 99	
Sex with women			
Q017	Have you ever had sex with a woman? [Including vaginal or anal sex]	No 0 Yes 1 Refuse to answer 99	0, 99 → Q028
Q018	In the past 12 months, have you had sex with a woman? [Including vaginal or anal sex]	No 0 Yes 1 Refuse to answer 99	0, 99 → Q028
Q019	In the past 12 months, have you had sex with a woman not in exchange for money or gifts? [Including vaginal or anal sex]	No 0 Yes 1 Refuse to answer 99	0→ Q022
Q020	In the past 3 month, how many women did you have sex with not in exchange for money or gifts?	Number of women: Refuse to answer 99	
Q021	In the past 3 month, how often did you use a condom when you had vaginal or anal	Never had sex with a woman 0 Always 1 Often 2	0, 1→ Q024

Questionnaire; Integrated Biological and Behavioral Survey among Transgender Population in Cambodia, 2015, Version 2.0, 17 September 2015 Page 3 of 11

	sex with women not in exchange for money or gifts?	Sometimes 3 Never 4 Refuse to answer 99	
Q022	In the past 3 month, if you did not always use a condom when you had sex with women not in exchange for money or gifts? What were the reasons? [Multiple Answers]	We are in a relationship 1 She is not HIV/STI infected 2 Too high to use a condom 3 No condom available 4 Feel better without a condom 5 I am HIV-infected 6 I penetrated, so I am not at risk 7 Partner refused 8 Other (Specify.....) 9 Refuse to answer 99	
Q023	In the past 12 months, have you had sex with a woman in exchange for money or gifts? [Including vaginal or anal sex]	No 0 Yes 1 Refuse to answer 99	0→ Q028
Q024	In the past 3 months, how many women did you have sex with in exchange for money or gifts? [Including vaginal or anal sex]	Number of women: Refuse to answer 99	
Q025	Where did you meet women with whom you had sex with in exchange for money or gifts??	Park 1 Street 2 Bar/discotheque/cafe 3 Beer Garden/Restaurant 4 Massage Parlor 5 Karaoke 6 Online (Facebook, Line, Website) 7 Guesthouse/hotel 8 Workplace/school 9 Other (Specify.....) 10	
Q026	In the past 12 months, how often did you use a condom when you had vaginal or anal sex with women in exchange for money or gifts?	Never had sex 0 Always 1 Often 2 Sometimes 3 Never 4 Refuse to answer 99	
Q027	In the past 3 months, if you did not always use a condom when you had sex with women not in exchange for money or gifts? What were the reasons? [Multiple Answers]	We are in a relationship 1 She is not HIV/STI infected 2 Too high to use a condom 3 No condom available 4 Feel better without a condom 5 I am HIV-infected 6 I penetrated, so I am not at risk 7 Partner refused 8 Other (Specify.....) 9	

		Refuse to answer 99	
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Sex with men			
Q028	Have you ever had anal sex with a man?	No 0 Yes 1 Refuse to answer 99	0, 99→ Q042
Q029	In the past 12 months, have you had anal sex with a man?	No 0 Yes 1 Refuse to answer 99	0, 99→ Q042
Q030	In the past 12 months, what was your usual role in anal sex with a man?	Insertive 1 Receptive 2 Both 3 Refuse to answer 99	
Q031	In the past 3 months, have you had sex with men not in exchange for money or gifts?	No 0 Yes 1 Refuse to answer 99	0→ Q033
Q032	In the past 3 months, how many men you had sex with not in exchange for money or gifts?	Number of men: Refuse to answer 99	
Q033	In the past 12 months, how often did you use a condom when you had anal sex with men not in exchange for money or gifts?	Never had sex 0 Always 1 Often 2 Sometimes 3 Never 4 Refuse to answer 99	0, 1→ Q035
Q034	If not always, why not? [Multiple Answers]	We are in a relationship 1 He is not HIV/STI infected 2 I was too drunk or too high with drugs 3 No condom available 4 Feel better without a condom 5 I am HIV-infected 6 I penetrated, so I am not at risk 7 Partner refused 8 Other (Specify.....) 9 Refuse to answer 99	
Q035	In the past 12 months, have you had sex with men in exchange for money or gifts?	No 0 Yes 1 Refuse to answer 99	0→ Q040
Q036	In the past 3 month, how many men you had sex with in exchange for money or	Number of men: Refuse to answer 99	

	gifts?		
Q037	Where did you meet the men you had sex with in exchange for money or gifts?	Park 1 Street 2 Bar/discotheque/cafe 3 Beer Garden/Restaurant 4 Massage Parlor 5 Karaoke 6 Online (Facebook, Line, Website) 7 Guesthouse/hotel 8 Workplace/School 9 Other (Specify.....) 10	
Q038	In the past 12 months, how often did you use a condom when you had anal sex with men you paid for sex?	Never had anal sex 0 Always 1 Often 2 Sometimes 3 Never 4 Refuse to answer 99	0, 1→ Q615
Q039	If not always, why not? [Multiple Answers]	We are in a relationship 1 He is not HIV/STI infected 2 I was drunk or too high with drugs 3 No condom available 4 Feel better without a condom 5 I am HIV-infected 6 I penetrated, so I am not at risk 7 Partner refused 8 Other (Specify.....) 10 Refuse to answer 99	

Access to condoms and lubricant			
No.	Questions and filters	Coding categories	Skip to
Q040	In this last 12 months, where did you get condoms? [Multiple Answers]	Friends/outreach workers 1 Condom peer sale representative 2 Pharmacy/drug store/clinic 3 Condom outlets 4 Mart/mini-mart 5 Groceries 6 Hotel/guesthouse 7 Other (Specify.....) 8 Refuse to answer 99	
Q041	In this last 12 months, where did you get lubricant?	Friends/outreach workers 1 Condom peer sale representative 2 Pharmacy/drug store/clinic 3	

	[Multiple Answers]	Condom outlets 4 Mart/mini-mart 5 Groceries 6 Hotel/guesthouse 7 Other (Specify.....) 8 Refuse to answer 99	
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SECTION 4. STI AND HIV TESTING			
No.	Questions and filters	Coding categories	Skip to
Q042	In the past 12 months, have you experienced the following symptoms? [Multiple Answers]	Cuts or sores in the genital area 1 Swelling in the genital area 2 Abnormal urethral discharge 3 Symptoms on the anus 4 Symptom in the mouth/throat 5 Refuse to answer 99	
Q043	Where did you first go for treatment the last time you had any above-mentioned symptoms? (Only one response)	Never had any symptoms 0 Pharmacy 1 Private clinic/hospital 2 Public Hospital/STD clinic 3 NGO Clinic/hospital 4 Traditional doctor 5 Didn't get care 6 Other (Specify.....) 7 Refuse to answer 99	
Q044	Have you ever been tested for HIV?	No 0 Yes 1 Refuse to answer 99	0→ Q049
Q045	In past 6 months, have you been tested for HIV?	No 0 Yes 1 Refuse to answer 99	
Q046	How long has it been since you received the last HIV test?	Number of months.....	
Q047	If a rapid self-test kit for HIV was available and you could get the result within 20 minutes at home, would you use it?	No 0 Yes 1 Refuse to answer 99	
Q048	Where did you have your most recent HIV test?	Private facilities 1 Public facilities 2 NGO facilities 3 NGO outreach workers at community or HTC 4 Other (Specify.....) 5 Refuse to answer 99	

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Q049	The last time you got tested for HIV, did you receive the result of the test?	No 0 Yes 1 Refuse to answer 99	
Q050	What is your current HIV status? (You could choose not to answer)	Positive 1 Negative 2 I don't know my status 3 Refuse to answer 99	2, 3 → Q057
Q051	Are you currently on ART?	No 0 Yes 1 I was but discontinued 2 I am on treatment for opportunistic infections 3 Refuse to answer 99	0 → Q056
Q052	Do you take ARV regularly as prescribed?	Regularly 1 Not regularly 2 Refuse to answer 99	
Q053	Where do you get ART services?	Public facilities 1 Private facilities 2 NGO facilities 3 Pharmacy 4 Other (Specify.....) 5 Refuse to answer 99	
Q054	If not, why are you not on ART?	Not needed 1 Not sure where to go 2 I am ashamed to go to health facility 3 I am afraid of being discriminated 4 I was treated badly in the past 5 Other (Specify.....) 6 Refuse to answer 99	

SECTION 5. ALCOHOL AND DRUG USE

Measurement of alcohol drinking

One standard measurement:

- A can/glass of beer, fermented palm juice (285 ml)
- A glass of wine (120 ml)
- A glass of whisky (30 ml)

No.	Questions and filters	Coding categories	Skip to
Q055	In the past 3 months, how often did you drink at least one can of beer or one glass of any types of wine?	Never 1 Once a month or less 2 2-4 times a month 3 2-3 times a week 4 4 or more times a week 5	1 → Q064

Questionnaire; Integrated Biological and Behavioral Survey among Transgender Population in Cambodia, 2015, Version 2.0, 17 September 2015 Page 8 of 11

		Don't know 98 Refuse to answer 99																																
Q056	In the past 3 months, how many standard drinks containing alcohol (a can of beer or a glass of any types of wine) did you have on a typical day on which you drank alcohol?	1 - 2 1 3 - 4 2 5 - 6 3 7 - 9 4 10 or more 5 Don't know 98 Refuse to answer 99																																
Q057	In the past 3 months, how often did you have more than 5 drinks in one day or night?	Never 1 Less than once a month 2 Once a month 3 1, 2, or 3 times a week 4 4 or more times a week 5 Don't know 98 Refuse to answer 99																																
Q058	In the past 3 months, how often have you got drunk from alcohol?	Never 1 Less than once a month 2 Once a month 3 Every week (1-3 times/week) 4 Every day/almost every day(4 or more times a week) 5 Don't know 98 Refuse to answer 99																																
Q059	<p>In the past 12 months, have you tried any of the following drugs? (Ask one by one – CIRCLE YES OR NO) [Multiple answers]</p> <table border="1"> <thead> <tr> <th></th> <th>Types of drug</th> <th>Yes</th> <th>No</th> </tr> </thead> <tbody> <tr> <td>1</td> <td>Marijuana</td> <td>1</td> <td>0</td> </tr> <tr> <td>2</td> <td>Heroin/Opium</td> <td>1</td> <td>0</td> </tr> <tr> <td>3</td> <td>Yama (amphetamine)</td> <td>1</td> <td>0</td> </tr> <tr> <td>4</td> <td>Crystal, Ice (Methamphetamine)</td> <td>1</td> <td>0</td> </tr> <tr> <td>5</td> <td>Ecstasy</td> <td>1</td> <td>0</td> </tr> <tr> <td>6</td> <td>Inhalants (glue, paint, petrol, spray can)</td> <td>1</td> <td>0</td> </tr> <tr> <td>7</td> <td>Other</td> <td>1</td> <td>0</td> </tr> </tbody> </table>			Types of drug	Yes	No	1	Marijuana	1	0	2	Heroin/Opium	1	0	3	Yama (amphetamine)	1	0	4	Crystal, Ice (Methamphetamine)	1	0	5	Ecstasy	1	0	6	Inhalants (glue, paint, petrol, spray can)	1	0	7	Other	1	0
	Types of drug	Yes	No																															
1	Marijuana	1	0																															
2	Heroin/Opium	1	0																															
3	Yama (amphetamine)	1	0																															
4	Crystal, Ice (Methamphetamine)	1	0																															
5	Ecstasy	1	0																															
6	Inhalants (glue, paint, petrol, spray can)	1	0																															
7	Other	1	0																															

Q060	In the past 3 months, did you inject any illicit drugs?	Heroin 1 Yama 2 Crystal, Ice (Methamphetamine) 3 Never injected any drug 4 Refuse to answer 99
Q061	In the past 3 months, did you have sex during/after using illicit drugs?	Heroin 1 Yama 2 Ice, Amphetamine 3 Never used drug before having sex 4 Refuse to answer 99

SECTION 9. EXPOSURE TO INTERVENTION PROGRAMS			
No.	Questions and filters	Coding categories	Skip to
Q062	In the past 3 months, have you been reached by NGO staff delivering HIV services?	No 0 Yes 1 Don't know 98 Refuse to answer 99	
Q063	In the past 3 months, what kind of services have you received (Check all that apply).	HIV/health education and/or materials 1 Condoms 2 Lubricants 3 VCCT/STI testing 4 Law support services 5 Other health services 6 Other (Specify.....) 7 Refuse to answer 99	
Q064	Do you have UIC card?	Yes 1 No 0 Refuse to answer 99	
Q065	In the past 12 months, how often did you visit MStyle/Srey Sros club(s)?	... times Refuse to answer 99	
Q066	Have you used online services developed for people like you (e.g. Facebook, website)?	No 0 Yes 1 Refuse to answer 99	0 -> stop interview
Q067	Have you found any sexual partner on an Internet website or any online program?	Never 0 Yes, always 1 Yes, sometimes 2 Yes, occasionally 3	

		Refuse to answer 99	
Q068	Have you accessed MStyle website in the past 6 months? (Attach the picture of MStyle webpage)	Never 0 Yes, always 1 Yes, sometimes 2 Yes, occasionally 3 Refuse to answer 99	

["Thank you very much for answering these questions. Let us repeat that your answers are totally confidential and there is no way anyone will learn what you told us.]

For peer review only

BMJ Open

HIV prevalence and factors associated with HIV infection among transgender women in Cambodia: Results from a national integrated biological and behavioral survey.

Journal:	<i>BMJ Open</i>
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Primary Subject Heading:	HIV/AIDS
Secondary Subject Heading:	Infectious diseases, Epidemiology, Public health
Keywords:	Transgender women, HIV infection, Risk factors, Prevention, Integrated Biological and Behavioral Survey, Cambodia

SCHOLARONE™
Manuscripts

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3 1 **HIV prevalence and factors associated with HIV infection among transgender women in Cam-**
4 **odia: Results from a national integrated biological and behavioral survey**
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9 4 Chhim Srean,¹ Chanrith Ngin,² Pheak Chhoun,² Sovannary Tuot,² Cheaty Ly,³ Phalkun Mun,⁴
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1
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3 30 **ABSTRACT**

4
5 31 **Objective:** To examine factors associated with HIV infection among transgender women in
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7 32 Cambodia.

8
9 33 **Design:** Cross-sectional study

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11 34 **Settings:** HIV high-burden sites including the capital city and 12 provinces

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13 35 **Participants:** This study included 1,375 sexually-active transgender women with a mean age of
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15 36 25.9 years (SD= 7.1), recruited by using respondent driven sampling for structured question-
16
17 37 naire interviews and rapid finger-prick HIV testing.

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19 38 **Primary outcome measure:** HIV infection detected by using Determine™ antibody test.

20
21 39 **Results:** HIV prevalence among this population was 5.9%. After adjustment for other covariates,
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23 40 participants living in urban areas were twice as likely to be HIV infected as those living in rural
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25 41 areas. Participants with primary education were 1.7 times as likely to be infected compared to
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27 42 those with high school education. HIV infection increased with age; compared to those aged 18-
28
29 43 24, the odds of being HIV infected were twice as high among transgender women aged 25-34
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31 44 and 2.8 times higher among those aged ≥ 35 . Self-injection of gender affirming hormones was
32
33 45 associated with a four-fold increase in the odds of HIV infection. A history of genital sores over
34
35 46 the previous 12 months increased the odds of HIV infection by three-fold. Transgender women
36
37 47 with stronger feminine identity, dressing as a woman all the time were twice as likely to be HIV
38
39 48 infected compared to those who did not dress as a woman all the time. Having never used
40
41 49 online services developed for transgender women in the past six months was also associated
42
43 50 with higher odds of being HIV infected.

44
45 51 **Conclusions :** Transgender women in Cambodia are at high risk of HIV. To achieve the goal of
46
47 52 eliminating HIV in Cambodia, effective combination prevention strategies addressing the above
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49 53 risk factors among transgender women should be strengthened.

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52 55 **Key words:** Transgender women, HIV infection, Risk factors, Prevention, Integrated Biological
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54 56 and Behavioral Survey, Cambodia.

