Dynamics of immunoglobulin sequence diversity in HIV-1 infected individuals

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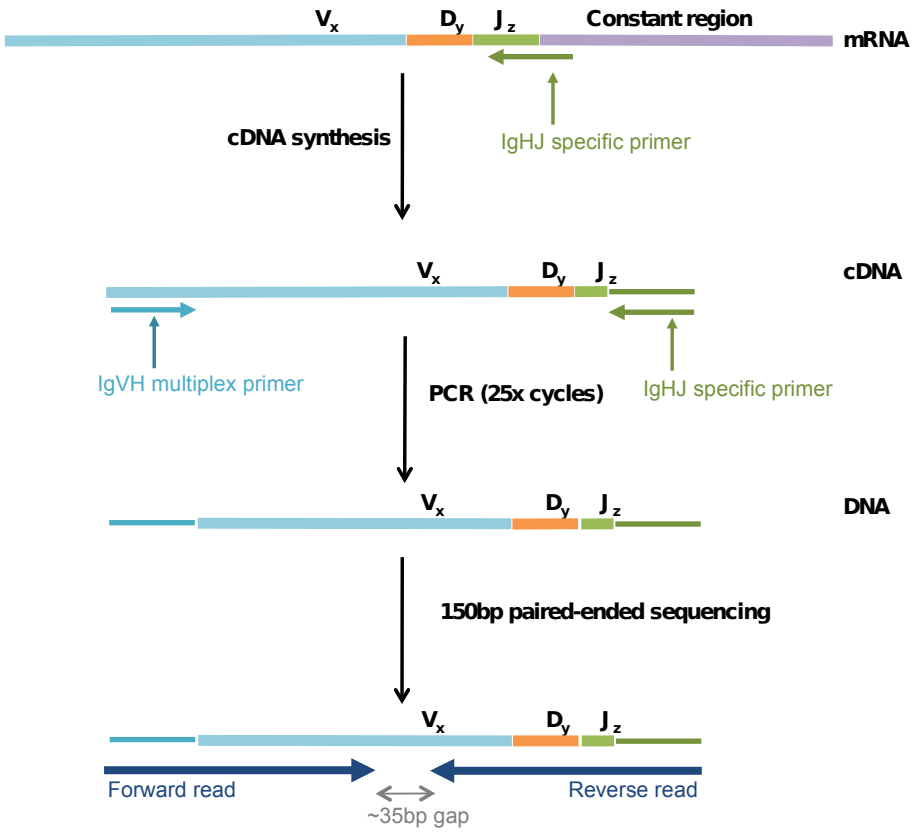
Supplemental information

