

Supplementary Table 1. Correlation between cervical and serum biomarkers as continuous variables adjusted by repeated measures among participants. The correlation matrix shows estimated Spearman rank-order correlation coefficients (top of the grid) with p-values (bottom of the grid) calculated by t-distributions. The coefficient indicates the degree of correlation which is ranked weak if a coefficient is between -0.2 and 0.2 and strong if <-0.7, >0.7). The p-value indicates the chance of zero correlation coefficient between two biomarkers.

	Cervical Biomarkers										Serum Biomarkers			
	BD2	SLPI	IL-1RA	IL-1 β	IL-6	IL-8	MIP-3 α	RANTES	ICAM-1	VEGF	sCD14	CRP	IL-6	IL-7
BD2		0.0272	0.25733	0.19236	0.13071	0.2556	0.22339	-0.205	-0.1403	0.15259	0.04608	-0.0105	-0.0473	-0.004
		0.1881	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0257	0.6104	0.0221	0.8479
SLPI			0.23074	0.30286	0.46831	0.44998	0.56425	0.0763	0.2796	0.39932	-0.1711	0.13425	-0.1447	-0.0591
			<.0001	<.0001	<.0001	<.0001	<.0001	0.0002	<.0001	<.0001	<.0001	<.0001	<.0001	0.0042
IL-1RA				0.34591	0.29607	0.24634	0.36045	-0.1206	0.14265	0.39848	0.00271	0.00915	-0.0053	0.04161
				<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.8958	0.6581	0.7993	0.044
IL-1 β					0.52279	0.62527	0.46096	0.01093	0.22328	0.5375	-0.1061	0.09958	-0.0095	0.02462
					<.0001	<.0001	<.0001	0.5968	<.0001	<.0001	<.0001	<.0001	0.6461	0.2334
IL-6						0.69766	0.64149	0.10954	0.33106	0.62496	-0.1362	0.13037	-0.1042	-0.0417
						<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0434
IL-8							0.62738	0.04736	0.20064	0.65417	-0.1188	0.12726	-0.0727	-0.0024
							<.0001	0.0218	<.0001	<.0001	<.0001	<.0001	0.0004	0.9077
MIP-3 α								0.11866	0.30056	0.49819	-0.109	0.11568	-0.111	-0.0135
								<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.5149
RANTES									0.53644	0.00471	0.00933	-0.0402	-0.0707	-0.0986
									<.0001	0.8197	0.6517	0.0517	0.0006	<.0001
ICAM-1										0.23479	-0.0613	0.01135	-0.0217	-0.037
										<.0001	0.003	0.5829	0.2946	0.0732
VEGF											-0.0831	0.10944	-0.0499	0.00777
											<.0001	<.0001	0.0156	0.7069
sCD14												-0.0152	0.16132	0.09685
												0.4613	<.0001	<.0001
CRP													0.15961	0.09542
													<.0001	<.0001
IL-6														0.20992
														<.0001
IL-7														

Supplementary **Table 2.** Characteristics of participant visits by HSV-2 status among HIV negative visits

Characteristics	HSV-2 negative ¹ visits (n=1505)	HSV-2 incident ² visits (n=633)	HSV-2 established ³ visits (n=978)	Total (n=3116)	P-value
	n (%)	n (%)	n (%)	n (%)	
Site					
Uganda	518 (34.42)	318 (50.24)	294 (30.06)	1130 (36.26)	
Zimbabwe	987 (65.58)	315 (49.76)	684 (69.94)	1986 (63.74)	<.001
Age at screening					
18-24	989 (65.71)	383 (60.51)	469 (47.96)	1841 (59.08)	
25+	516 (34.29)	250 (39.49)	509 (52.04)	1275 (40.92)	<.001
HC group					
COC	535 (35.55)	230 (36.33)	398 (40.74)	1163 (37.34)	
DMPA	545 (36.21)	179 (28.28)	280 (28.66)	1004 (32.23)	
NH	425 (28.24)	224 (35.39)	299 (30.6)	948 (30.43)	<.001
Currently pregnant	111 (7.4)	42 (6.68)	99 (10.16)	252 (8.12)	<.001
Currently Breastfeeding	243 (16.15)	98 (15.51)	118 (12.07)	459 (14.74)	<.001
2+ sexual partners	29 (1.93)	11 (1.74)	12 (1.23)	52 (1.67)	<.001
Current smoking	3 (0.2)	1 (0.16)	4 (0.41)	8 (0.26)	<.001
# unprotected acts					
15+	339 (22.52)	133 (21.01)	234 (23.93)	706 (22.66)	
8-14	390 (25.91)	181 (28.59)	237 (24.23)	808 (25.93)	
1-7	401 (26.64)	131 (20.7)	210 (21.47)	742 (23.81)	
0 or no sex act	375 (24.92)	188 (29.7)	297 (30.37)	860 (27.6)	<.001
Any RTI/STI	593 (40.9)	633 (100)	978 (100)	2204 (72)	<.001
BV (Nugent score 7-10)	391 (27.08)	166 (27.35)	272 (29.76)	829 (27.96)	<.001
Candidiasis	169 (11.3)	66 (10.59)	100 (10.49)	335 (10.91)	<.001
Chlamydia	32 (2.15)	19 (3.09)	10 (1.06)	61 (2)	<.001
Gonorrhoea	33 (2.22)	19 (3.09)	13 (1.37)	65 (2.13)	<.001
Trichomonas	42 (2.83)	22 (3.57)	17 (1.81)	81 (2.66)	<.001