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3 59 **Strengths and limitations of this study**
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- 7 61 ▪ This is a rare and the second ever Integrated Biological and Behavioral Survey (IBBS)
8 among transgender women—one of the most vulnerable and under-studied populations
9 62 in Cambodia and globally.
10 63
11 64 ▪ This study covered HIV high-burden sites including the capital city and 12 provinces,
12 thus providing more nationally generalizable results.
13 65
14 66 ▪ This study identified important risk factors for HIV infection among transgender women,
15 which require being addressed in order to reduce HIV infection among this key popula-
16 67 tion.
17 68
18 69 ▪ Limitations of the study included potential bias of self-reported measures, participant
19 70 sampling, the cross-sectional nature of the data that limits causation inferences, and the
20 71 backward selection of variables in multivariable analysis.
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75 INTRODUCTION

76 Globally, transgender women are at high risk for HIV infection, and little is known about the
77 burden of HIV infection and its related factors in this population. A 2013 systematic review
78 showed that the global HIV prevalence among transgender women was 19%, with low- and
79 middle-income countries having a slightly lower prevalence of 18% compared to 22% in high-
80 income countries.¹ The probability of being infected with HIV among transgender women was
81 approximately 49 times higher than among the general adult population.¹ Epidemiologically,
82 transgender women share some HIV risk factors with other populations, such as ulcerative sex-
83 ually transmitted infections (STIs),^{2,3} multiple substance use, victimization, intimate partner vio-
84 lence,⁴ unprotected sex,⁵ multiple sex partners,⁶ and injecting drug use.⁵⁻⁷

85 However, transgender women encounter additional and unique biological, social, cul-
86 tural, legal and economic issues, which increase their vulnerability to HIV. Transgender women
87 are marginalized legally and economically through discriminative legislation and practice, which
88 accelerates their vulnerability to HIV infection by confining their job options to sex work. Sex
89 work in turn exacerbates their stigmatization and alienation,⁸ and further increases their HIV
90 risk.^{9,10} Stigmatization and discrimination further increase the risk of HIV infection by prevent-
91 ing transgender women from seeking essential health information and services.¹¹ Depression
92 and low self-esteem, which are common psychological consequences of stigma, further weak-
93 ens the bargaining power for condom use in sexual relationships, resulting in heightened vul-
94 nerability to HIV infection.^{5,12-15} Transgender women who have sex with men often engage in
95 receptive anal intercourse, which increases their risk of HIV acquisition, compared to insertive
96 sexual partners.² Syndemics of illicit drug use, abuse by family members, and mental health of-
97 ten occur together;¹⁶ and are often associated with less likelihood of transgender women to use
98 condoms during sex with commercial partners.¹⁷ In addition, the confluence of arbitrary ar-
99 rests,^{18,19} violence,^{16,20} economic pressures,²¹ and other social problems further aggravate
100 mental health risks among transgender women, causing them to engage in risky sexual behav-
101 iors.^{6,17}

102 In Southeast Asia, a systematic review of studies from Indonesia, Thailand and Vietnam
103 demonstrated that transgender women are particularly at high HIV risk, with an estimated HIV

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3 104 prevalence of 26.1%, 12.2% and 6.7%, respectively.¹ In Cambodia, up to 2012, transgender
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5 105 women were traditionally included in HIV prevention programs targeting men who have sex
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7 106 with men (MSM). As a result, their unique needs were not reflected in HIV programs' goals.²²
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9 107 There was also a lack of transgender women-specific data to quantify the number reached by
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11 108 outreach programs and services. In 2012, the first Integrated Biological and Behavioral Survey
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13 109 (IBBS) that specifically included transgender women in Cambodia found an HIV prevalence of
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15 110 4.2% among this population,²² which was six times higher than the 0.7% prevalence among the
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17 111 general adult population aged 15-45,²³ and about twice higher than the 2.1% among MSM.²⁴
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19 112 This 2012 study also found that older age (>35 years), residing in Siem Reap province (a major
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21 113 tourist site), having sex during or after drug use, not using a condom during the last anal sex,
22
23 114 and low self-esteem were associated with HIV infection among transgender women.²²

24 115 Recognizing the differences in HIV vulnerability and prevention needs between
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26 116 transgender women and MSM, in 2013 the National Center for HIV/AIDS, Dermatology and STD
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28 117 (NCHADS) separated these two populations in its revised Standard Operating Procedure (SOP)
29
30 118 known as Boosted Continuum of Prevention, Care and Treatment (B-CoPCT).²⁵ Transgender
31
32 119 women have since been considered a distinct at-risk group for HIV surveillance and programing.
33
34 120 The National B-CoPCT approach aims to increase the uptake of HIV testing and counseling (HTC)
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36 121 and other related services, ensure the quality of outreach and strengthen service delivery to
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38 122 meet specific needs of key populations, including transgender women, in-order to achieve
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40 123 Cambodia's 3.0 goal (i.e. zero new HIV infection, zero discrimination, and zero AIDS-related
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42 124 deaths) by 2020.

43 125 Three years following the first IBBS in 2012,²² the TGIBBS-2016 was conducted between
44
45 126 December 2015 and February 2016 to explore the HIV prevalence and identify risk factors asso-
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47 127 ciated with HIV infection among transgender women in Cambodia. The study findings will help
48
49 128 determine ways in which services and policies can be tailored for this key population.

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52 130 **METHODS**

54 131 **Study sites**

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3 132 Data collection was conducted in the capital city of Phnom Penh and 12 provinces namely Bat-
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5 133 tambang, Banteay Meanchey, Kampong Cham, Kandal, Koh Kong, Kampong Chhnang, Kampong
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7 134 Speu, Prey Veng, Preah Sihanouk, Siem Reap, Svay Rieng and Tbong Khmum. These sites were
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9 135 purposively selected from the 23 HIV high-burden operational districts identified by NCHADS.²⁶
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138 **Sample size and sampling procedures**

139 The sample size calculation was based on an assumption that HIV prevalence would have
140 changed between the first and the current IBBS. The minimum sample size required for this
141 study was 1,380, which would result in sufficient power to detect a priori significant difference
142 in HIV prevalence, as expressed in the following null and alternative hypotheses. The hypothesis
143 test was that of one-sample proportion in comparison to the null hypothesis.

144 The null hypothesis (H_0): HIV prevalence among transgender women remains constant at
145 4.2%, similar to that found in the 2012 IBBS.

146 The alternative hypothesis (H_a): HIV prevalence among transgender women has decreased
147 by 1.2% from 4.2% in 2012 to 3.0% in the present IBBS.

148

149 The sample size was calculated using the following formula and assumptions:

$$n = \frac{p_0q_0 \left(z_{1-\alpha} + z_{1-\beta} \sqrt{p_1q_1/p_0q_0} \right)^2}{(p_1 - p_0)^2}$$

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151 ▪ P_0 = estimated proportion in H_0 (In the most recent survey, HIV prevalence among TG
152 women was 4.2% in 2012.)

153 ▪ P_1 = estimated proportion in H_a (The expected HIV prevalence among TG women in the
154 current study is about 3.0 %)

155 ▪ $Z_{(1-\alpha)}$ = significance level at 5% in response to one-sided test (Z score = 1.645)

156 ▪ $Z(1-\beta)$ = power level of 80% (Z score = 0.83)

157 ▪ Refusal rate of 10%

158 ▪ Design effect is assumed to be one.

159

This sample size was stratified by study site. Roughly half of the estimated transgender women in each study site were recruited. However, in five provinces where the estimated number of transgender women was smaller than 100, all transgender women were recruited. Potential participants were included in the study if they: (1) were biologically male at birth and self-identified as a woman or third gender, (2) were Khmer-speaking, (3) were at least 18 years old at the time of screening, (4) reported having had sex with at least one man within the past 12 months and (5) were able and willing to provide an informed consent.

Among the 13 study sites, participant recruitment was conducted in 20 locations (six locations in Phnom Penh and 14 locations in the remaining provinces). The number of the selected locations was determined based on the required sample size in each study site. Participants were recruited through a Respondent Driven Sampling (RDS) method. First, four “seeds” (two seeds aged 18 to 24 and two seeds 25 or older) who were well connected with other transgender women in each location were selected by outreach workers from implementing partners (local NGOs) based in the selected locations. These seeds had to meet the above-mentioned eligibility criteria for participation and have an established and large social network consisting of 10 or more other transgender women in their given location. Eligibility to participate as a seed was determined by the leader of the data collection team, using a paper-based eligibility form.

Second, each seed was given a personal identity number (PIN) and enrolled in the study. Third, each seed was given three coupons and asked to refer three additional transgender women. US\$2 was given to each seed for a successful referral. Each seed was expected to extend to three to six “recruitment waves” in each location. If the initial seeds did not recruit participants, or if enrollment was halted because all recruitment chains had “dried up” (i.e. stopped recruiting), additional seeds were selected. In total, 80 seeds were selected by the outreach workers, and a total of 1,375 transgender women were enrolled in the study. Referred participants were initially screened by the data collectors for eligibility.

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187 **Data collection training and procedures**

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3 188 Data were collected by three teams; each team comprised of one field supervisor, five inter-
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5 189 viewers, one lab technician and one counselor. Lab technicians and counselors were from the
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7 190 Municipal or Provincial AIDS and STI Program of the study sites. Data collection teams were
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9 191 trained for three days on study protocol, research ethics, interview techniques and data collec-
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11 192 tion procedures provided by the principle investigators and research coordinators.

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13 193 To determine HIV prevalence, all participants received onsite rapid finger-prick testing.
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15 194 Pre-test counseling was provided by qualified, well-trained counselors working for voluntary
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17 195 confidential counseling and testing (VCCT) centers. Participants could receive their HIV-test re-
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19 196 sult verbally after the questionnaire interview. A blood sample was obtained from each partici-
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21 197 pant by a trained laboratory technician through finger-prick and tested for HIV using Deter-
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23 198 mine™ test, in keeping with the national protocol.²⁸ Post-test counseling was provided for each
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25 199 participant regardless of their HIV test result by the same counselor who conducted the pre-
26
27 200 test counseling in accordance with NCHADS HIV testing guidelines.²⁸ Participants who were HIV
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29 201 reactive and did not know their HIV status were referred by the counselor for confirmatory
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31 202 testing at the nearest VCCT center. The quality of HIV tests was monitored using quality control
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33 203 samples. Discordant results between the screening and confirmatory tests were investigated to
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35 204 elucidate potential causes and minimize potential erroneous results. In the case of human er-
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37 205 ror, individuals performing screening tests were retrained before resuming their involvement in
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39 206 the study.

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41 207 After HIV testing, the participant was interviewed by a well-trained interviewer using a
42
43 208 computer-assisted survey instrument (CASI). The questionnaire was set up in Qualtrics, a web-
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45 209 based application, and run on an Android tablet. HIV testing and interviews were conducted in
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47 210 the selected 20 locations including drop-in centers, private houses and offices of implementing
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49 211 partners, depending on participants' convenience.

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213 **Questionnaire development and measures**

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53 214 The questionnaire was developed using standardized and validated tools adapted from TGIBBS-
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55 215 2012,²² FHI 360's guideline for Behavioral Surveillance Survey (BSS)²⁹ and NCHADS's Boosted-
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57 216 CoPCT²⁵ to measure key variables related to the objectives of the study. The questionnaire was
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3 217 initially drafted in English and translated into Khmer, the national language of Cambodia. Con-
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5 218 sultative meetings to improve the draft questionnaire were held with representatives of
6
7 219 transgender women, communities and NGOs working with transgender women, as well as re-
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9 220 searchers and practitioners working on HIV and AIDS in Cambodia. The questionnaire was pre-
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11 221 tested with 20 transgender women in Phnom Penh and finalized based on findings.

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13 222 The questionnaire collected information on socio-economic characteristics, transgender
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15 223 identity and related experiences, sexual behaviors and condom use with different types of sex-
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17 224 ual partners, HIV/STI screening and care seeking behaviors, substance use and exposure to HIV
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19 225 programs. For details of the questionnaire, please see Supplementary File 1.

20 226

21 227 **Data analyses**

22 228 HIV prevalence was calculated by dividing the total number of participants with HIV reactive
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24 229 test results with the total number of participants. To examine the associated factors of HIV in-
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26 230 fection, we conducted both bivariate and multivariable analyses. In the bivariate analyses, we
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28 231 compared all characteristics and behavioral variables of participants with a reactive test result
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30 232 to those of participants with a non-reactive test result. Among participants who tested HIV pos-
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32 233 itive, additional analyses were conducted to assess whether there was a significant difference in
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34 234 sexual behaviors of those who were aware of their HIV-positive status and of those who were
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36 235 not. Chi-square test or Fisher's exact test (for an expected cell value of ≤ 5) was used for cate-
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38 236 gorical variables, while Student's *t*-test was used for continuous variables. To facilitate the
39
40 237 model, some continuous variables, such as age were transformed to categorical variables. A
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42 238 multivariable logistic regression model was constructed to examine independent factors associ-
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44 239 ated with HIV infection. Variables with a significance level of $p < 0.05$ in the bivariate analyses
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46 240 were simultaneously included in the model. Backward elimination method was then used to
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48 241 eliminate variables with the highest *p*-value one-by-one from the model. STATA Version 12.0
49
50 242 for Windows (Stata Corp, TX, USA) was used to conduct the data analyses.

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52 244 **ETHICAL STATEMENT**

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3 245 Participation in this study was voluntary, and a written informed consent was obtained from
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5 246 each study participant after a detailed description of the study objective and procedures was
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7 247 explained to them. Participants were informed that they could stop responding to questions
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9 248 and discontinue their participation at any time. Interviews were conducted at a private place,
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11 249 and confidentiality was enhanced by assigning a unique and anonymous code to each partici-
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13 250 pant. No personal identifiers were contained in the questionnaires or dataset. The study proto-
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15 251 col was approved by the National Ethics Committee for Health Research (NECHR) of the Minis-
16
17 252 try of Health, Cambodia (No. 420 NECHR) and FHI 360's Protection of Human Subjects Commit-
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19 253 tee (PHSC No. 713897).

20 254

21 255 **RESULTS**

22 256 **HIV prevalence**

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26 257 Of 1,375 participants tested, 81 (5.9%) had an HIV reactive test; of whom, 42 (52%) were not
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28 258 aware of their HIV status prior to the study. Out of the 39 cases who already knew their status,
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30 259 37 (94.9%) were currently on HIV treatment, with the other two reporting having dropped out
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32 260 of the treatment.

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34 262 **Socio-demographic characteristics**

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37 263 As shown in Table 1, the majority of the participants (83.4%) were recruited from urban com-
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39 264 munities; 53.0% were younger than 25 years old and 97.2% had never been married. More than
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41 265 two-thirds of participants (68.6%) had completed high school, and 9.1% had a higher education.
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43 266 The most common main occupations reported by the participants were hair dress-
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45 267 ers/beauticians (35.1%), laborers/farmers (17.5%) and entertainment workers (14.8%). More
46
47 268 than one-third of participants (38.6%) reported an average monthly income in the past six
48
49 269 months of US\$ 100-199, while 16.5% reported it to be more than US\$ 300. Regarding gender
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51 270 identity, 42.2% identified themselves as female, while 57.2% identified themselves as third
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53 271 gender. Almost half (48.0%) of the participants reported dressing as a woman all the time; 45%
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55 272 ever used hormone/non-hormone substance; and 14.0% ever injected hormones.

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273 HIV prevalence was significantly higher among participants living in urban communities
 274 compared to participants living in rural communities (6.5% vs. 2.6%; $p= 0.02$). The HIV preva-
 275 lence was also significantly different by age group: 3.0% of participants in the age group of 18-
 276 24, 8.2% in 25-34, 13.1% in 35-44 and 11.4% in 45 or older ($p< 0.001$). HIV prevalence was also
 277 significantly higher among those who never attended school or dropped out of primary school
 278 when compared to those who at least attended high school or higher (10.4% vs. 8.0%; $p=$
 279 <0.001). In addition, HIV prevalence was significantly higher among transgender women who
 280 were NGO staff (mainly HIV focused NGOs) (20.6%) and those who were unemployed (9.4%) ($p=$
 281 0.03). HIV prevalence was significantly higher among transgender women who reported dress-
 282 ing as women all the time (8.5% vs. 3.5%, $p= 0.001$), among those who ever used female hor-
 283 mones (8.1% vs. 4.1%, $p= 0.02$) and among those who ever injected hormones (9.1% vs. 5.2%,
 284 $p= 0.002$). Moreover, HIV prevalence was significantly higher among transgender women who
 285 reported never using online services developed specifically for MSM or transgender women,
 286 such as Facebook group pages or various websites (9.5% vs. 3.9%, $p< 0.001$).

287

288 **Table 1** Comparison of socio-demographic, gender identity and hormone use characteristics of
 289 transgender women with and without a reactive HIV test

Socio-demographics, gender identity and hormone use	Total (<i>n</i> = 1375)	HIV test result		<i>P</i> -value*
		Reactive (<i>n</i> = 81)	Non-reactive (<i>n</i> = 1294)	
	<i>n</i> (%)	<i>n</i> (%)	<i>n</i> (%)	
Community type				
Urban	1146 (83.4)	75 (6.5)	1071 (93.5)	0.02
Rural	229 (16.6)	6 (2.6)	223 (97.4)	
Age in years				
18- 24	729 (53.0)	22 (3.0)	707 (97.0)	<0.001

25-34	503 (36.6)	41 (8.2)	462 (91.8)	
≥35	143 (10.4)	18 (12.6)	125 (87.4)	
Current marital status				
Married	7 (0.5)	0 (0.0)	7 (100)	0.47
Widowed/divorced/separated	18 (1.3)	1 (5.6)	17 (94.4)	
Never married	1334 (97.2)	78 (5.9)	1256 (94.1)	
Refused to answer	16 (1.2)	2 (15.4)	11 (84.6)	
Years of formal education completed				
Primary (0-6 years)	307 (22.3)	32 (10.4)	275 (89.6)	<0.001
High school or higher (>7 years)	1068 (77.7)	49 (4.6)	1019 (95.4)	
Main occupation				
Unemployed	64 (4.7)	6 (9.4)	58 (90.6)	0.03
Hair dresser/beautician	482 (35.1)	24 (5.0)	458 (95.0)	
Office worker	50 (3.6)	3 (6.0)	47 (94.0)	
Labor/farmer	241 (17.5)	15 (6.2)	226 (93.8)	
Seller	149 (10.8)	10 (6.7)	139 (93.3)	
Entertainment Worker	203 (14.8)	10 (4.9)	193 (95.1)	
Student	108 (7.9)	1 (0.9)	107 (99.1)	
NGO staff	34 (2.5)	7 (20.6)	27 (79.4)	
Other	44 (3.2)	5 (11.4)	39 (88.6)	
Monthly income in the past 6 months (USD)				

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4	< 100	351 (25.6)	22 (6.3)	329 (93.7)	0.70
5					
6	100-199	530 (38.6)	29 (5.5)	501 (94.5)	
7					
8					
9	200-299	266 (19.3)	19 (7.1)	247 (92.9)	
10					
11	≥300	226 (16.5)	11 (4.9)	215 (95.1)	
12					
13					
14	Gender identity (self-identified)				
15					
16	Female	580 (42.2)	29 (5.0)	551 (95.0)	0.35
17					
18	Third gender	786 (57.2)	52 (6.6)	734 (93.4)	
19					
20	Uncertain	8 (0.6)	0 (0.0)	8 (100)	
21					
22					
23					
24	Frequency of dressing as a woman				
25					
26	All the time	660 (48.0)	56 (8.5)	604 (91.5)	0.001
27					
28	Not all the time	715 (52.0)	25 (3.5)	689 (96.5)	
29					
30					
31					
32	Ever injected hormones				
33					
34	No	1123 (81.7)	58 (5.2)	1065 (94.8)	0.02
35					
36	Yes	252 (18.3)	23 (9.1)	229 (90.9)	
37					
38					
39	Ever self-injected hormones				
40					
41	No	1,358 (98.8)	77 (5.7)	1281 (94.3)	0.002
42					
43	Yes	17 (1.2)	4 (25.5)	13 (76.5)	
44					
45					
46					
47	Ever shared needles when injecting hormones/beauty substances				
48					
49	No	1355 (98.5)	78 (5.8)	550 (94.2)	0.33
50					
51	Yes	20 (3.2)	2 (10.0)	18 (90.0)	
52					
53					
54					
55	Ever used online services developed for MSM/TG (e.g. Facebook, website)				
56					
57	No	483 (35.1)	46 (9.5)	437 (90.5)	<0.001
58					
59					
60					

Yes 892 (64.9) 35 (3.9) 857 (96.1)

290 *Abbreviations: HIV, human immunodeficiency virus; USD, United States dollar.*

291 **Chi-square or Fisher's exact test was used as appropriate.*

292

293 Sexual behaviors

294 Sexual behaviors among transgender women with reactive and non-reactive HIV test are shown
 295 in Table 2. An overwhelming majority of study participants (87.5%) reported only having had
 296 receptive anal sex in the past 12 months. Of the total, 86.0% reported having had anal sex with
 297 at least one man within the previous three months, with the median number of male sex part-
 298 ners in the past three months being three (IQR 1-9). Of those who were sexually active with
 299 men in the past three months, 61.9% reported having used a condom at last sex with a man.