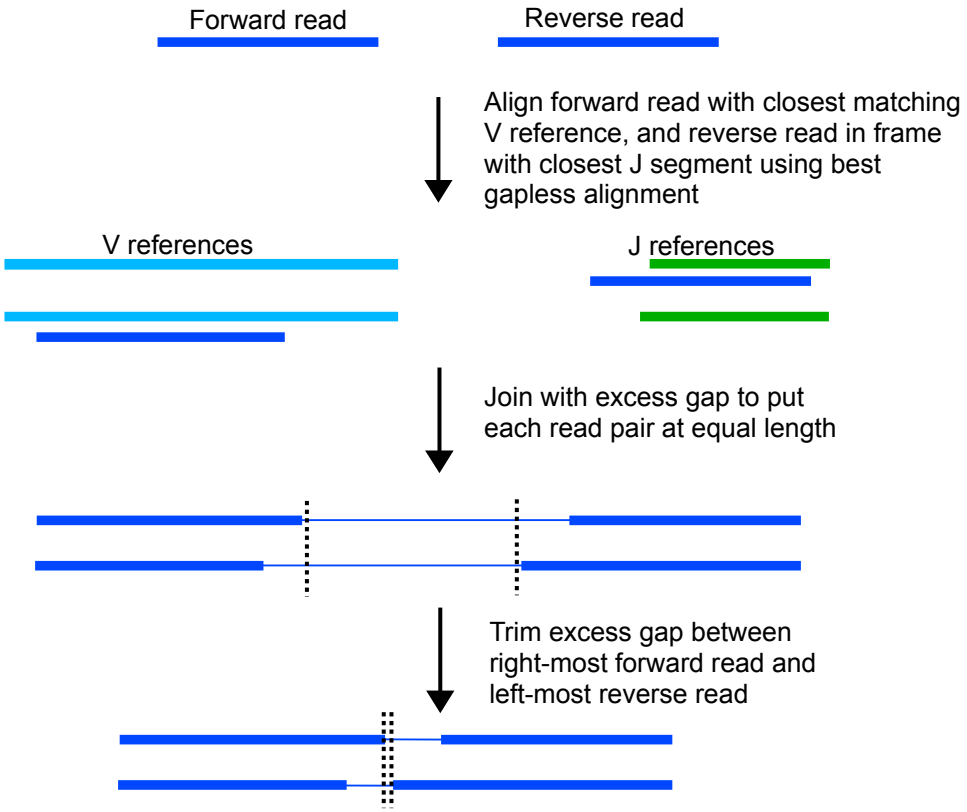
S1: Human B-cell receptor PCR primers

Primer	**Sequence**	
JH reverse	CTTACCTGAGGAGACGGTGACC	
VH1-FR1 forward	GGCCTCAGTGAAGGTCTCCTGCAAG	
VH2-FR1 forward	GTCTGGTCCTACGCTGGTGAAACCC	
VH3-FR1 forward	CTGGGGGGTCCCTGAGACTCTCCTG	FR1 primer set*
VH4-FR1 forward	CTTCGGAGACCCTGTCCCTCACCTG	
VH5-FR1 forward	CGGGGAGTCTCTGAACATCTCCTGT	
VH6-FR1 forward	TCGCAGACCCTCTCACTCACCTGTG	

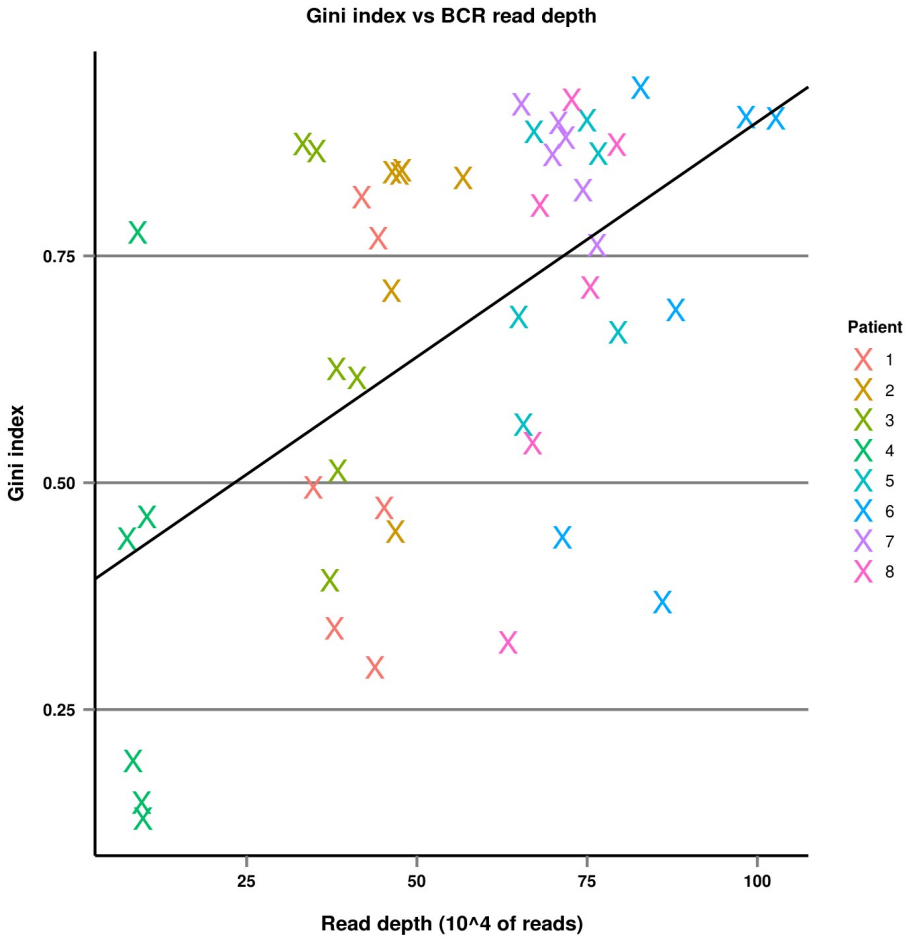
S2: Illustration of the BCR sequencing pipeline



S3: Illustration of the sequence alignment pipeline



S4: Relationship between Gini index and read depth between patients



S4: Each point represents a sampling time point and is coloured by patient (see key). The fitted linear regression has a slope of 5.2×10^{-3} ($p = 5.69 \times 10^{-5}$) and a correlation coefficient of 0.55. This relationship remained significant when patient means were used to correct for temporal autocorrelation ($p < 0.03$).

