Supplemental Information

Supplementary Materials and Methods

Production of lentivirus-packed target genes

293T cells (2×10^6) were transfected using Lipofectamine 2000 (Life Technologies) with 0.45 µg pCMV- Δ R8.91, 0.05 µg pMD.G, and 0.5 µg MRJ-L shRNA vector or a MRJ-L expression plasmid to generate pseudotyped lentiviruses carrying the corresponding genes. Cellular supernatants were harvested at 72 h post-transfection. Viral titer was determined using the protocol provided by the RNAi Core Facility, Academia Sinica, Taiwan (<u>http://rnai.genmed.sinica.edu.tw/protocols</u>).

Fluorescence Confocal Imaging

HeLa cells were co-transfected with pEGFP-Vpr and pcDNA-HA-MRJ expression vectors on glass cover slides. After 24 h, cells were fixed with ice-cold acetone for 10 min and stained with anti-HA antibody (Roche, Palo Alto, CA, USA) for 1 h, washed three times with PBS, and subsequently incubated with AlexaFluor-conjugated anti-mouse antibody for 30 min. After washing two times with PBS, cells were stained with 3 μM 4', 6'-diamidino-2-phenylindole (DAPI) at room temperature for 5 min and

observed under confocal microscopy (Leica TCS SP2).

Supplementary Tables

Supplementary Table 1

Demographic data of HIV-1 positive and negative participants from commercial MSM community.

	HIV-1 (+)	HIV-1 (-)	Total	Р
Variable	N=20	N=30	N=50	value
	n(%)	n(%)	n(%)	
Age				0.079
<30	12 (60)	19 (63)	31 (62)	
≥30	8 (40)	6 (20)	14 (28)	
NA	0 (0)	5 (17)	5 (10)	
Sexual orientation				0.440
Heterosexual	1 (5)	1 (3)	2 (4)	
Homosexual	18 (90)	29 (97)	47 (94)	
Bisexual	1 (5)	0 (0)	1 (2)	
NA	0 (0)	0 (0)	0 (0)	
Marital status				0.594
Single	17 (85)	27 (90)	44 (88)	
Married	0 (0)	0 (0)	0 (0)	
Divorced/Separated/Widowed	3 (15)	3 (10)	6 (12)	
Education				0.768
<junior high="" school<="" td=""><td>0 (0)</td><td>0 (0)</td><td>0 (0)</td><td></td></junior>	0 (0)	0 (0)	0 (0)	
≥Senior high school	19 (95)	29 (97)	48 (96)	
NA	1 (5)	1 (3)	2 (4)	
Occupation				0.464

Student	4 (20)	8 (27)	12 (24)	
Government employees	5 (25)	3 (10)	8 (16)	
Office worker	7 (35)	13 (43)	20 (40)	
Professional	2 (10)	2 (7)	4 (8)	
Unemployed/ Other	1 (5)	4 (13)	5 (10)	
NA	1 (5)	0 (0)	1 (2)	
Number of sexual partners				0.116
1	5 (25)	5 (17)	10 (20)	
2-3	9 (45)	22 (73)	31 (62)	
≥ 4	1 (5)	2 (7)	3 (6)	
NA	3 (15)	0 (0)	3 (6)	
Role and protection during anal				
intercourse				0.07
Exclusively insertive	2	4 (13)	4 (8)	
with condom	0			
Exclusively insertive	2		3 (6)	
without regular condom	0	3 (10)		
Exclusively receptive /			15 (30)	
Versatile with condom	6 (30)	9 (30)		
Exclusively receptive /				
Versatile without	12 (60)	10 (33)	22 (44)	
regular condom				
Oral sex	0 (0)	3 (10)	3 (6)	

NA	2 (10)	1 (3)	3 (6)		
Times of sexual contact per week				0.364	
<1	3 (15)	9 (30)	12 (24)		
1-3	7 (35)	12 (40)	19 (38)		
>3	4 (20)	2 (7)	6 (12)		
NA	6 (30)	7 (23)	13 (26)		
Recreational drug usage				0.451	
No	9 (45)	17 (57)	26 (52)		
Yes	8 (40)	7 (23)	15 (30)		
NA	3 (15)	6 (20)	9 (18)		
Oil- or petroleum-based lubricants during sexual intercourse					
Saliva	8 (40)	14 (47)	22 (44)		
Oil-based ^a	6 (30)	5 (17)	11 (22)		
NA	6 (30)	11 (37)	17 (34)		
History of having sexually transmitted diseases					
No	8 (40)	24 (80)	32 (64)		
Yes	2 (10)	0 (0)	2 (4)		
NA	10 (50)	6 (20)	16 (32)		
Frequency of lubricant usage				0.719	
Always	14 (70)	24 (80)	38 (76)		
Frequently/Occasionally/	2 (15)	2 (10)			
Rarely/Never	3 (15)	3 (10)	6 (12)		
NA	3 (15)	3 (10)	6 (12)		

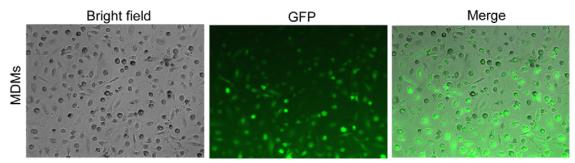
NA, not available.

Supplementary Table 2

Risk factors for HIV-1 infection were analyzed by using Bayesian multiple logistic regression.

	Odds Ratio	2.5% credible interval	97.5% credible interval
MRJ levels			
MRJ Low	1		
MRJ Medium	7.601	1.37	69.25
MRJ High	41.57	4.285	705.7
Role of sexual behavior			
Insertive anal sex	1		
Receptive anal sex	43.99	2.681	2957
Oral sex	0.2145	8.58E-05	73.13

Supplementary Figure 1



Supplementary Fig. 1. Lentiviral transduction efficiency in primary MDMs. MDMs at day 7 of differentiation were transduced with lentivirus packed with GFP mRNA. The transduction efficiency was about 50-70% as measured by the percentage of cells expressing GFP.