300 A total of 94.8% of participants who had anal sex in the prior three months reported
 301 having at least one non-transactional male sex partner within the past three months; of which,
 302 62.1% reported always using condoms with their non-transactional male sex partners. Of partic-
 303 ipants who had sex in the past three months, 41.8% reported having sex with at least one man
 304 in exchange for money or gift; of whom 60.0% reported always using condoms with the part-
 305 ners.

306 Table 2 also shows that HIV prevalence was significantly higher among participants who
 307 reported having had anal sex with a man in the past three months (6.5% vs. 2.1%, $p= 0.02$) and
 308 among those who reported having had sex with at least one man in exchange for money or gift
 309 in the prior three months (8.1% vs. 5.4%, $p= 0.04$) compared to participants who did not report
 310 these sexual behaviors.

311

312 **Table 2** Comparisons of sexual behaviors among transgender women with reactive and non-
 313 reactive HIV test

Sexual behaviors	Total (<i>n</i> = 1375)	HIV test result		<i>P</i> -value*
		Reactive (<i>n</i> = 81)	Non-reactive (<i>n</i> = 1294)	
	<i>n</i> (%)	<i>n</i> (%)	<i>n</i> (%)	

Role in anal sex with a man (past 12 months)

Insertive	29 (2.2)	1 (3.5)	28 (96.5)	0.73
Receptive	1145 (87.5)	72 (6.3)	1073 (93.7)	
Both	135 (10.3)	7 (5.2)	128 (94.8)	

Had anal sex with a man (past 3 months)

No	192 (14.0)	4 (2.1)	188 (97.9)	0.02
Yes	1183 (86.0)	77 (6.5)	1106 (93.5)	

Number of male sexual partners (past 3 months)

<2	512 (37.3)	24 (4.7)	488 (95.3)	0.14
≥2	861 (62.7)	57 (6.6)	804 (93.4)	

Used a condom at last sex (past 3 months)

No	451 (38.1)	27 (6.0)	423 (94.0)	0.57
Yes	732 (61.9)	50 (6.8)	682 (93.2)	

Had anal sex with a man, not in exchange for money or gift (past 3 months)

No	61 (5.2)	5 (8.2)	56 (91.8)	0.37
Yes	1122 (94.8)	72 (6.4)	1050 (93.6)	

Number of male sexual partner, not in exchange for money or gift (past 3 months)

< 2	357 (31.9)	24 (6.7)	333 (93.3)	0.78
≥2	763 (68.1)	48 (6.3)	715 (93.7)	

Condom use with male sexual partner not in exchange for money or gift (past 3 months)

Not always	697 (62.1)	50 (7.2)	647 (92.8)	0.19
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Always	425 (37.9)	22 (5.2)	403 (94.8)	
Had anal sex with a man in exchange for money or gift (past 3 months)				
No	688 (58.2)	37 (5.4)	651 (94.6)	0.04
Yes	495 (41.8)	40 (8.1)	455 (91.9)	
Number male sexual partners in exchange for money/gifts (past 3 months)				
< 2	161 (32.5)	12 (7.5)	149 (92.6)	0.72
≥2	334 (67.5)	28 (8.4)	306 (91.6)	
Condom use with male sexual partner in exchange for money or gifts (past 3 months)				
Not always	164 (40.0)	12 (7.3)	152 (92.7)	0.56
Always	246 (60.0)	22 (8.9)	224 (91.1)	

314 *Abbreviation: HIV, human immunodeficiency virus.*

315 **Chi-square test or Fisher's exact test was used as appropriate.*

317 **Sexual behaviors and awareness of HIV status**

318 Among the participants who had HIV reactive tests, additional analyses were conducted to see
 319 if there were differences in sexual behaviors among those who self-reported a positive status
 320 and those who did not. Participants who did not report or were unaware of their positive status
 321 were significantly less likely to report using a condom at last sexual intercourse (52.4% vs.
 322 79.0%, $p=0.01$). No other significant differences were found.

324 **Sexually transmitted infections (STIs)**

Overall, 14.0% of participants reported having had at least one STI symptom in the past 12 months. Anal and perianal symptoms were most commonly reported (6.1%). As shown in Table 3, HIV prevalence was significantly higher among participants who reported having had an STI symptom compared to that among participants who did not have an STI symptom in the past 12 months (11.4% vs. 5.0%; $p < 0.001$). HIV prevalence was also significantly higher among participants who reported having had an ulceration or sore in the genital area in the past 12 months compared to that among participants who did not have it (15.2% vs. 5.6%; $p < 0.001$).

Table 3 Comparisons of STI symptoms among transgender women with reactive and non-reactive HIV test

STI symptoms	Total (<i>n</i> = 1375)	HIV test result		P-value*
		Reactive (<i>n</i> = 81)	Non-reactive (<i>n</i> = 1294)	
	n (%)	n (%)	n (%)	
Had experienced any STI symptom (past 12 months)				
No	1182 (86.0)	59 (5.0)	1123 (95.0)	<0.001
Yes	193 (14.0)	22 (11.4)	171 (88.6)	
Ulceration or sores around the genitals (past 12 months)				
No	1329 (96.7)	74 (5.6)	1255(94.4)	<0.001
Yes	46 (3.4)	7 (15.2)	39 (84.8)	
Swelling around the genitals (past 12 months)				
No	1359 (98.8)	80 (5.9)	1279 (94.1)	1.000
Yes	16 (1.2)	1 (6.3)	15 (93.7)	
Abnormal urethral discharge (past 12 months)				

No	1339 (97.4)	77 (5.8)	1262(94.2)	0.16
Yes	36 (2.6)	4 (11.1)	32 (88.9)	

STI symptoms around the anus (past 12 months)

No	1291 (93.9)	73 (5.6)	1218 (94.4)	0.15
Yes	84 (6.1)	8 (9.52)	76 (90.48)	

STI symptoms in the mouth or throat (past 12 months)

No	1336 (97.2)	76 (5.7)	1260 (94.3)	0.07
Yes	39 (2.8)	5 (12.82)	34 (87.18)	

335 *Abbreviations: HIV, human immunodeficiency virus; STI, sexually transmitted infections.*

336 **Chi-square or Fisher's exact test was used as appropriate.*

337

338 **Substance use**

339 As shown in Table 5, the majority (75.9%) of participants reported drinking at least one can of
 340 beer or a glass of wine in the past 3 months, while 10.1% reported using amphetamine-type
 341 stimulants (Yama, Crystal Ice, Ecstasy), and 0.9% reported using other drugs (marijuana, heroin,
 342 etc.) in the previous 12 months. Of total, 1.5% reported having injected any illicit drugs in the
 343 past three months.

344 Table 4 also shows that HIV prevalence was significantly higher among participants who
 345 did not drink alcohol when compared to that among participants who reported drinking at least
 346 one can of beer or a glass of wine in the past three months (8.5% vs. 5.1%; $p= 0.02$). Moreover,
 347 HIV prevalence was significantly higher among participants who reported injecting any illicit
 348 drugs in the past three months compared to participants who did not (15.0% vs. 5.8%, $p= 0.01$).

349

350 **Table 4** Comparisons of substance use among transgender women with reactive and non-
 351 reactive HIV test

352

Substance use	Total (n= 1375)	HIV test result		P-value*
		Reactive (n= 81)	Non-reactive (n= 1294)	
	n (%)	n (%)	n (%)	
Drank at least one can of beer or glass of wine in the past 3 months				
No	331 (24.1)	28 (8.5)	303 (91.5)	0.02
Yes	1042 (75.9)	53 (5.1)	989 (94.9)	
Frequency of having more than 5 drinks in one day in the past 3 months				
Never more than five drinks	336 (24.4)	28 (8.3)	308 (91.7)	0.17
Less than once a month	771 (56.1)	40 (5.2)	731 (94.8)	
1-3 times a week	157 (11.4)	9 (5.7)	148 (94.3)	
4 or more times a week	111 (8.1)	4 (3.6)	107 (96.4)	
Used illicit drugs in the past 12 months				
Never	1224 (89.0)	72(5.9)	1152 (94.1)	0.74
Yes, ATS (yama, crystal ice, ecstasy)	139 (10.1)	8 (5.8)	131 (94.2)	
Other (marijuana, heroin, etc.)	12 (0.9)	1 (8.3)	11 (91.7)	
Injected any illicit drugs in the past 3 months				
No	1355 (98.6)	78 (5.8)	1277 (94.2)	0.01
Yes	20 (1.5)	3 (15.0)	17 (85.0)	
Had sex during/after using illicit drugs in the past 3 months				
No	1286 (93.5)	76 (5.9)	1210 (94.1)	1.00

Yes 89 (6.5) 5 (5.6) 84 (94.4)

353 *Abbreviation: ATS, amphetamine-type stimulant; HIV, human immunodeficiency virus.*

354 **Chi-square or Fisher's exact test was used as appropriate.*

356 **Factors associated with HIV infection**

357 Table 5 presents independent factors associated with HIV infection in multivariable logistic re-
 358 gression analyses. After adjustment for other covariates, participants living in urban areas were
 359 twice as likely to be HIV infected as those living in rural areas (AOR= 2.7, 95% CI= 1.1-6.5). Par-
 360 ticipants with primary education were 1.7 times as likely to be infected compared to those with
 361 high school education (AOR=1.7, 95% CI= 1.0-2.9). HIV infection increased with age; compared
 362 to those aged 18-24, the odds of being HIV infected were twice as high among transgender
 363 women aged 25-34 (AOR= 2.1, 95% CI= 1.2-3.6) and 2.8 times higher among those aged ≥ 35
 364 (AOR= 2.8, 95% CI= 1.3-6.1). Self-injection of gender affirming hormones was associated with a
 365 four-fold increase in the odds of HIV infection (AOR= 4.4, 95% CI= 1.1-17.3). A history of genital
 366 sores over the previous 12 months increased the odds of HIV infection by three-fold (AOR= 3.0,
 367 95% CI= 1.2-7.8). Transgender women with stronger feminine identity, dressing as a woman all
 368 the time were twice as likely to be HIV infected compared to those who did not dress as a
 369 woman all the time (AOR= 2.1, 95% CI= 1.2-3.8). Having never used online services developed
 370 for transgender women in the past six months was also associated with higher odds of being
 371 HIV infected (AOR= 1.9, 95% CI= 1.2-3.2).

372
 373 **Table 5** Factors associated with HIV infection in multivariate logistic regression model

Variables in the final model	AOR (95% CI)	P-value
Community type		
Rural	Reference	
Urban	2.7 (1.1-6.5)	0.03
Formal education attained		

Primary (0–6 years)	1.7 (1.1-2.9)	0.04
High school or higher (≥7)	Reference	
Age in years		
<25	Reference	
25-34	2.1 (1.2-3.6)	0.01
≥35	2.6 (1.3-5.4)	0.01
Frequency of express and/or dressing as a woman		
Not all the time	Reference	
All the time	2.1 (1.2-3.8)	0.01
Ever self-injected hormone		
No	Reference	
Yes	4.4 (1.1-17.3)	0.03
Ulcerations or sores in the genital area in the past 12 months		
No	Reference	
Yes	3.0 (1.2-7.8)	0.02
Used online services developed MSM/TG in the past 6 months (e.g. Facebook, website)		
No	1.9 (1.2-3.2)	0.01
Yes	Reference	

374 *Abbreviations: AOR, adjusted odds ratio; CI, confidence interval; HIV, human immunodeficiency virus.*

375 **Variables in the table were the ones that remained statistically significant after several steps of model*

376 *fitting.*

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3 378 **DISCUSSION**
4

5 379 This study reveals an HIV prevalence of 5.9% among transgender women in Cambodia. This
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7 380 prevalence was about 20 times higher than the 0.3% among women attending antenatal care
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9 381 clinics in 2014 who represent the general adult population aged 15-45,³⁰ and about 2.5 times
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11 382 higher than the 2.3% among MSM in 2014.²⁶ This observed prevalence emphasizes that
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13 383 transgender women in Cambodia are at high risk of HIV, and is consistent with global literature
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15 384 regarding the high burden of HIV among this transgender population.¹

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17 385 Although statistically non-significant ($p= 0.13$), the prevalence found in this study was
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19 386 higher than the prevalence of 4.2% reported in the smaller ($n= 891$) TGIBBS conducted in
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21 387 2012²² and was therefore contrary to our hypothesized expectations of reduction in HIV preva-
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23 388 lence among this population. Recent progress in Cambodia, where 73% of all estimated HIV
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25 389 positive people or 93% of those who have been aware of their positive status are in care,³¹ had
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27 390 prompted us to hypothesize the lower prevalence. Although we have no concrete evidence re-
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29 391 garding the non-significant increase of HIV prevalence, we suggest that it may be related to the
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31 392 sampling variation in the two surveys. The previous study recruited participants only from the
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33 393 capital city and five provinces (Phnom Penh, Battambang, Banteay Meanchey, Kampong Cham,
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35 394 Siem Reap and Preah Sihanouk),²² whereas our study expanded to additional eight provinces.

36 395 More than half (52%) of those who had HIV reactive tests reported that they had never
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38 396 learned about their HIV positive status before they participated in the study. We believe that
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40 397 this self-reporting of the HIV status is realistic. Since we were concerned that those who were
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42 398 already aware of their HIV status may be unwilling to report their status, we decided to use
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44 399 well-trained counselors who all were from the government's Voluntary Confidential Counseling
45
46 400 and Testing (VCCT) centers. These counselors had many years of experience in providing VCCT
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48 401 services and were better equipped to cope with difficult situations. In Cambodia, we believe
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50 402 people are more likely to trust their counselors, as opposed to interviewers. However, as in all
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52 403 self-reported measures, the potential for response bias cannot be entirely ruled out.

53 404 This study also reports important findings related to factors associated with HIV infec-
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55 405 tion among transgender women in Cambodia. First, the HIV prevalence among transgender
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57 406 women residing in urban communities was twice as high when compared to that among
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3 407 transgender women living in rural communities. This finding is similar to that in a previous
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5 408 study that found a higher HIV prevalence in transgender women living in urban areas of Siem
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7 409 Reap and Phnom Penh, compared to that among transgender women living in other provinc-
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9 410 es.²² Findings from our study suggest that these differences may not necessarily be attributable
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11 411 to common sexual risk practices. The mean number of sexual partners (8.4 vs. 7.9; $p= 0.70$) and
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13 412 the mean number of male commercial sexual partners (0.7 vs. 0.7; $p= 0.48$) in the past three
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15 413 months among transgender women living in urban and rural communities were not significantly
16
17 414 different. In addition, the rates of inconsistent condom use with male non-commercial sexual
18
19 415 partners in the past three months were also similar in the two groups (58.0% vs. 62%; $p= 0.32$).

20 416 Given the failure of sexual risk behaviors to sufficiently account for the difference in HIV
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22 417 prevalence, these data suggest that a higher HIV prevalence among sexual partners of
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24 418 transgender women in urban communities may possibly explain the higher probability of urban
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26 419 transgender women to get infected, although this may not be stated with certainty. This is par-
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28 420 ticularly relevant given that previous studies among all transgender women found a higher
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30 421 prevalence of HIV in urban areas.²² Studies among MSM in Cambodia have also found a higher
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32 422 prevalence of HIV in urban areas.^{24 32} In addition, a recent national sentinel survey among
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34 423 women attending antenatal care clinics found that HIV prevalence among women in urban are-
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36 424 as was consistently high,³⁰ implying that HIV prevalence in urban areas among most popula-
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38 425 tions, who potentially include transgender women and their partners, is high. It is also possible
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40 426 that transgender population in urban communities face unique or additional risks compared to
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42 427 other members of this population in rural areas,²² which could also contribute to the difference
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44 428 in HIV prevalence between urban and rural communities. Still, it is possible that those who knew
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46 429 they were positive may have moved to urban centers for care and treatment, which unfortunately can-
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48 430 not be examined further in this cross-sectional study. Further research is needed with regard to this
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50 431 observation.