¹ HSV-2 negative visits were contributed by 382 women remaining HSV-2 negative (1157 visits) and 334 women that HSV-2 seroconverted during the study contributed (348 negative visits occurring at least 3 months before the HSV-2 seroconversion visit).

²The 633 visits defined as 'HSV-2 incident visits' were contributed by 334 women who passed inclusion criteria and included all available visits by these women that occurred at and <6 months (180 days) after the first HSV-2 positive test.

³The 978 visits defined as 'HSV-2 established' were contributed by: 1) 303 women who were HSV-2 positive at enrollment (528 visits available at least 6 months from the enrollment visit), and 2) 334 women who acquired HSV-2 during the study (450 visits available at ≥ 6 months from the HSV-2 seroconversion visit).

Supplementary Table 3. Estimated odds ratios (OR) of HSV-2 acquisition for levels of concomitantly imbalanced systemic and cervical Immunity measured at the HSV-2 negative visits from individuals remaining HSV-2 negative and the HSV-2 seroconverters at the visit prior to the incident visit.

Systemic + Cervical Biomarker Groups	#↑↓ ¹ / #Sample used ²	OR ³ (95% CI)	P-value
Concomitant serum sCD14 ↑ + cervical ↑↓			
IL-1β↑, IL-6↑	48 / 703	0.39 (0.14, 1.11)	0.077
IL-8↑, MIP-3α↑	52 / 703	0.68 (0.30, 1.55)	0.360
VEGF↓, ICAM-1↓	47 / 703	2.02 (1.04, 3.90)	0.037
SLPI↓	84 / 703	1.85 (1.09, 3.12)	0.022
BD-2↑	72 / 702	2.18 (1.26, 3.75)	0.005
IL-1RA↓	67 / 703	1.34 (0.72, 2.46)	0.354
RANTES↑	87 / 703	1.30 (0.75, 2.25)	0.351
Concomitant serum CRP ↓ + cervical ↑↓			
IL-1β↑, IL-6↑	114 / 703	0.75 (0.43, 1.32)	0.322
IL-8↑, MIP-3α↑	125 / 703	1.15 (0.71, 1.88)	0.568
VEGF↓, ICAM-1↓	96 / 703	0.96 (0.55, 1.69)	0.892
SLPI↓	169 / 703	1.36 (0.89, 2.10)	0.156
BD-2↑	168 / 702	1.82 (1.20, 2.76)	0.005
IL-1RA↓	179 / 703	1.18 (0.77, 1.82)	0.446
RANTES↑	207 / 703	1.21 (0.80, 1.83)	0.357
Concomitant serum IL6 ↑ + cervical ↑↓			
IL-1β↑, IL-6↑	121 / 703	0.44 (0.24, 0.83)	0.012
IL-8↑, MIP-3α↑	132 / 703	0.88 (0.53, 1.45)	0.612
VEGF↓, ICAM-1↓	89 / 703	1.94 (1.16, 3.23)	0.011
SLPI↓	164 / 703	1.65 (1.08, 2.53)	0.020
BD-2↑	156 / 702	1.41 (0.91, 2.18)	0.125
IL-1RA↓	156 / 703	1.41 (0.91, 2.19)	0.123
RANTES↑	174 / 703	1.07 (0.69, 1.66)	0.766

¹ ↑ indicates activated (levels above median) and ↓ indicates suppressed (levels below median) immunity, with the exception of sCD14 which is activated if its concentration is above top quartile, for all HIV-1 and HSV-2 negative visits.

² HSV-2 negative visits from individuals remaining HSV-2 negative and HSV-2 seroconverters at the visit prior to the incident visit.

³ Odds ratios (OR) and 95% confidence intervals (CI) estimated likelihood of HSV-2 seroconversion with biomarker levels activated/suppressed as indicated by arrows assessed concomitantly.