S5: Gini index and read depth

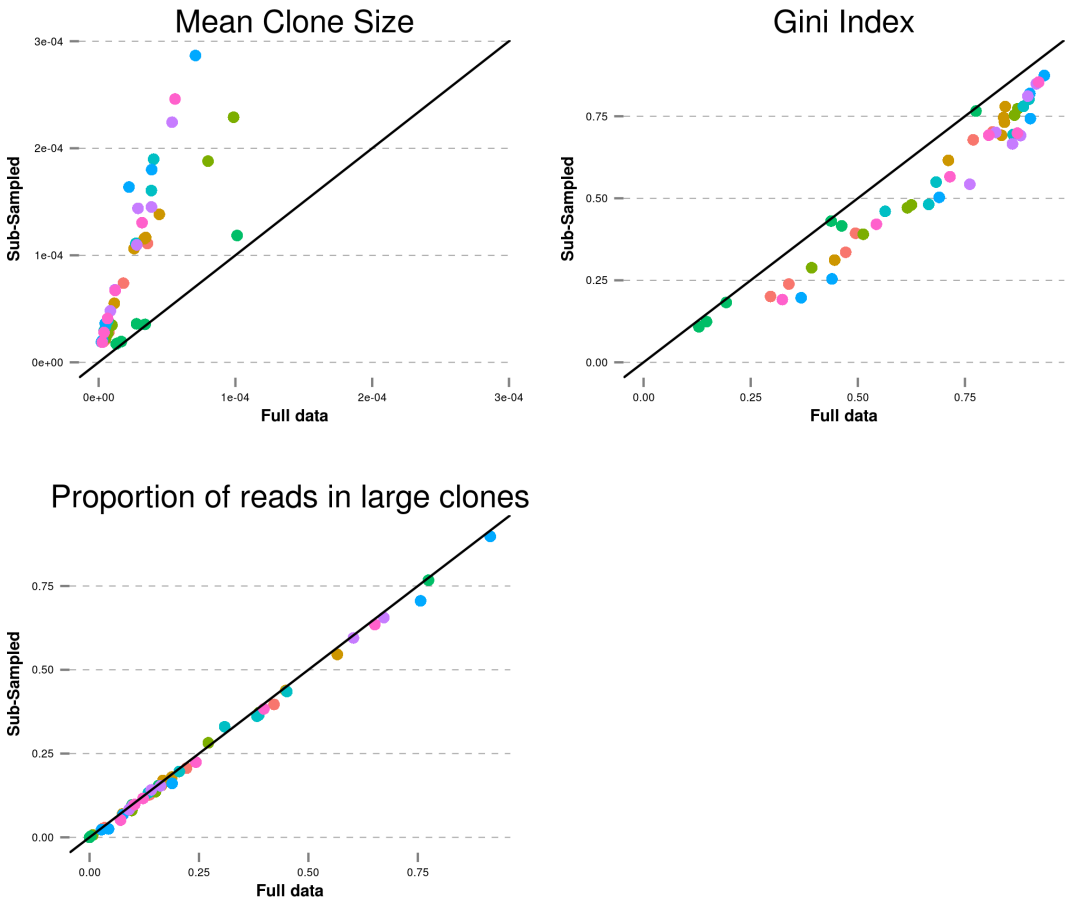
We model the effect of read depth on Gini index using a simple population of B cells consisting of a single dominant clone – occupying one third of all B cells – and a large pool of unique B cells, which comprise the rest of the system. As sequencing depth increases, one third of each sample joins the dominant clone, which grows larger. However, all other clones remain unique, leading to a larger population of singleton clones. This means the ratio of reads in the dominant clone to each of the singletons grows with sampling depth, leading to a more skewed size distribution and higher Gini index. Specific values are provided in the table below.

Depth (total reads)	Largest clone (reads)	No. of Singletons	Gini index
15	5	10	0.24
30	10	20	0.29
45	15	30	0.30
60	20	40	0.31
75	25	50	0.31

S6: CD4+ cell counts and seroconversion information

		CD4+ counts					
Patient	*Estimated days since seroconversion at week 0*	*Week 4*	*Week 16*	*Week 24*	*Week 52*	*Week 60*	*Week 108*
1	95	470	NA	440	480	50	470
2	34	NA	430	460	470	410	NA
3	12	550	690	630	530	440	NA
4	76	1610	1230	1180	1200	870	1460
5	93	830	620	850	500	650	510
6	68	490	550	480	600	740	500
7	35	590	570	660	470	360	NA
8	44	770	390	NA	660	320	NA

S7: Effect of sub-sampling on BCR statistics



S7: BCR statistics of HIV+ patients calculated from full data versus data sub-sampled data. Read depth of full data had a median of 567,936 and a range of 74,861 to 1,040,936 reads per patient per time point. Sub-sampled data were randomly sub-sampled to a depth of 70,000 reads per patient per time point. Dots are coloured by patient.