51 432 Second, HIV prevalence among older transgender women (≥ 25 years) was significantly
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53 433 higher, compared to that among their younger counterparts. This finding is similar to those of
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55 434 previous studies in Cambodia²² and Thailand.³ A possible explanation is that older transgender
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3 435 women had been exposed to risks for a longer period of time; the number of their sexual part-
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5 436 ners accumulated over time and thus increased cumulative probability of HIV acquisition.²²
6

7 437 Third, our study suggests that more overt feminine expression (e.g., those who dressed
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9 438 as a woman all the time) was associated with a higher HIV prevalence. A study in the United
10
11 439 States found that transgender women who disclosed their female gender identity possessed a
12
13 440 higher HIV prevalence.³³ In our study, these transgender women who dressed as a woman all
14
15 441 the time had higher numbers of both male non-commercial (mean= 8.1 vs. 5.1; $p= 0.004$) and
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17 442 male commercial sexual partners (mean= 3.7 vs. 1.6; $p< 0.001$), compared to those who did not.
18
19 443 Making their female status visible could render more chances to meet with men. However, it
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21 444 could make them difficult to find a decent job due to stigmatization and discrimination, thus
22
23 445 leading them to engage in sex work. This finding is in line with existing literature showing that
24
25 446 transgender women try to have sex with men –both casual and paid sex– to prove or validate
26
27 447 their female gender identity.^{34–36} In these contexts, “sex work provides both desired gender af-
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29 448 firmation and economic stability, often with greater financial rewards for sex without a con-
30
31 449 dom.”³⁴ The desire to affirm their gender identity and attractiveness to men also incentivizes
32
33 450 them to engage in casual sex with multiple partners.¹⁵ This sexual behavior may make them
34
35 451 more prone to HIV infection.

36 452 There is a tendency to self-inject gender-affirming hormones, often with shared needles,
37
38 453 among transgender women in Cambodia.³⁷ This risky practice was associated with an increased
39
40 454 risk of HIV infection, implying that self-injecting was performed through unsafe means, and that
41
42 455 injecting by a trained health or other professional might mitigate the likelihood of HIV infection.
43
44 456 Although hormone injection to augment femininity is becoming more common, it can potential-
45
46 457 ly cause adverse health effects among transgender women.^{3 38} A Thai study also found that
47
48 458 transgender women who injected hormone to make them more feminine had a higher HIV
49
50 459 prevalence.³

51 460 Fourth, transgender women who completed or dropped out primary school had a higher
52
53 461 HIV prevalence than those with high-school education. This finding confirms results of the
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55 462 TGIBBS 2012 in Cambodia²² and other studies^{6 39} that associated low education with high HIV
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57 463 prevalence among transgender women. This has important implications for HIV programs to
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2
3 464 ensure that poorly educated transgender women are reached with education, information,
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5 465 communication and skills related to HIV prevention and other health-related services.
6

7 466 Fifth, transgender women with self-reported STI symptoms (having ulcerations or sores
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9 467 in the genital area in the past 12 months) had a greater HIV prevalence. This is in line with exist-
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11 468 ing evidence showing that STIs promote HIV transmission via a variety of biological mecha-
12
13 469 nisms.⁴⁰ Transgender women with high rates of STIs, particularly ulcerative genital diseases, are
14
15 470 at high risk of HIV acquisition.²³
16

17 471 Finally, transgender women who reported never using online services developed for
18
19 472 transgender women or MSM had an increased risk of HIV infection. Low utilization of online
20
21 473 services by key populations in Cambodia is iterated in other studies.⁴¹ Non-users of online ser-
22
23 474 vices, which tend to provide HIV information, education and communication, might have riskier
24
25 475 sexual behaviors. Our data suggest that transgender women who did not use online services
26
27 476 had higher numbers of overall male sexual partners (10.2 vs. 6.3, $p= 0.002$) and male commer-
28
29 477 cial sexual partners (0.8 vs. 0.6, $p= 0.001$) in the past three months. Also, they had a higher rate
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31 478 of inconsistent condom use with male commercial sexual partners in the past three months
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33 479 (48.5% vs. 34.4%, $p= 0.004$). Studies in other settings confirm that transgender women with
34
35 480 multiple sexual partners are exposed to a riskier level of HIV infection.^{6,15} This finding suggests
36
37 481 that access to community-based services tailored to transgender population can reduce their
38
39 482 vulnerability to HIV as demonstrated in other settings.⁴² Sustaining and increasing the coverage
40
41 483 of internet and peer-led community-based services for this population is particularly important,
42
43 484 given that they are often unable to access health services due to stigmatization and discrimina-
44
45 485 tion,^{16,41} even in health care settings.¹⁸ As in other settings globally,⁹ only half of those who
46
47 486 were infected with HIV were aware of their status, demonstrating the need to intensify HIV
48
49 487 testing, including self-testing at the community level, which transgender women in Cambodia
50
51 488 are willing to use.⁴³ Our study also found that once diagnosed, the coverage of ART was high in
52
53 489 this population.
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55 490

56 491 **LIMITATIONS OF THE STUDY**

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3 492 Our study covered only the capital city and 12 provinces, which contain the highest numbers of
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5 493 transgender women. Since cities and provinces with fewer TG were left out, these results may
6
7 494 not be generalized to all transgender women nationally. Second, the initial participant seeds
8
9 495 were identified and recruited by outreach workers of community-based organizations, which
10
11 496 could introduce bias towards transgender women under their programs, leading to a recruit-
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13 497 ment bias. This problem could be exacerbated by outreach workers interviewing some partici-
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15 498 pants who had received services from their NGO, which could have induced the participants'
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17 499 responses. Third, this study employed a self-reporting questionnaire on sensitive health and
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19 500 sexual behaviors, which may have been limited by social desirability bias. Fourth, albeit mini-
20
21 501 mal, the monetary incentive given to the participants to recruit seeds may have affected their
22
23 502 genuine motivation to partake in the study, which could influence their responses. Fifth, we
24
25 503 used the backward selection of variables in the multivariable analysis. By using this method, we
26
27 504 possibly dropped one or more variables that could be significant if we kept them until the final
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29 505 model. Finally, as this study was cross-sectional, it reports associations at a given time, and may
30
31 506 not be construed to be reporting causal relationships.

32 507

33 508 **CONCLUSIONS**

34
35 509 This study demonstrates that HIV prevalence among transgender women in Cambodia remains
36
37 510 persistently high. With the prevalence rate at 5.9%, higher prevalence was observed among
38
39 511 transgender women who resided in urban areas, of older age, with low education levels, with
40
41 512 previous genital sores, with a history of self-injected hormone and those who had never used
42
43 513 online services developed for transgender women or MSM. Therefore, to eliminate new HIV
44
45 514 infections among transgender women, tailored interventions need to focus on these sub-
46
47 515 populations and attendant risk factors.

48 516

49
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6
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10
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14
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29

30 535

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34
35 538 **Patient consent** Obtained
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39 540 **Ethics approval** The National Ethics Committee for Health Research (NE-CHR) of the Ministry of
40
41 541 Health, Cambodia (No. 420 NECHR) and FHI 360's Protection of Human Subjects Committee
42
43 542 (PHSC No. 713897).
44

45 543

46 544 **Consent for publication**

47
48 545 Consent to publish was obtained from participants as stated in the informed consent form.
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52 547 **Data sharing statement** Data used for this study cannot be made available in the manuscript,
53
54 548 the supplemental files or a public repository due to the ethical restriction stated in the agree-
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3 549 ment with the ethical committees. However, they can be accessed upon request from the Prin-
4
5 550 cipal Investigator (Dr. Siyan Yi) at siyan@doctor.com.
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For peer review only

Participant ID Code:

Integrated Biological and Behavioral Survey of Transgender Women in Cambodia, 2016

Date of interview: day.....month.....year.....

Province/City:

- | | | |
|---------------------|---------------------|------------------|
| 1. Phnom Pehn | 6. Preah Sihanouk | 11. Kandal |
| 2. Kampong Cham | 7. Prey Veng | 12. Koh Kong |
| 3. Battambang | 8. Svay Rieng | 13. Thbong Khmum |
| 4. Siem Reap | 9. Kampong Spue | |
| 5. Banteay Meanchey | 10. Kampong Chhnang | |

Interview Location (City/NGOs):

Name of administrative districts (current residence):

Status of administrative districts:

- 1- Urban
- 2- Rural

SECTION 1: SOCIODEMOGRAPHIC CHARACTERISTICS			
No.	Questions and filters	Coding categories	Skip to
Now I would like to ask you some questions related to your personal information.			
Q001	How old are you?	Age in completed years:	
Q002	What is your current marital status? (only one response)	Married and living together 1 Married ,but not living together 2 Widowed, Divorced or separated 3 Not married, not living with any partner 4 Not married, living with sweetheart (female) 5 Not married but living with male lover 6 Other (Specify.....) 7	
Q003	For how long have you been living in the current city?Years <u>Record 0</u> if not living in this city <u>Record 1</u> if living in this city 1 year or less Refuse to answer 99	
Q004	In the past 6 months, on average, how much money	Amount of money (in US\$): Refuse to answer 99	

	did you make per month? (exchange rate: 4000 riel per 1 US\$)		
Q005	How many years of formal education have you completed?	Number of years:	
		<u>Record 0</u> if never attending school.	
		Refuse to answer	99
Q006	What is your current job (main source of income)? (only one response) <i>Note: If you are both studying and having a paid job, report your main source of income.</i>	Unemployed 0 Hair dresser/Beautician 1 Government officer 2 Laborer (factory, construction work) 3 Seller 4 Entertainment Worker (bar, karaoke, etc.) 5 Sex worker 6 Student 7 NGO staff 8 Private company staff 9 Farmer/fisherman 10 Artist 11 Other (Specify.....) 12	

SECTION 2. TRANSGENDER IDENTITY & EXPERIENCES

Q007	What gender do you consider yourself?	Female 1 Male 2 Third gender 3 Uncertain 4 Refuse to answer 99	
Q008	How often do you express or dress yourself as a woman?	All the time 1 Often 2 Sometimes 3 Rarely 4 Refuse to answer 99	
Q009	How many friends in your social network are also transgender?	Number:	
Q010	Have you ever taken hormones or any substances for beauty purposes?	No 1 Yes 2 Refuse to answer 99	2→ Q014
Q011	What are the methods you have used to take hormones or any substances for beauty purposes? [multiple answer]	Pills 1 Injections 2 Skin patches 3 Other (Specify.....) 4 Refuse to answer 99	
Q012	If you have taken hormones or any substances for beauty	Never injected 0 Injected by myself 1	

	purposes by injection, how have you received it?	Injected by skilled personnel (beauty clinic) 2 Injected by non-skilled personnel 3 Other (Specify.....) 4	
Q013	If you have injected hormones or any substances for beauty purposes, have you shared needle?	Never injected 0 No 1 Yes 2 Refuse to answer 99	
Q014	Have you ever had any operation to change any parts of your body to become a woman?	No 1 Yes 2 Refuse to answer 99	

Section 3. SEXUAL PARTNERS AND SEXUAL HISTORY			
No.	Questions and filters	Coding categories	Skip to
	Now I would like to ask questions about your recent sexual relationship with various kinds of sexual partners.		
Q015	Have you ever had sex with any partner in your life time? [Including vaginal, or anal sex]	No 0 Yes 1 Refuse to answer 99	
Q016	How likely do you think you are infected with HIV?	Very Likely 1 Likely 2 Unlikely 3 Very Unlikely 4 Refuse to answer 99	
Sex with women			
Q017	Have you ever had sex with a woman? [Including vaginal or anal sex]	No 0 Yes 1 Refuse to answer 99	0, 99 → Q028
Q018	In the past 12 months, have you had sex with a woman? [Including vaginal or anal sex]	No 0 Yes 1 Refuse to answer 99	0, 99 → Q028
Q019	In the past 12 months, have you had sex with a woman not in exchange for money or gifts? [Including vaginal or anal sex]	No 0 Yes 1 Refuse to answer 99	0→ Q022
Q020	In the past 3 month, how many women did you have	Number of women:	

Questionnaire; Integrated Biological and Behavioral Survey among Transgender Population in Cambodia, 2015, Version 2.0, 17 September 2015 Page 3 of 11

	sex with not in exchange for money or gifts?	Refuse to answer 99	
Q021	In the past 3 month, how often did you use a condom when you had vaginal or anal sex with women not in exchange for money or gifts?	Never had sex with a woman 0 Always 1 Often 2 Sometimes 3 Never 4 Refuse to answer 99	0, 1→ Q024
Q022	In the past 3 month, if you did not always use a condom when you had sex with women not in exchange for money or gifts? What were the reasons? [Multiple Answers]	We are in a relationship 1 She is not HIV/STI infected 2 Too high to use a condom 3 No condom available 4 Feel better without a condom 5 I am HIV-infected 6 I penetrated, so I am not at risk 7 Partner refused 8 Other (Specify.....) 9 Refuse to answer 99	
Q023	In the past 12 months, have you had sex with a woman in exchange for money or gifts? [Including vaginal or anal sex]	No 0 Yes 1 Refuse to answer 99	0→ Q028
Q024	In the past 3 months, how many women did you have sex with in exchange for money or gifts? [Including vaginal or anal sex]	Number of women: Refuse to answer 99	
Q025	Where did you meet women with whom you had sex with in exchange for money or gifts??	Park 1 Street 2 Bar/discotheque/cafe 3 Beer Garden/Restaurant 4 Massage Parlor 5 Karaoke 6 Online (Facebook, Line, Website) 7 Guesthouse/hotel 8 Workplace/school 9 Other (Specify.....) 10	
Q026	In the past 12 months, how often did you use a condom when you had vaginal or anal sex with women in exchange for money or gifts?	Never had sex 0 Always 1 Often 2 Sometimes 3 Never 4 Refuse to answer 99	

Q027	In the past 3 months, if you did not always use a condom when you had sex with women not in exchange for money or gifts? What were the reasons? [Multiple Answers]	We are in a relationship 1 She is not HIV/STI infected 2 Too high to use a condom 3 No condom available 4 Feel better without a condom 5 I am HIV-infected 6 I penetrated, so I am not at risk 7 Partner refused 8 Other (Specify.....) 9 Refuse to answer 99	
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Sex with men			
Q028	Have you ever had anal sex with a man?	No 0 Yes 1 Refuse to answer 99	0, 99→ Q042
Q029	In the past 12 months, have you had anal sex with a man?	No 0 Yes 1 Refuse to answer 99	0, 99→ Q042
Q030	In the past 12 months, what was your usual role in anal sex with a man?	Insertive 1 Receptive 2 Both 3 Refuse to answer 99	
Q031	In the past 3 months, have you had sex with men not in exchange for money or gifts?	No 0 Yes 1 Refuse to answer 99	0→ Q033
Q032	In the past 3 months, how many men you had sex with not in exchange for money or gifts?	Number of men: Refuse to answer 99	
Q033	In the past 12 months, how often did you use a condom when you had anal sex with men not in exchange for money or gifts?	Never had sex 0 Always 1 Often 2 Sometimes 3 Never 4 Refuse to answer 99	0, 1→ Q035
Q034	If not always, why not? [Multiple Answers]	We are in a relationship 1 He is not HIV/STI infected 2 I was too drunk or too high with drugs 3 No condom available 4 Feel better without a condom 5 I am HIV-infected 6 I penetrated, so I am not at risk 7 Partner refused 8 Other (Specify.....) 9 Refuse to answer 99	

Q035	In the past 12 months, have you had sex with men in exchange for money or gifts?	No 0 Yes 1 Refuse to answer 99	0→ Q040
Q036	In the past 3 month, how many men you had sex with in exchange for money or gifts?	Number of men: Refuse to answer 99	
Q037	Where did you meet the men you had sex with in exchange for money or gifts?	Park 1 Street 2 Bar/discotheque/cafe 3 Beer Garden/Restaurant 4 Massage Parlor 5 Karaoke 6 Online (Facebook, Line, Website) 7 Guesthouse/hotel 8 Workplace/School 9 Other (Specify.....) 10	
Q038	In the past 12 months, how often did you use a condom when you had anal sex with men you paid for sex?	Never had anal sex 0 Always 1 Often 2 Sometimes 3 Never 4 Refuse to answer 99	0, 1→ Q615
Q039	If not always, why not? [Multiple Answers]	We are in a relationship 1 He is not HIV/STI infected 2 I was drunk or too high with drugs 3 No condom available 4 Feel better without a condom 5 I am HIV-infected 6 I penetrated, so I am not at risk 7 Partner refused 8 Other (Specify.....) 10 Refuse to answer 99	

Access to condoms and lubricant			
No.	Questions and filters	Coding categories	Skip to
Q040	In this last 12 months, where did you get condoms? [Multiple Answers]	Friends/outreach workers 1 Condom peer sale representative 2 Pharmacy/drug store/clinic 3 Condom outlets 4 Mart/mini-mart 5 Groceries 6 Hotel/guesthouse 7 Other (Specify.....) 8	

		Refuse to answer 99	
Q041	In this last 12 months, where did you get lubricant? [Multiple Answers]	Friends/outreach workers 1 Condom peer sale representative 2 Pharmacy/drug store/clinic 3 Condom outlets 4 Mart/mini-mart 5 Groceries 6 Hotel/guesthouse 7 Other (Specify.....) 8 Refuse to answer 99	

SECTION 4. STI AND HIV TESTING			
No.	Questions and filters	Coding categories	Skip to
Q042	In the past 12 months, have you experienced the following symptoms? [Multiple Answers]	Cuts or sores in the genital area 1 Swelling in the genital area 2 Abnormal urethral discharge 3 Symptoms on the anus 4 Symptom in the mouth/throat 5 Refuse to answer 99	
Q043	Where did you first go for treatment the last time you had any above-mentioned symptoms? (Only one response)	Never had any symptoms 0 Pharmacy 1 Private clinic/hospital 2 Public Hospital/STD clinic 3 NGO Clinic/hospital 4 Traditional doctor 5 Didn't get care 6 Other (Specify.....) 7 Refuse to answer 99	
Q044	Have you ever been tested for HIV?	No 0 Yes 1 Refuse to answer 99	0→ Q049
Q045	In past 6 months, have you been tested for HIV?	No 0 Yes 1 Refuse to answer 99	
Q046	How long has it been since you received the last HIV test?	Number of months.....	
Q047	If a rapid self-test kit for HIV was available and you could get the result within 20 minutes at home, would you use it?	No 0 Yes 1 Refuse to answer 99	

Q048	Where did you have your most recent HIV test?	Private facilities 1 Public facilities 2 NGO facilities 3 NGO outreach workers at community or HTC 4 Other (Specify.....) 5 Refuse to answer 99	
Q049	The last time you got tested for HIV, did you receive the result of the test?	No 0 Yes 1 Refuse to answer 99	
Q050	What is your current HIV status? (You could choose not to answer)	Positive 1 Negative 2 I don't know my status 3 Refuse to answer 99	2, 3 → Q057
Q051	Are you currently on ART?	No 0 Yes 1 I was but discontinued 2 I am on treatment for opportunistic infections 3 Refuse to answer 99	0 → Q056
Q052	Do you take ARV regularly as prescribed?	Regularly 1 Not regularly 2 Refuse to answer 99	
Q053	Where do you get ART services?	Public facilities 1 Private facilities 2 NGO facilities 3 Pharmacy 4 Other (Specify.....) 5 Refuse to answer 99	
Q054	If not, why are you not on ART?	Not needed 1 Not sure where to go 2 I am ashamed to go to health facility 3 I am afraid of being discriminated 4 I was treated badly in the past 5 Other (Specify.....) 6 Refuse to answer 99	

SECTION 5. ALCOHOL AND DRUG USE

Measurement of alcohol drinking			
One standard measurement: - A can/glass of beer, fermented palm juice (285 ml) - A glass of wine (120 ml) - A glass of whisky (30 ml)			
No.	Questions and filters	Coding categories	Skip to
Q055	In the past 3 months, how often did you drink at least one can of beer or one glass of any types of wine?	Never 1 Once a month or less 2 2-4 times a month 3 2-3 times a week 4 4 or more times a week 5 Don't know 98 Refuse to answer 99	1→ Q064
Q056	In the past 3 months, how many standard drinks containing alcohol (a can of beer or a glass of any types of wine) did you have on a typical day on which you drank alcohol?	1 - 2 1 3 - 4 2 5 - 6 3 7 - 9 4 10 or more 5 Don't know 98 Refuse to answer 99	
Q057	In the past 3 months, how often did you have more than 5 drinks in one day or night?	Never 1 Less than once a month 2 Once a month 3 1, 2, or 3 times a week 4 4 or more times a week 5 Don't know 98 Refuse to answer 99	
Q058	In the past 3 months, how often have you got drunk from alcohol?	Never 1 Less than once a month 2 Once a month 3 Every week (1-3 times/week) 4 Every day/almost every day(4 or more times a week) 5 Don't know 98 Refuse to answer 99	

Q059	In the past 12 months, have you tried any of the following drugs? (Ask one by one - CIRCLE YES OR NO) [Multiple answers]			
		Types of drug	Yes	No
	1	Marijuana	1	0
	2	Heroin/Opium	1	0
	3	Yama (amphetamine)	1	0
	4	Crystal, Ice (Methamphetamine)	1	0
	5	Ecstasy	1	0
	6	Inhalants (glue, paint, petrol, spray can)	1	0
	7	Other	1	0
Q060	In the past 3 months, did you inject any illicit drugs?			
		Heroin	1	
		Yama	2	
		Crystal, Ice (Methamphetamine)	3	
		Never injected any drug	4	
	Refuse to answer	99		
Q061	In the past 3 months, did you have sex during/after using illicit drugs?			
		Heroin	1	
		Yama	2	
		Ice, Amphetamine	3	
		Never used drug before having sex	4	
	Refuse to answer	99		

SECTION 9. EXPOSURE TO INTERVENTION PROGRAMS			
No.	Questions and filters	Coding categories	Skip to
Q062	In the past 3 months, have you been reached by NGO staff delivering HIV services?	No 0 Yes 1 Don't know 98 Refuse to answer 99	
Q063	In the past 3 months, what kind of services have you received (Check all that apply).	HIV/health education and/or materials 1 Condoms 2 Lubricants 3 VCCT/STI testing 4 Law support services 5 Other health services 6 Other (Specify.....) 7 Refuse to answer 99	
Q064	Do you have UIC card?	Yes 1 No 0	

		Refuse to answer 99	
Q065	In the past 12 months, how often did you visit MStyle/Srey Sros club(s)?	... times Refuse to answer 99	
Q066	Have you used online services developed for people like you (e.g. Facebook, website)?	No 0 Yes 1 Refuse to answer 99	0 -> stop interview
Q067	Have you found any sexual partner on an Internet website or any online program?	Never 0 Yes, always 1 Yes, sometimes 2 Yes, occasionally 3 Refuse to answer 99	
Q068	Have you accessed MStyle website in the past 6 months? (Attach the picture of MStyle webpage)	Never 0 Yes, always 1 Yes, sometimes 2 Yes, occasionally 3 Refuse to answer 99	

["Thank you very much for answering these questions. Let us repeat that your answers are totally confidential and there is no way anyone will learn what you told us.]

BMJ Open

HIV prevalence and factors associated with HIV infection among transgender women in Cambodia: Results from a national integrated biological and behavioral survey

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Keywords:	Transgender women, HIV infection, Risk factors, Prevention, Integrated Biological and Behavioral Survey, Cambodia

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Manuscripts

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3 1 **HIV prevalence and factors associated with HIV infection among transgender women in Cam-**
4 **odia: Results from a national integrated biological and behavioral survey**
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9 4 Chhim Srean,¹ Chanrith Ngin,² Pheak Chhoun,² Sovannary Tuot,² Cheaty Ly,³ Phalkun Mun,⁴
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3 30 **ABSTRACT**

4
5 31 **Objective:** To examine factors associated with HIV infection among transgender women in
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7 32 Cambodia.

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9 33 **Design:** Cross-sectional study

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11 34 **Settings:** HIV high-burden sites including the capital city and 12 provinces

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13 35 **Participants:** This study included 1,375 sexually-active transgender women with a mean age of
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15 36 25.9 years (SD= 7.1), recruited by using respondent driven sampling for structured question-
16
17 37 naire interviews and rapid finger-prick HIV testing.

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19 38 **Primary outcome measure:** HIV infection detected by using Determine™ antibody test.

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21 39 **Results:** HIV prevalence among this population was 5.9%. After adjustment for other covariates,
22
23 40 participants living in urban areas were twice as likely to be HIV infected as those living in rural
24
25 41 areas. Participants with primary education were 1.7 times as likely to be infected compared to
26
27 42 those with high school education. HIV infection increased with age; compared to those aged 18-
28
29 43 24, the odds of being HIV infected were twice as high among transgender women aged 25-34
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31 44 and 2.8 times higher among those aged ≥ 35 . Self-injection of gender affirming hormones was
32
33 45 associated with a four-fold increase in the odds of HIV infection. A history of genital sores over
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35 46 the previous 12 months increased the odds of HIV infection by three-fold. Transgender women
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37 47 with stronger feminine identity, dressing as a woman all the time were twice as likely to be HIV
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39 48 infected compared to those who did not dress as a woman all the time. Having never used
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41 49 online services developed for transgender women in the past six months was also associated
42
43 50 with higher odds of being HIV infected.

44
45 51 **Conclusions :** Transgender women in Cambodia are at high risk of HIV. To achieve the goal of
46
47 52 eliminating HIV in Cambodia, effective combination prevention strategies addressing the above
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49 53 risk factors among transgender women should be strengthened.

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51 54
52 55 **Key words:** Transgender women, HIV infection, Risk factors, Prevention, Integrated Biological
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54 56 and Behavioral Survey, Cambodia.

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3 59 **Strengths and limitations of this study**
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- This is a rare and the second ever Integrated Biological and Behavioral Survey (IBBS) among transgender women—one of the most vulnerable and under-studied populations in Cambodia and globally.
 - This study covered HIV high-burden sites including the capital city and 12 provinces, thus providing more nationally generalizable results.
 - This study identified important risk factors for HIV infection among transgender women, which require being addressed in order to reduce HIV infection among this key population.
 - Limitations of the study included potential bias of self-reported measures, participant sampling, the cross-sectional nature of the data that limits causation inferences, and the backward selection of variables in multivariable analysis.

75 INTRODUCTION

76 Globally, transgender women are at high risk for HIV infection, and little is known about the
77 burden of HIV infection and its related factors in this population. A 2013 systematic review
78 showed that the global HIV prevalence among transgender women was 19%, with low- and
79 middle-income countries having a slightly lower prevalence of 18% compared to 22% in high-
80 income countries.¹ The probability of being infected with HIV among transgender women was
81 approximately 49 times higher than among the general adult population.¹ Epidemiologically,
82 transgender women share some HIV risk factors with other populations, such as ulcerative sex-
83 ually transmitted infections (STIs),^{2,3} multiple substance use, victimization, intimate partner vio-
84 lence,⁴ unprotected sex,⁵ multiple sex partners,⁶ and injecting drug use.⁵⁻⁷

85 However, transgender women encounter additional and unique biological, social, cul-
86 tural, legal and economic issues, which increase their vulnerability to HIV. Transgender women
87 are marginalized legally and economically through discriminative legislation and practice, which
88 accelerates their vulnerability to HIV infection by confining their job options to sex work. Sex
89 work in turn exacerbates their stigmatization and alienation,⁸ and further increases their HIV
90 risk.^{9,10} Stigmatization and discrimination further increase the risk of HIV infection by prevent-
91 ing transgender women from seeking essential health information and services.¹¹ Depression
92 and low self-esteem, which are common psychological consequences of stigma, further weak-
93 ens the bargaining power for condom use in sexual relationships, resulting in heightened vul-
94 nerability to HIV infection.^{5,12-15} Transgender women who have sex with men often engage in
95 receptive anal intercourse, which increases their risk of HIV acquisition, compared to insertive
96 sexual partners.² Syndemics of illicit drug use, abuse by family members, and mental health of-
97 ten occur together;¹⁶ and are often associated with less likelihood of transgender women to use
98 condoms during sex with commercial partners.¹⁷ In addition, the confluence of arbitrary ar-
99 rests,^{18,19} violence,^{16,20} economic pressures,²¹ and other social problems further aggravate
100 mental health risks among transgender women, causing them to engage in risky sexual behav-
101 iors.^{6,17}

102 In Southeast Asia, a systematic review of studies from Indonesia, Thailand and Vietnam
103 demonstrated that transgender women are particularly at high HIV risk, with an estimated HIV

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3 104 prevalence of 26.1%, 12.2% and 6.7%, respectively.¹ In Cambodia, up to 2012, transgender
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5 105 women were traditionally included in HIV prevention programs targeting men who have sex
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7 106 with men (MSM). As a result, their unique needs were not reflected in HIV programs' goals.²²
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9 107 There was also a lack of transgender women-specific data to quantify the number reached by
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11 108 outreach programs and services. In 2012, the first Integrated Biological and Behavioral Survey
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13 109 (IBBS) that specifically included transgender women in Cambodia found an HIV prevalence of
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15 110 4.2% among this population,²² which was six times higher than the 0.7% prevalence among the
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17 111 general adult population aged 15-45,²³ and about twice higher than the 2.1% among MSM.²⁴
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19 112 This 2012 study also found that older age (>35 years), residing in Siem Reap province (a major
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21 113 tourist site), having sex during or after drug use, not using a condom during the last anal sex,
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23 114 and low self-esteem were associated with HIV infection among transgender women.²²

24 115 Recognizing the differences in HIV vulnerability and prevention needs between
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26 116 transgender women and MSM, in 2013 the National Center for HIV/AIDS, Dermatology and STD
27
28 117 (NCHADS) separated these two populations in its revised Standard Operating Procedure (SOP)
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30 118 known as Boosted Continuum of Prevention, Care and Treatment (B-CoPCT).²⁵ Transgender
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32 119 women have since been considered a distinct at-risk group for HIV surveillance and programing.
33
34 120 The National B-CoPCT approach aims to increase the uptake of HIV testing and counseling (HTC)
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36 121 and other related services, ensure the quality of outreach and strengthen service delivery to
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38 122 meet specific needs of key populations, including transgender women, in-order to achieve
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40 123 Cambodia's 3.0 goal (i.e. zero new HIV infection, zero discrimination, and zero AIDS-related
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42 124 deaths) by 2020.

43 125 This study was conducted to explore the HIV prevalence and identify risk factors associ-
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45 126 ated with HIV infection among transgender women in Cambodia. The study findings will help
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47 127 determine ways in which services and policies can be tailored for this key population.
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50 129 **METHODS**

51 130 **Study design and sites**

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54 131 Between December 2015 and February 2016, a cross-sectional study was conducted in the capi-
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56 132 tal city of Phnom Penh and 12 provinces namely Battambang, Banteay Meanchey, Kampong
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3 133 Cham, Kandal, Koh Kong, Kampong Chhnang, Kampong Speu, Prey Veng, Preah Sihanouk, Siem
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5 134 Reap, Svay Rieng and Tbong Khmum. These sites were purposively selected from the 23 HIV
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7 135 high-burden operational districts identified by NCHADS.^{26 27} The study was a combination of a
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9 136 biological (blood test) and behavioral (face-to-face interview) survey using Respondent Driven
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11 137 Sampling (RDS) method to reach the target population.

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14 139 **Sample size and sampling procedures**

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16 140 The sample size calculation was based on an assumption that HIV prevalence would have
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18 141 changed between the first and the current IBBS. The minimum sample size required for this
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20 142 study was 1,380, which would result in sufficient power to detect a priori significant difference
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22 143 in HIV prevalence, as expressed in the following null and alternative hypotheses. The hypothesis
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24 144 test was that of one-sample proportion in comparison to the null hypothesis.

25
26 145 The null hypothesis (H_0): HIV prevalence among transgender women remains constant at
27
28 146 4.2%, similar to that found in the 2012 IBBS.

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30 147 The alternative hypothesis (H_a): HIV prevalence among transgender women has decreased
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32 148 by 1.2% from 4.2% in 2012 to 3.0% in the present IBBS.

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34 149

35 150 The sample size was calculated using the following formula and assumptions:

$$n = \frac{p_0q_0 \left(z_{1-\alpha} + z_{1-\beta} \sqrt{p_1q_1/p_0q_0} \right)^2}{(p_1 - p_0)^2}$$

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43 152 ▪ P_0 = estimated proportion in H_0 (In the most recent survey, HIV prevalence among TG
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45 153 women was 4.2% in 2012.)

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47 154 ▪ P_1 = estimated proportion in H_a (The expected HIV prevalence among TG women in the
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49 155 current study is about 3.0 %)

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51 156 ▪ $Z_{(1-\alpha)}$ = significance level at 5% in response to one-sided test (Z score = 1.645)

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53 157 ▪ $Z(1-\beta)$ = power level of 80% (Z score = 0.83)

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55 158 ▪ Refusal rate of 10%

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57 159 ▪ Design effect is assumed to be one.

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5 161 This sample size was stratified by study site. Roughly half of the estimated transgender
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7 162 women in each study site were recruited. However, in five provinces where the estimated
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9 163 number of transgender women was smaller than 100, all transgender women were recruited.
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11 164 Potential participants were included in the study if they: (1) were biologically male at birth and
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13 165 self-identified as a woman or third gender, (2) were Khmer-speaking, (3) were at least 18 years
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15 166 old at the time of screening, (4) reported having had sex with at least one man within the past
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17 167 12 months and (5) were able and willing to provide an informed consent.

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19 168 Among the 13 study sites, data collection was conducted in 20 locations (six locations in
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21 169 Phnom Penh and 14 locations in the remaining provinces). The number of the selected loca-
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23 170 tions was determined based on the required sample size in each study site. Our participants,
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25 171 including the seeds and the people the seeds referred, had to meet the eligibility criteria. These
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27 172 participants were recruited from the entire transgender population in the study sites. However,
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29 173 the initial seeds at each site were recruited through our implementing partners. First, four
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31 174 “seeds” (two seeds aged 18 to 24 and two seeds 25 or older) who were well connected with
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33 175 other transgender women in each location were selected by outreach workers from imple-
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35 176 menting partners (local NGOs) based in the selected locations. These seeds had to meet the
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37 177 above-mentioned eligibility criteria for participation and have an established and large social
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39 178 network consisting of 10 or more other transgender women in their given location. Eligibility to
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41 179 participate as a seed was determined by the leader of the data collection team, using a paper-
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43 180 based eligibility form.

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45 181 Second, each seed was given a personal identity number (PIN) and enrolled in the study.
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47 182 Third, each seed was given three coupons and asked to refer three additional transgender
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49 183 women. US\$2 was given to each seed for a successful referral. Each seed was expected to ex-
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51 184 tend to three to six “recruitment waves” in each location. If the initial seeds did not recruit
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53 185 participants, or if enrollment was halted because all recruitment chains had “dried up” (i.e.
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55 186 stopped recruiting), additional seeds were selected. In total, 80 seeds were selected by the out-
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57 187 reach workers, and a total of 1,375 transgender women were enrolled in the study. Referred
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59 188 participants were initially screened by the data collectors for eligibility.
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5 190 **Data collection training**

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7 191 Data were collected by three teams; each team comprised of one field supervisor, five inter-
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9 192 viewers, one lab technician and one counselor. Lab technicians and counselors were from the
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11 193 Municipal or Provincial AIDS and STI Program of the study sites. Data collection teams were
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13 194 trained for three days on study protocol, research ethics, interview techniques and data collec-
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15 195 tion procedures provided by the principle investigators and research coordinators.

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18 197 **Data collection procedures**19
20 198 *Biological data collection*

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22 199 To determine HIV prevalence, all participants received onsite rapid finger-prick testing. Pre-test
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24 200 counseling was provided by qualified, well-trained counselors working for voluntary confiden-
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26 201 tial counseling and testing (VCCT) centers. Participants could receive their HIV-test result ver-
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28 202 bally after the questionnaire interview. A blood sample was obtained from each participant by a
29
30 203 trained laboratory technician through finger-prick and tested for HIV using Determine™ test, in
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32 204 keeping with the national protocol.²⁸ Post-test counseling was provided for each participant
33
34 205 regardless of their HIV test result by the same counselor who conducted the pre-test counseling
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36 206 in accordance with NCHADS HIV testing guidelines.²⁸ Participants who were HIV reactive and
37
38 207 did not know their HIV status were referred by the counselor for confirmatory testing at the
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40 208 nearest VCCT center. The quality of HIV tests was monitored using quality control samples. Dis-
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42 209 cordant results between the screening and confirmatory tests were investigated to elucidate
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44 210 potential causes and minimize potential erroneous results. In the case of human error, individ-
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46 211 uals performing screening tests were retrained before resuming their involvement in the study.

47 212 After HIV testing, the participant was interviewed by a well-trained interviewer using a
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49 213 computer-assisted survey instrument (CASI). The questionnaire was set up in Qualtrics, a web-
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51 214 based application, and run on an Android tablet. HIV testing and interviews were conducted in
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53 215 the selected 20 locations including drop-in centers, private houses and offices of implementing
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55 216 partners, depending on participants' convenience.

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218 *Questionnaire development and measures*

219 The questionnaire was developed using standardized and validated tools adapted from TGIBBS-
220 2012,²² FHI 360's guideline for Behavioral Surveillance Survey (BSS)²⁹ and NCHADS's Boosted-
221 CoPCT²⁵ to measure key variables related to the objectives of the study. The questionnaire was
222 initially drafted in English and translated into Khmer, the national language of Cambodia. Con-
223 sultative meetings to improve the draft questionnaire were held with representatives of
224 transgender women, communities and NGOs working with transgender women, as well as re-
225 searchers and practitioners working on HIV and AIDS in Cambodia. The questionnaire was pre-
226 tested with 20 transgender women in Phnom Penh and finalized based on findings.

227 The questionnaire collected information on socio-economic characteristics, transgender
228 identity and related experiences, sexual behaviors and condom use with different types of sex-
229 ual partners, HIV/STI screening and care seeking behaviors, substance use and exposure to HIV
230 programs. For details of the questionnaire, please see Supplementary File 1.

231

232 **Data analyses**

233 HIV prevalence was calculated by dividing the total number of participants with HIV reactive
234 test results with the total number of participants. To examine the associated factors of HIV in-
235 fection, we conducted both bivariate and multivariable analyses. In the bivariate analyses, we
236 compared all characteristics and behavioral variables of participants with a reactive test result
237 to those of participants with a non-reactive test result. Among participants who tested HIV pos-
238 itive, additional analyses were conducted to assess whether there was a significant difference in
239 sexual behaviors of those who were aware of their HIV-positive status and of those who were
240 not. Chi-square test or Fisher's exact test (for an expected cell value of ≤ 5) was used for cate-
241 gorical variables, while Student's *t*-test was used for continuous variables. To facilitate the
242 model, some continuous variables, such as age were transformed to categorical variables. A
243 multivariable logistic regression model was constructed to examine independent factors associ-
244 ated with HIV infection. Variables with a significance level of $p < 0.05$ in the bivariate analyses
245 were simultaneously included in the model. Backward elimination method was then used to

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3 246 eliminate variables with the highest p -value one-by-one from the model. STATA Version 12.0
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5 247 for Windows (Stata Corp, TX, USA) was used to conduct the data analyses.
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9 249 **ETHICAL STATEMENT**

10 250 Participation in this study was voluntary, and a written informed consent was obtained from
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12 251 each study participant after a detailed description of the study objective and procedures was
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14 252 explained to them. Participants were informed that they could stop responding to questions
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16 253 and discontinue their participation at any time. Interviews were conducted at a private place,
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18 254 and confidentiality was enhanced by assigning a unique and anonymous code to each partici-
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20 255 pant. No personal identifiers were contained in the questionnaires or dataset. The study proto-
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22 256 col was approved by the National Ethics Committee for Health Research (NECHR) of the Minis-
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24 257 try of Health, Cambodia (No. 420 NECHR) and FHI 360's Protection of Human Subjects Commit-
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26 258 tee (PHSC No. 713897).
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29
30 260 **RESULTS**

31 261 **HIV prevalence**

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33 262 Of 1,375 participants tested, 81 (5.9%) had an HIV reactive test; of whom, 42 (52%) were not
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35 263 aware of their HIV status prior to the study. Out of the 39 cases who already knew their status,
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37 264 37 (94.9%) were currently on HIV treatment, with the other two reporting having dropped out
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39 265 of the treatment.
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43 267 **Socio-demographic characteristics**

44 268 As shown in Table 1, the majority of the participants (83.4%) were recruited from urban com-
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46 269 munities; 53.0% were younger than 25 years old and 97.2% had never been married. More than
47
48 270 two-thirds of participants (68.6%) had completed high school, and 9.1% had a higher education.
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50 271 The most common main occupations reported by the participants were hair dress-
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52 272 ers/beauticians (35.1%), laborers/farmers (17.5%) and entertainment workers (14.8%). More
53
54 273 than one-third of participants (38.6%) reported an average monthly income in the past six
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56 274 months of US\$ 100-199, while 16.5% reported it to be more than US\$ 300. Regarding gender
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identity, 42.2% identified themselves as female, while 57.2% identified themselves as third gender. Almost half (48.0%) of the participants reported dressing as a woman all the time; 45% ever used hormone/non-hormone substance; and 14.0% ever injected hormones.

HIV prevalence was significantly higher among participants living in urban communities compared to participants living in rural communities (6.5% vs. 2.6%; $p= 0.02$). The HIV prevalence was also significantly different by age group: 3.0% of participants in the age group of 18-24, 8.2% in 25-34, 13.1% in 35-44 and 11.4% in 45 or older ($p< 0.001$). HIV prevalence was also significantly higher among those who never attended school or dropped out of primary school when compared to those who at least attended high school or higher (10.4% vs. 8.0%; $p=<0.001$). In addition, HIV prevalence was significantly higher among transgender women who were NGO staff (mainly HIV focused NGOs) (20.6%) and those who were unemployed (9.4%) ($p= 0.03$). HIV prevalence was significantly higher among transgender women who reported dressing as women all the time (8.5% vs. 3.5%, $p= 0.001$), among those who ever used female hormones (8.1% vs. 4.1%, $p= 0.02$) and among those who ever injected hormones (9.1% vs. 5.2%, $p= 0.002$). Moreover, HIV prevalence was significantly higher among transgender women who reported never using online services developed specifically for MSM or transgender women, such as Facebook group pages or various websites (9.5% vs. 3.9%, $p< 0.001$).

Table 1 Comparison of socio-demographic, gender identity and hormone use characteristics of transgender women with and without a reactive HIV test

Socio-demographics, gender identity and hormone use	Total (<i>n</i> = 1375)	HIV test result		<i>P</i> -value*
		Reactive (<i>n</i> = 81)	Non-reactive (<i>n</i> = 1294)	
	<i>n</i> (%)	<i>n</i> (%)	<i>n</i> (%)	
Community type				
Urban	1146 (83.4)	75 (6.5)	1071 (93.5)	0.02
Rural	229 (16.6)	6 (2.6)	223 (97.4)	

Age in years

18- 24	729 (53.0)	22 (3.0)	707 (97.0)	<0.001
25-34	503 (36.6)	41 (8.2)	462 (91.8)	
≥35	143 (10.4)	18 (12.6)	125 (87.4)	

Current marital status

Married	7 (0.5)	0 (0.0)	7 (100)	0.47
Widowed/divorced/separated	18 (1.3)	1 (5.6)	17 (94.4)	
Never married	1334 (97.2)	78 (5.9)	1256 (94.1)	
Refused to answer	16 (1.2)	2 (15.4)	11 (84.6)	

Years of formal education completed

Primary (0-6 years)	307 (22.3)	32 (10.4)	275 (89.6)	<0.001
High school or higher (>7 years)	1068 (77.7)	49 (4.6)	1019 (95.4)	

Main occupation

Unemployed	64 (4.7)	6 (9.4)	58 (90.6)	0.03
Hair dresser/beautician	482 (35.1)	24 (5.0)	458 (95.0)	
Office worker	50 (3.6)	3 (6.0)	47 (94.0)	
Labor/farmer	241 (17.5)	15 (6.2)	226 (93.8)	
Seller	149 (10.8)	10 (6.7)	139 (93.3)	
Entertainment Worker	203 (14.8)	10 (4.9)	193 (95.1)	
Student	108 (7.9)	1 (0.9)	107 (99.1)	
NGO staff	34 (2.5)	7 (20.6)	27 (79.4)	

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Other	44 (3.2)	5 (11.4)	39 (88.6)	
Monthly income in the past 6 months (USD)				
< 100	351 (25.6)	22 (6.3)	329 (93.7)	0.70
100-199	530 (38.6)	29 (5.5)	501 (94.5)	
200-299	266 (19.3)	19 (7.1)	247 (92.9)	
≥300	226 (16.5)	11 (4.9)	215 (95.1)	
Gender identity (self-identified)				
Female	580 (42.2)	29 (5.0)	551 (95.0)	0.35
Third gender	786 (57.2)	52 (6.6)	734 (93.4)	
Uncertain	8 (0.6)	0 (0.0)	8 (100)	
Frequency of dressing as a woman				
All the time	660 (48.0)	56 (8.5)	604 (91.5)	0.001
Not all the time	715 (52.0)	25 (3.5)	689 (96.5)	
Ever injected hormones				
No	1123 (81.7)	58 (5.2)	1065 (94.8)	0.02
Yes	252 (18.3)	23 (9.1)	229 (90.9)	
Ever self-injected hormones				
No	1,358 (98.8)	77 (5.7)	1281 (94.3)	0.002
Yes	17 (1.2)	4 (25.5)	13 (76.5)	
Ever shared needles when injecting hormones/beauty substances				
No	1355 (98.5)	78 (5.8)	550 (94.2)	0.33

Yes	20 (3.2)	2 (10.0)	18 (90.0)	
Ever used online services developed for MSM/TG (e.g. Facebook, website)				
No	483 (35.1)	46 (9.5)	437 (90.5)	<0.001
Yes	892 (64.9)	35 (3.9)	857 (96.1)	

Abbreviations: HIV, human immunodeficiency virus; USD, United States dollar.

*Chi-square or Fisher's exact test was used as appropriate.

Sexual behaviors

Sexual behaviors among transgender women with reactive and non-reactive HIV test are shown in Table 2. An overwhelming majority of study participants (87.5%) reported only having had receptive anal sex in the past 12 months. Of the total, 86.0% reported having had anal sex with at least one man within the previous three months, with the median number of male sex partners in the past three months being three (IQR 1-9). Of those who were sexually active with men in the past three months, 61.9% reported having used a condom at last sex with a man.

A total of 94.8% of participants who had anal sex in the prior three months reported having at least one non-transactional male sex partner within the past three months; of which, 62.1% reported always using condoms with their non-transactional male sex partners. Of participants who had sex in the past three months, 41.8% reported having sex with at least one man in exchange for money or gift; of whom 60.0% reported always using condoms with the partners.

Table 2 also shows that HIV prevalence was significantly higher among participants who reported having had anal sex with a man in the past three months (6.5% vs. 2.1%, $p=0.02$) and among those who reported having had sex with at least one man in exchange for money or gift in the prior three months (8.1% vs. 5.4%, $p=0.04$) compared to participants who did not report these sexual behaviors.

Table 2 Comparisons of sexual behaviors among transgender women with reactive and non-reactive HIV test

Sexual behaviors	Total	HIV test result
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	(n= 1375)	Reactive (n= 81)	Non-reactive (n= 1294)	
	n (%)	n (%)	n (%)	P-value*
Role in anal sex with a man (past 12 months)				
Insertive	29 (2.2)	1 (3.5)	28 (96.5)	0.73
Receptive	1145 (87.5)	72 (6.3)	1073 (93.7)	
Both	135 (10.3)	7 (5.2)	128 (94.8)	
Had anal sex with a man (past 3 months)				
No	192 (14.0)	4 (2.1)	188 (97.9)	0.02
Yes	1183 (86.0)	77 (6.5)	1106 (93.5)	
Number of male sexual partners (past 3 months)				
<2	512 (37.3)	24 (4.7)	488 (95.3)	0.14
≥2	861 (62.7)	57 (6.6)	804 (93.4)	
Used a condom at last sex (past 3 months)				
No	451 (38.1)	27 (6.0)	423 (94.0)	0.57
Yes	732 (61.9)	50 (6.8)	682 (93.2)	
Had anal sex with a man, not in exchange for money or gift (past 3 months)				
No	61 (5.2)	5 (8.2)	56 (91.8)	0.37
Yes	1122 (94.8)	72 (6.4)	1050 (93.6)	
Number of male sexual partner, not in exchange for money or gift (past 3 months)				
< 2	357 (31.9)	24 (6.7)	333 (93.3)	0.78

≥2	763 (68.1)	48 (6.3)	715 (93.7)	
Condom use with male sexual partner not in exchange for money or gift (past 3 months)				
Not always	697 (62.1)	50 (7.2)	647 (92.8)	0.19
Always	425 (37.9)	22 (5.2)	403 (94.8)	
Had anal sex with a man in exchange for money or gift (past 3 months)				
No	688 (58.2)	37 (5.4)	651 (94.6)	0.04
Yes	495 (41.8)	40 (8.1)	455 (91.9)	
Number male sexual partners in exchange for money/gifts (past 3 months)				
< 2	161 (32.5)	12 (7.5)	149 (92.6)	0.72
≥2	334 (67.5)	28 (8.4)	306 (91.6)	
Condom use with male sexual partner in exchange for money or gifts (past 3 months)				
Not always	164 (40.0)	12 (7.3)	152 (92.7)	0.56
Always	246 (60.0)	22 (8.9)	224 (91.1)	

319 *Abbreviation: HIV, human immunodeficiency virus.*

320 **Chi-square test or Fisher's exact test was used as appropriate.*

322 **Sexual behaviors and awareness of HIV status**

323 Among the participants who had HIV reactive tests, additional analyses were conducted to see
 324 if there were differences in sexual behaviors among those who self-reported a positive status
 325 and those who did not. Participants who did not report or were unaware of their positive status
 326 were significantly less likely to report using a condom at last sexual intercourse (52.4% vs.
 327 79.0%, $p=0.01$). No other significant differences were found.

329 **Sexually transmitted infections (STIs)**

330 Overall, 14.0% of participants reported having had at least one STI symptom in the past 12
 331 months. Anal and perianal symptoms were most commonly reported (6.1%). As shown in Table
 332 3, HIV prevalence was significantly higher among participants who reported having had an STI
 333 symptom compared to that among participants who did not have an STI symptom in the past
 334 12 months (11.4% vs. 5.0%; $p < 0.001$). HIV prevalence was also significantly higher among par-
 335 ticipants who reported having had an ulceration or sore in the genital area in the past 12
 336 months compared to that among participants who did not have it (15.2% vs. 5.6%; $p < 0.001$).

337
 338 **Table 3** Comparisons of STI symptoms among transgender women with reactive and non-
 339 reactive HIV test

STI symptoms	Total (<i>n</i> = 1375)	HIV test result		P-value*
		Reactive (<i>n</i> = 81)	Non-reactive (<i>n</i> = 1294)	
	n (%)	n (%)	n (%)	
Had experienced any STI symptom (past 12 months)				
No	1182 (86.0)	59 (5.0)	1123 (95.0)	<0.001
Yes	193 (14.0)	22 (11.4)	171 (88.6)	
Ulceration or sores around the genitals (past 12 months)				
No	1329 (96.7)	74 (5.6)	1255(94.4)	<0.001
Yes	46 (3.4)	7 (15.2)	39 (84.8)	
Swelling around the genitals (past 12 months)				
No	1359 (98.8)	80 (5.9)	1279 (94.1)	1.000
Yes	16 (1.2)	1 (6.3)	15 (93.7)	
Abnormal urethral discharge (past 12 months)				

No	1339 (97.4)	77 (5.8)	1262(94.2)	0.16
Yes	36 (2.6)	4 (11.1)	32 (88.9)	

STI symptoms around the anus (past 12 months)

No	1291 (93.9)	73 (5.6)	1218 (94.4)	0.15
Yes	84 (6.1)	8 (9.52)	76 (90.48)	

STI symptoms in the mouth or throat (past 12 months)

No	1336 (97.2)	76 (5.7)	1260 (94.3)	0.07
Yes	39 (2.8)	5 (12.82)	34 (87.18)	

Abbreviations: HIV, human immunodeficiency virus; STI, sexually transmitted infections.

*Chi-square or Fisher's exact test was used as appropriate.

Substance use

As shown in Table 5, the majority (75.9%) of participants reported drinking at least one can of beer or a glass of wine in the past 3 months, while 10.1% reported using amphetamine-type stimulants (Yama, Crystal Ice, Ecstasy), and 0.9% reported using other drugs (marijuana, heroin, etc.) in the previous 12 months. Of total, 1.5% reported having injected any illicit drugs in the past three months.

Table 4 also shows that HIV prevalence was significantly higher among participants who did not drink alcohol when compared to that among participants who reported drinking at least one can of beer or a glass of wine in the past three months (8.5% vs. 5.1%; $p = 0.02$). Moreover, HIV prevalence was significantly higher among participants who reported injecting any illicit drugs in the past three months compared to participants who did not (15.0% vs. 5.8%, $p = 0.01$).

Table 4 Comparisons of substance use among transgender women with reactive and non-reactive HIV test

Substance use	Total (n= 1375)	HIV test result		P-value*
		Reactive (n= 81)	Non-reactive (n= 1294)	
	n (%)	n (%)	n (%)	
Drank at least one can of beer or glass of wine in the past 3 months				
No	331 (24.1)	28 (8.5)	303 (91.5)	0.02
Yes	1042 (75.9)	53 (5.1)	989 (94.9)	
Frequency of having more than 5 drinks in one day in the past 3 months				
Never more than five drinks	336 (24.4)	28 (8.3)	308 (91.7)	0.17
Less than once a month	771 (56.1)	40 (5.2)	731 (94.8)	
1-3 times a week	157 (11.4)	9 (5.7)	148 (94.3)	
4 or more times a week	111 (8.1)	4 (3.6)	107 (96.4)	
Used illicit drugs in the past 12 months				
Never	1224 (89.0)	72(5.9)	1152 (94.1)	0.74
Yes, ATS (yama, crystal ice, ecstasy)	139 (10.1)	8 (5.8)	131 (94.2)	
Other (marijuana, heroin, etc.)	12 (0.9)	1 (8.3)	11 (91.7)	
Injected any illicit drugs in the past 3 months				
No	1355 (98.6)	78 (5.8)	1277 (94.2)	0.01
Yes	20 (1.5)	3 (15.0)	17 (85.0)	
Had sex during/after using illicit drugs in the past 3 months				
No	1286 (93.5)	76 (5.9)	1210 (94.1)	1.00

Yes 89 (6.5) 5 (5.6) 84 (94.4)

Abbreviation: ATS, amphetamine-type stimulant; HIV, human immunodeficiency virus.

*Chi-square or Fisher's exact test was used as appropriate.

Factors associated with HIV infection

Table 5 presents independent factors associated with HIV infection in multivariable logistic regression analyses. After adjustment for other covariates, participants living in urban areas were twice as likely to be HIV infected as those living in rural areas (AOR= 2.7, 95% CI= 1.1-6.5). Participants with primary education were 1.7 times as likely to be infected compared to those with high school education (AOR=1.7, 95% CI= 1.0-2.9). HIV infection increased with age; compared to those aged 18-24, the odds of being HIV infected were twice as high among transgender women aged 25-34 (AOR= 2.1, 95% CI= 1.2-3.6) and 2.8 times higher among those aged ≥ 35 (AOR= 2.8, 95% CI= 1.3-6.1). Self-injection of gender affirming hormones was associated with a four-fold increase in the odds of HIV infection (AOR= 4.4, 95% CI= 1.1-17.3). A history of genital sores over the previous 12 months increased the odds of HIV infection by three-fold (AOR= 3.0, 95% CI= 1.2-7.8). Transgender women with stronger feminine identity, dressing as a woman all the time were twice as likely to be HIV infected compared to those who did not dress as a woman all the time (AOR= 2.1, 95% CI= 1.2-3.8). Having never used online services developed for transgender women in the past six months was also associated with higher odds of being HIV infected (AOR= 1.9, 95% CI= 1.2-3.2).

Table 5 Factors associated with HIV infection in multivariate logistic regression model

Variables in the final model	AOR (95% CI)	P-value
Community type		
Rural	Reference	
Urban	2.7 (1.1-6.5)	0.03
Formal education attained		

Primary (0–6 years)	1.7 (1.1-2.9)	0.04
High school or higher (≥7)	Reference	
Age in years		
<25	Reference	
25-34	2.1 (1.2-3.6)	0.01
≥35	2.6 (1.3-5.4)	0.01
Frequency of express and/or dressing as a woman		
Not all the time	Reference	
All the time	2.1 (1.2-3.8)	0.01
Ever self-injected hormone		
No	Reference	
Yes	4.4 (1.1-17.3)	0.03
Ulcerations or sores in the genital area in the past 12 months		
No	Reference	
Yes	3.0 (1.2-7.8)	0.02
Used online services developed MSM/TG in the past 6 months (e.g. Facebook, website)		
No	1.9 (1.2-3.2)	0.01
Yes	Reference	

379 *Abbreviations: AOR, adjusted odds ratio; CI, confidence interval; HIV, human immunodeficiency virus.*

380 **Variables in the table were the ones that remained statistically significant after several steps of model*

381 *fitting.*

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DISCUSSION

This study reveals an HIV prevalence of 5.9% among transgender women in Cambodia. This prevalence was about 20 times higher than the 0.3% among women attending antenatal care clinics in 2014 who represent the general adult population aged 15-45,³⁰ and about 2.5 times higher than the 2.3% among MSM in 2014.²⁶ This observed prevalence emphasizes that transgender women in Cambodia are at high risk of HIV, and is consistent with global literature regarding the high burden of HIV among this transgender population.¹

Although statistically non-significant ($p= 0.13$), the prevalence found in this study was higher than the prevalence of 4.2% reported in the smaller ($n= 891$) TGIBBS conducted in 2012²² and was therefore contrary to our hypothesized expectations of reduction in HIV prevalence among this population. Recent progress in Cambodia, where 73% of all estimated HIV positive people or 93% of those who have been aware of their positive status are in care,³¹ had prompted us to hypothesize the lower prevalence. Although we have no concrete evidence regarding the non-significant increase of HIV prevalence, we suggest that it may be related to the sampling variation in the two surveys. The previous study recruited participants only from the capital city and five provinces (Phnom Penh, Battambang, Banteay Meanchey, Kampong Cham, Siem Reap and Preah Sihanouk),²² whereas our study expanded to additional eight provinces.

More than half (52%) of those who had HIV reactive tests reported that they had never learned about their HIV positive status before they participated in the study. We believe that this self-reporting of the HIV status is realistic. Since we were concerned that those who were already aware of their HIV status may be unwilling to report their status, we decided to use well-trained counselors who all were from the government's Voluntary Confidential Counseling and Testing (VCCT) centers. These counselors had many years of experience in providing VCCT services and were better equipped to cope with difficult situations. In Cambodia, we believe people are more likely to trust their counselors, as opposed to interviewers. However, as in all self-reported measures, the potential for response bias cannot be entirely ruled out.

This study also reports important findings related to factors associated with HIV infection among transgender women in Cambodia. First, the HIV prevalence among transgender women residing in urban communities was twice as high when compared to that among

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2
3 412 transgender women living in rural communities. This finding is similar to that in a previous
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5 413 study that found a higher HIV prevalence in transgender women living in urban areas of Siem
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7 414 Reap and Phnom Penh, compared to that among transgender women living in other provinc-
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9 415 es.²² Findings from our study suggest that these differences may not necessarily be attributable
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11 416 to common sexual risk practices. The mean number of sexual partners (8.4 vs. 7.9; $p= 0.70$) and
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13 417 the mean number of male commercial sexual partners (0.7 vs. 0.7; $p= 0.48$) in the past three
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15 418 months among transgender women living in urban and rural communities were not significantly
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17 419 different. In addition, the rates of inconsistent condom use with male non-commercial sexual
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19 420 partners in the past three months were also similar in the two groups (58.0% vs. 62%; $p= 0.32$).

20 421 Given the failure of sexual risk behaviors to sufficiently account for the difference in HIV
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22 422 prevalence, these data suggest that a higher HIV prevalence among sexual partners of
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24 423 transgender women in urban communities may possibly explain the higher probability of urban
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26 424 transgender women to get infected, although this may not be stated with certainty. This is par-
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28 425 ticularly relevant given that previous studies among all transgender women found a higher
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30 426 prevalence of HIV in urban areas.²² Studies among MSM in Cambodia have also found a higher
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32 427 prevalence of HIV in urban areas.^{24 32} In addition, a recent national sentinel survey among
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34 428 women attending antenatal care clinics found that HIV prevalence among women in urban are-
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36 429 as was consistently high,³⁰ implying that HIV prevalence in urban areas among most popula-
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38 430 tions, who potentially include transgender women and their partners, is high. It is also possible
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40 431 that transgender population in urban communities face unique or additional risks compared to
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42 432 other members of this population in rural areas,²² which could also contribute to the difference
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44 433 in HIV prevalence between urban and rural communities. Still, it is possible that those who knew
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46 434 they were positive may have moved to urban centers for care and treatment, which unfortunately can-
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48 435 not be examined further in this cross-sectional study. Further research is needed with regard to this
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50 436 observation.

51 437 Second, HIV prevalence among older transgender women (≥ 25 years) was significantly
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53 438 higher, compared to that among their younger counterparts. This finding is similar to those of
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55 439 previous studies in Cambodia²² and Thailand.³ A possible explanation is that older transgender
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3 440 women had been exposed to risks for a longer period of time; the number of their sexual part-
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5 441 ners accumulated over time and thus increased cumulative probability of HIV acquisition.²²
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7 442 Third, our study suggests that more overt feminine expression (e.g., those who dressed
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9 443 as a woman all the time) was associated with a higher HIV prevalence. A study in the United
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11 444 States found that transgender women who disclosed their female gender identity possessed a
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13 445 higher HIV prevalence.³³ In our study, these transgender women who dressed as a woman all
14
15 446 the time had higher numbers of both male non-commercial (mean= 8.1 vs. 5.1; $p= 0.004$) and
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17 447 male commercial sexual partners (mean= 3.7 vs. 1.6; $p< 0.001$), compared to those who did not.
18
19 448 Making their female status visible could render more chances to meet with men. However, it
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21 449 could make them difficult to find a decent job due to stigmatization and discrimination, thus
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23 450 leading them to engage in sex work. This finding is in line with existing literature showing that
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25 451 transgender women try to have sex with men –both casual and paid sex– to prove or validate
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27 452 their female gender identity.^{34–36} In these contexts, “sex work provides both desired gender af-
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29 453 firmation and economic stability, often with greater financial rewards for sex without a con-
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31 454 dom.”³⁴ The desire to affirm their gender identity and attractiveness to men also incentivizes
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33 455 them to engage in casual sex with multiple partners.¹⁵ This sexual behavior may make them
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35 456 more prone to HIV infection.

36 457 There is a tendency to self-inject gender-affirming hormones, often with shared needles,
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38 458 among transgender women in Cambodia.³⁷ This risky practice was associated with an increased
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40 459 risk of HIV infection, implying that self-injecting was performed through unsafe means, and that
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42 460 injecting by a trained health or other professional might mitigate the likelihood of HIV infection.
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44 461 Although hormone injection to augment femininity is becoming more common, it can potential-
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46 462 ly cause adverse health effects among transgender women.^{3 38} A Thai study also found that
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48 463 transgender women who injected hormone to make them more feminine had a higher HIV
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50 464 prevalence.³

51 465 Fourth, transgender women who completed or dropped out primary school had a higher
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53 466 HIV prevalence than those with high-school education. This finding confirms results of the
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55 467 TGIBBS 2012 in Cambodia²² and other studies^{6 39} that associated low education with high HIV
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57 468 prevalence among transgender women. This has important implications for HIV programs to
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3 469 ensure that poorly educated transgender women are reached with education, information,
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5 470 communication and skills related to HIV prevention and other health-related services.
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7 471 Fifth, transgender women with self-reported STI symptoms (having ulcerations or sores
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9 472 in the genital area in the past 12 months) had a greater HIV prevalence. This is in line with exist-
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11 473 ing evidence showing that STIs promote HIV transmission via a variety of biological mecha-
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13 474 nisms.⁴⁰ Transgender women with high rates of STIs, particularly ulcerative genital diseases, are
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15 475 at high risk of HIV acquisition.²³

16 476 Finally, transgender women who reported never using online services developed for
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18 477 transgender women or MSM had an increased risk of HIV infection. Low utilization of online
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20 478 services by key populations in Cambodia is iterated in other studies.⁴¹ Non-users of online ser-
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22 479 vices, which tend to provide HIV information, education and communication, might have riskier
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24 480 sexual behaviors. Our data suggest that transgender women who did not use online services
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26 481 had higher numbers of overall male sexual partners (10.2 vs. 6.3, $p= 0.002$) and male commer-
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28 482 cial sexual partners (0.8 vs. 0.6, $p= 0.001$) in the past three months. Also, they had a higher rate
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30 483 of inconsistent condom use with male commercial sexual partners in the past three months
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32 484 (48.5% vs. 34.4%, $p= 0.004$). Unfortunately, in this study, we did not collect the details about
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34 485 the type of the online services, which could refer to dating, health and social services, or anothe-
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36 486 r type of online service. Studies in other settings confirm that transgender women with multi-
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38 487 ple sexual partners are exposed to a riskier level of HIV infection.^{6 15} This finding suggests that
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40 488 access to community-based services tailored to transgender population can reduce their vul-
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42 489 nerability to HIV as demonstrated in other settings.⁴² Sustaining and increasing the coverage of
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44 490 internet and peer-led community-based services for this population is particularly important,
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46 491 given that they are often unable to access health services due to stigmatization and discrimina-
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48 492 tion,^{16 41} even in health care settings.¹⁸ As in other settings globally,⁹ only half of those who
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50 493 were infected with HIV were aware of their status, demonstrating the need to intensify HIV
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52 494 testing, including self-testing at the community level, which transgender women in Cambodia
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54 495 are willing to use.⁴³ Our study also found that once diagnosed, the coverage of ART was high in
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56 496 this population.
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498 **LIMITATIONS OF THE STUDY**

499 Our study covered only the capital city and 12 provinces, which contain the highest numbers of
500 transgender women. Since cities and provinces with fewer TG were left out, these results may
501 not be generalized to all transgender women nationally. Second, the initial participant seeds
502 were identified and recruited by outreach workers of community-based organizations, which
503 could introduce bias towards transgender women under their programs, leading to a recruit-
504 ment bias. This problem could be exacerbated by outreach workers interviewing some partici-
505 pants who had received services from their NGO, which could have induced the participants'
506 responses. Third, this study employed a self-reporting questionnaire on sensitive health and
507 sexual behaviors, which may have been limited by social desirability bias. Fourth, albeit mini-
508 mal, the monetary incentive given to the participants to recruit seeds may have affected their
509 genuine motivation to partake in the study, which could influence their responses. Fifth, we
510 used the backward selection of variables in the multivariable analysis. By using this method, we
511 possibly dropped one or more variables that could be significant if we kept them until the final
512 model. Finally, as this study was cross-sectional, it reports associations at a given time, and may
513 not be construed to be reporting causal relationships.

514

515 **CONCLUSIONS**

516 This study demonstrates that HIV prevalence among transgender women in Cambodia remains
517 persistently high. With the prevalence rate at 5.9%, higher prevalence was observed among
518 transgender women who resided in urban areas, of older age, with low education levels, with
519 previous genital sores, with a history of self-injected hormone and those who had never used
520 online services developed for transgender women or MSM. Therefore, to eliminate new HIV
521 infections among transgender women, tailored interventions need to focus on these sub-
522 populations and attendant risk factors.

523

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15
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17
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19
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36 545 **Patient consent** Obtained

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39
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41
42 548 Health, Cambodia (No. 420 NECHR) and FHI 360's Protection of Human Subjects Committee
43
44 549 (PHSC No. 713897).

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48 551 **Consent for publication**
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50 552 Consent to publish was obtained from participants as stated in the informed consent form.

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54 554 **Data sharing statement** Data used for this study cannot be made available in the manuscript,
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56 555 the supplemental files or a public repository due to the ethical restriction stated in the agree-

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3 556 ment with the ethical committees. However, they can be accessed upon request from the Prin-
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5 557 cipal Investigator (Dr. Siyan Yi) at siyan@doctor.com.
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Participant ID Code:

Integrated Biological and Behavioral Survey of Transgender Women in Cambodia, 2016

Date of interview: day.....month.....year.....

Province/City:

- | | | |
|---------------------|---------------------|------------------|
| 1. Phnom Pehn | 6. Preah Sihanouk | 11. Kandal |
| 2. Kampong Cham | 7. Prey Veng | 12. Koh Kong |
| 3. Battambang | 8. Svay Rieng | 13. Thbong Khmum |
| 4. Siem Reap | 9. Kampong Spue | |
| 5. Banteay Meanchey | 10. Kampong Chhnang | |

Interview Location (City/NGOs):

Name of administrative districts (current residence):

Status of administrative districts:

- 1- Urban
- 2- Rural

SECTION 1: SOCIODEMOGRAPHIC CHARACTERISTICS			
No.	Questions and filters	Coding categories	Skip to
Now I would like to ask you some questions related to your personal information.			
Q001	How old are you?	Age in completed years:	
Q002	What is your current marital status? (only one response)	Married and living together 1 Married ,but not living together 2 Widowed, Divorced or separated 3 Not married, not living with any partner 4 Not married, living with sweetheart (female) 5 Not married but living with male lover 6 Other (Specify.....) 7	
Q003	For how long have you been living in the current city?Years <u>Record 0</u> if not living in this city <u>Record 1</u> if living in this city 1 year or less Refuse to answer 99	
Q004	In the past 6 months, on average, how much money	Amount of money (in US\$): Refuse to answer 99	

	did you make per month? (exchange rate: 4000 riel per 1 US\$)		
Q005	How many years of formal education have you completed?	Number of years: <u>Record 0</u> if never attending school. Refuse to answer 99	
Q006	What is your current job (main source of income)? (only one response) <i>Note: If you are both studying and having a paid job, report your main source of income.</i>	Unemployed 0 Hair dresser/Beautician 1 Government officer 2 Laborer (factory, construction work) 3 Seller 4 Entertainment Worker (bar, karaoke, etc.) 5 Sex worker 6 Student 7 NGO staff 8 Private company staff 9 Farmer/fisherman 10 Artist 11 Other (Specify.....) 12	

SECTION 2. TRANSGENDER IDENTITY & EXPERIENCES			
Q007	What gender do you consider yourself?	Female 1 Male 2 Third gender 3 Uncertain 4 Refuse to answer 99	
Q008	How often do you express or dress yourself as a woman?	All the time 1 Often 2 Sometimes 3 Rarely 4 Refuse to answer 99	
Q009	How many friends in your social network are also transgender?	Number:	
Q010	Have you ever taken hormones or any substances for beauty purposes?	No 1 Yes 2 Refuse to answer 99	2→ Q014
Q011	What are the methods you have used to take hormones or any substances for beauty purposes? [multiple answer]	Pills 1 Injections 2 Skin patches 3 Other (Specify.....) 4 Refuse to answer 99	
Q012	If you have taken hormones or any substances for beauty	Never injected 0 Injected by myself 1	

	purposes by injection, how have you received it?	Injected by skilled personnel (beauty clinic) 2 Injected by non-skilled personnel 3 Other (Specify.....) 4	
Q013	If you have injected hormones or any substances for beauty purposes, have you shared needle?	Never injected 0 No 1 Yes 2 Refuse to answer 99	
Q014	Have you ever had any operation to change any parts of your body to become a woman?	No 1 Yes 2 Refuse to answer 99	

Section 3. SEXUAL PARTNERS AND SEXUAL HISTORY			
No.	Questions and filters	Coding categories	Skip to
	Now I would like to ask questions about your recent sexual relationship with various kinds of sexual partners.		
Q015	Have you ever had sex with any partner in your life time? [Including vaginal, or anal sex]	No 0 Yes 1 Refuse to answer 99	
Q016	How likely do you think you are infected with HIV?	Very Likely 1 Likely 2 Unlikely 3 Very Unlikely 4 Refuse to answer 99	
Sex with women			
Q017	Have you ever had sex with a woman? [Including vaginal or anal sex]	No 0 Yes 1 Refuse to answer 99	0, 99 → Q028
Q018	In the past 12 months, have you had sex with a woman? [Including vaginal or anal sex]	No 0 Yes 1 Refuse to answer 99	0, 99 → Q028
Q019	In the past 12 months, have you had sex with a woman not in exchange for money or gifts? [Including vaginal or anal sex]	No 0 Yes 1 Refuse to answer 99	0→ Q022
Q020	In the past 3 month, how many women did you have	Number of women:	

Questionnaire; Integrated Biological and Behavioral Survey among Transgender Population in Cambodia, 2015, Version 2.0, 17 September 2015 Page 3 of 11

	sex with not in exchange for money or gifts?	Refuse to answer 99	
Q021	In the past 3 month, how often did you use a condom when you had vaginal or anal sex with women not in exchange for money or gifts?	Never had sex with a woman 0 Always 1 Often 2 Sometimes 3 Never 4 Refuse to answer 99	0, 1→ Q024
Q022	In the past 3 month, if you did not always use a condom when you had sex with women not in exchange for money or gifts? What were the reasons? [Multiple Answers]	We are in a relationship 1 She is not HIV/STI infected 2 Too high to use a condom 3 No condom available 4 Feel better without a condom 5 I am HIV-infected 6 I penetrated, so I am not at risk 7 Partner refused 8 Other (Specify.....) 9 Refuse to answer 99	
Q023	In the past 12 months, have you had sex with a woman in exchange for money or gifts? [Including vaginal or anal sex]	No 0 Yes 1 Refuse to answer 99	0→ Q028
Q024	In the past 3 months, how many women did you have sex with in exchange for money or gifts? [Including vaginal or anal sex]	Number of women: Refuse to answer 99	
Q025	Where did you meet women with whom you had sex with in exchange for money or gifts??	Park 1 Street 2 Bar/discotheque/cafe 3 Beer Garden/Restaurant 4 Massage Parlor 5 Karaoke 6 Online (Facebook, Line, Website) 7 Guesthouse/hotel 8 Workplace/school 9 Other (Specify.....) 10	
Q026	In the past 12 months, how often did you use a condom when you had vaginal or anal sex with women in exchange for money or gifts?	Never had sex 0 Always 1 Often 2 Sometimes 3 Never 4 Refuse to answer 99	

Q027	In the past 3 months, if you did not always use a condom when you had sex with women not in exchange for money or gifts? What were the reasons? [Multiple Answers]	We are in a relationship 1 She is not HIV/STI infected 2 Too high to use a condom 3 No condom available 4 Feel better without a condom 5 I am HIV-infected 6 I penetrated, so I am not at risk 7 Partner refused 8 Other (Specify.....) 9 Refuse to answer 99	
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Sex with men			
Q028	Have you ever had anal sex with a man?	No 0 Yes 1 Refuse to answer 99	0, 99→ Q042
Q029	In the past 12 months, have you had anal sex with a man?	No 0 Yes 1 Refuse to answer 99	0, 99→ Q042
Q030	In the past 12 months, what was your usual role in anal sex with a man?	Insertive 1 Receptive 2 Both 3 Refuse to answer 99	
Q031	In the past 3 months, have you had sex with men not in exchange for money or gifts?	No 0 Yes 1 Refuse to answer 99	0→ Q033
Q032	In the past 3 months, how many men you had sex with not in exchange for money or gifts?	Number of men: Refuse to answer 99	
Q033	In the past 12 months, how often did you use a condom when you had anal sex with men not in exchange for money or gifts?	Never had sex 0 Always 1 Often 2 Sometimes 3 Never 4 Refuse to answer 99	0, 1→ Q035
Q034	If not always, why not? [Multiple Answers]	We are in a relationship 1 He is not HIV/STI infected 2 I was too drunk or too high with drugs 3 No condom available 4 Feel better without a condom 5 I am HIV-infected 6 I penetrated, so I am not at risk 7 Partner refused 8 Other (Specify.....) 9 Refuse to answer 99	

Q035	In the past 12 months, have you had sex with men in exchange for money or gifts?	No 0 Yes 1 Refuse to answer 99	0→ Q040
Q036	In the past 3 month, how many men you had sex with in exchange for money or gifts?	Number of men: Refuse to answer 99	
Q037	Where did you meet the men you had sex with in exchange for money or gifts?	Park 1 Street 2 Bar/discotheque/cafe 3 Beer Garden/Restaurant 4 Massage Parlor 5 Karaoke 6 Online (Facebook, Line, Website) 7 Guesthouse/hotel 8 Workplace/School 9 Other (Specify.....) 10	
Q038	In the past 12 months, how often did you use a condom when you had anal sex with men you paid for sex?	Never had anal sex 0 Always 1 Often 2 Sometimes 3 Never 4 Refuse to answer 99	0, 1→ Q615
Q039	If not always, why not? [Multiple Answers]	We are in a relationship 1 He is not HIV/STI infected 2 I was drunk or too high with drugs 3 No condom available 4 Feel better without a condom 5 I am HIV-infected 6 I penetrated, so I am not at risk 7 Partner refused 8 Other (Specify.....) 10 Refuse to answer 99	

Access to condoms and lubricant			
No.	Questions and filters	Coding categories	Skip to
Q040	In this last 12 months, where did you get condoms? [Multiple Answers]	Friends/outreach workers 1 Condom peer sale representative 2 Pharmacy/drug store/clinic 3 Condom outlets 4 Mart/mini-mart 5 Groceries 6 Hotel/guesthouse 7 Other (Specify.....) 8	

		Refuse to answer 99	
Q041	In this last 12 months, where did you get lubricant? [Multiple Answers]	Friends/outreach workers 1 Condom peer sale representative 2 Pharmacy/drug store/clinic 3 Condom outlets 4 Mart/mini-mart 5 Groceries 6 Hotel/guesthouse 7 Other (Specify.....) 8 Refuse to answer 99	

SECTION 4. STI AND HIV TESTING			
No.	Questions and filters	Coding categories	Skip to
Q042	In the past 12 months, have you experienced the following symptoms? [Multiple Answers]	Cuts or sores in the genital area 1 Swelling in the genital area 2 Abnormal urethral discharge 3 Symptoms on the anus 4 Symptom in the mouth/throat 5 Refuse to answer 99	
Q043	Where did you first go for treatment the last time you had any above-mentioned symptoms? (Only one response)	Never had any symptoms 0 Pharmacy 1 Private clinic/hospital 2 Public Hospital/STD clinic 3 NGO Clinic/hospital 4 Traditional doctor 5 Didn't get care 6 Other (Specify.....) 7 Refuse to answer 99	
Q044	Have you ever been tested for HIV?	No 0 Yes 1 Refuse to answer 99	0→ Q049
Q045	In past 6 months, have you been tested for HIV?	No 0 Yes 1 Refuse to answer 99	
Q046	How long has it been since you received the last HIV test?	Number of months.....	
Q047	If a rapid self-test kit for HIV was available and you could get the result within 20 minutes at home, would you use it?	No 0 Yes 1 Refuse to answer 99	

1 2 3 4 5 6 7 8 9	Q048	Where did you have your most recent HIV test?	Private facilities 1 Public facilities 2 NGO facilities 3 NGO outreach workers at community or HTC 4 Other (Specify.....) 5 Refuse to answer 99	
10 11 12 13	Q049	The last time you got tested for HIV, did you receive the result of the test?	No 0 Yes 1 Refuse to answer 99	
14 15 16 17	Q050	What is your current HIV status? (You could choose not to answer)	Positive 1 Negative 2 I don't know my status 3 Refuse to answer 99	2, 3 → Q057
18 19 20 21 22 23 24 25	Q051	Are you currently on ART?	No 0 Yes 1 I was but discontinued 2 I am on treatment for opportunistic infections 3 Refuse to answer 99	0 → Q056
26 27 28 29	Q052	Do you take ARV regularly as prescribed?	Regularly 1 Not regularly 2 Refuse to answer 99	
30 31 32 33 34 35 36	Q053	Where do you get ART services?	Public facilities 1 Private facilities 2 NGO facilities 3 Pharmacy 4 Other (Specify.....) 5 Refuse to answer 99	
37 38 39 40 41 42 43 44 45	Q054	If not, why are you not on ART?	Not needed 1 Not sure where to go 2 I am ashamed to go to health facility 3 I am afraid of being discriminated 4 I was treated badly in the past 5 Other (Specify.....) 6 Refuse to answer 99	

SECTION 5. ALCOHOL AND DRUG USE

Measurement of alcohol drinking			
One standard measurement: - A can/glass of beer, fermented palm juice (285 ml) - A glass of wine (120 ml) - A glass of whisky (30 ml)			
No.	Questions and filters	Coding categories	Skip to
Q055	In the past 3 months, how often did you drink at least one can of beer or one glass of any types of wine?	Never 1 Once a month or less 2 2-4 times a month 3 2-3 times a week 4 4 or more times a week 5 Don't know 98 Refuse to answer 99	1→ Q064
Q056	In the past 3 months, how many standard drinks containing alcohol (a can of beer or a glass of any types of wine) did you have on a typical day on which you drank alcohol?	1 - 2 1 3 - 4 2 5 - 6 3 7 - 9 4 10 or more 5 Don't know 98 Refuse to answer 99	
Q057	In the past 3 months, how often did you have more than 5 drinks in one day or night?	Never 1 Less than once a month 2 Once a month 3 1, 2, or 3 times a week 4 4 or more times a week 5 Don't know 98 Refuse to answer 99	
Q058	In the past 3 months, how often have you got drunk from alcohol?	Never 1 Less than once a month 2 Once a month 3 Every week (1-3 times/week) 4 Every day/almost every day(4 or more times a week) 5 Don't know 98 Refuse to answer 99	

Q059	In the past 12 months, have you tried any of the following drugs? (Ask one by one - CIRCLE YES OR NO) [Multiple answers]			
		Types of drug	Yes	No
	1	Marijuana	1	0
	2	Heroin/Opium	1	0
	3	Yama (amphetamine)	1	0
	4	Crystal, Ice (Methamphetamine)	1	0
	5	Ecstasy	1	0
	6	Inhalants (glue, paint, petrol, spray can)	1	0
	7	Other	1	0
Q060	In the past 3 months, did you inject any illicit drugs?			
		Heroin	1	
		Yama	2	
		Crystal, Ice (Methamphetamine)	3	
		Never injected any drug	4	
	Refuse to answer	99		
Q061	In the past 3 months, did you have sex during/after using illicit drugs?			
		Heroin	1	
		Yama	2	
		Ice, Amphetamine	3	
		Never used drug before having sex	4	
	Refuse to answer	99		

SECTION 9. EXPOSURE TO INTERVENTION PROGRAMS			
No.	Questions and filters	Coding categories	Skip to
Q062	In the past 3 months, have you been reached by NGO staff delivering HIV services?	No 0 Yes 1 Don't know 98 Refuse to answer 99	
Q063	In the past 3 months, what kind of services have you received (Check all that apply).	HIV/health education and/or materials 1 Condoms 2 Lubricants 3 VCCT/STI testing 4 Law support services 5 Other health services 6 Other (Specify.....) 7 Refuse to answer 99	
Q064	Do you have UIC card?	Yes 1 No 0	

		Refuse to answer 99	
Q065	In the past 12 months, how often did you visit MStyle/Srey Sros club(s)?	... times Refuse to answer 99	
Q066	Have you used online services developed for people like you (e.g. Facebook, website)?	No 0 Yes 1 Refuse to answer 99	0 -> stop interview
Q067	Have you found any sexual partner on an Internet website or any online program?	Never 0 Yes, always 1 Yes, sometimes 2 Yes, occasionally 3 Refuse to answer 99	
Q068	Have you accessed MStyle website in the past 6 months? (Attach the picture of MStyle webpage)	Never 0 Yes, always 1 Yes, sometimes 2 Yes, occasionally 3 Refuse to answer 99	

["Thank you very much for answering these questions. Let us repeat that your answers are totally confidential and there is no way anyone will learn what you told us.]

STROBE Statement—Checklist of items that should be included in reports of *cross-sectional studies*

	Item No	Recommendation
Title and abstract	1	(a) Indicate the study's design with a commonly used term in the title or the abstract Confirmed (Lines 1-53) (b) Provide in the abstract an informative and balanced summary of what was done and what was found. Confirmed (Lines 30-53)
Introduction		
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported. Confirmed (Lines 75-122)
Objectives	3	State specific objectives, including any prespecified hypotheses. Confirmed (Lines 123-126)
Methods		
Study design	4	Present key elements of study design early in the paper. Confirmed (Line 128-135)
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection. Confirmed (Line 128-135)
Participants	6	(a) Give the eligibility criteria, and the sources and methods of selection of participants. Confirmed (Lines 137-186)
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable. Confirmed (Lines 195-228)
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group. Confirmed (Lines 195-228)
Bias	9	Describe any efforts to address potential sources of bias. Confirmed (Lines 188-193)
Study size	10	Explain how the study size was arrived at. Confirmed (138-157)
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why. Confirmed (Lines 231-237)
Statistical methods	12	(a) Describe all statistical methods, including those used to control for confounding. Confirmed (Lines 237-245) (b) Describe any methods used to examine subgroups and interactions. (Not applicable) (c) Explain how missing data were addressed (Not applicable) (d) If applicable, describe analytical methods taking account of sampling strategy. (Not applicable) (e) Describe any sensitivity analyses. (Not applicable)
Results		
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed. Confirmed (Lines 259-262) (b) Give reasons for non-participation at each stage (c) Consider use of a flow diagram
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders. Confirmed (Lines 264-275) (b) Indicate number of participants with missing data for each variable of interest. (Not applicable)
Outcome data	15*	Report numbers of outcome events or summary measures. Confirmed (259-260)
Main results	16	(a) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and

their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included. [Confirmed \(276-289, 309-313, 329-334, 347-351\)](#)

(b) Report category boundaries when continuous variables were categorized. [\(Not applicable\)](#)

(c) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period. [\(Not applicable\)](#)

Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses. Confirmed (320-325)
Discussion		
Key results	18	Summarise key results with reference to study objectives. Confirmed (Lines 381-493)
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias. Confirmed (Lines 495-510)
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence. Confirmed (Lines 513-519)
Generalisability	21	Discuss the generalisability (external validity) of the study results. Confirmed (Lines 496-502)
Other information		
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based. Confirmed (Lines 535-537)

*Give information separately for exposed and unexposed groups.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at <http://www.plosmedicine.org/>, Annals of Internal Medicine at <http://www.annals.org/>, and Epidemiology at <http://www.epidem.com/>). Information on the STROBE Initiative is available at www.strobe-statement.org.