SUPPLEMENTARY APPENDIX TO

Influence of B cell or Plasma Cell-targeted CAR-T Cell Therapy on Humoral Immunity and Vaccine Immunogenicity: The HICAR Study

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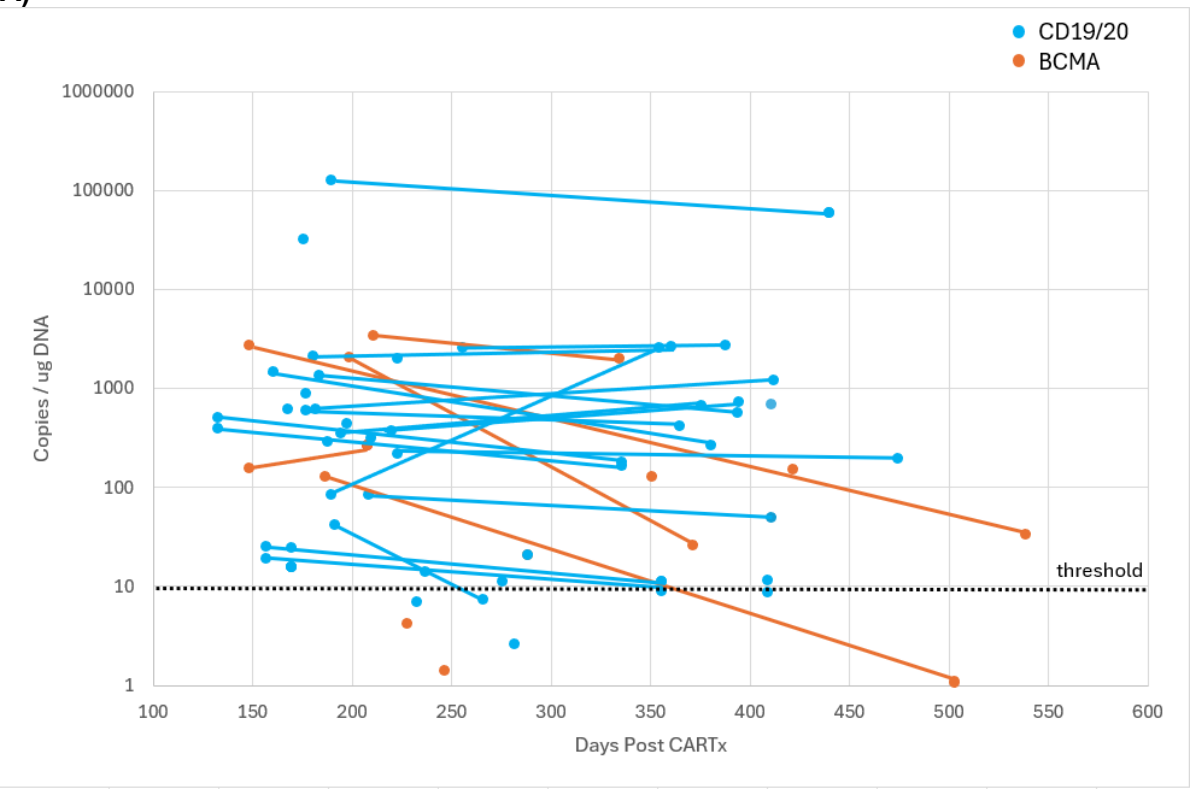
SUPPLEMENTAL FIGURES

A)



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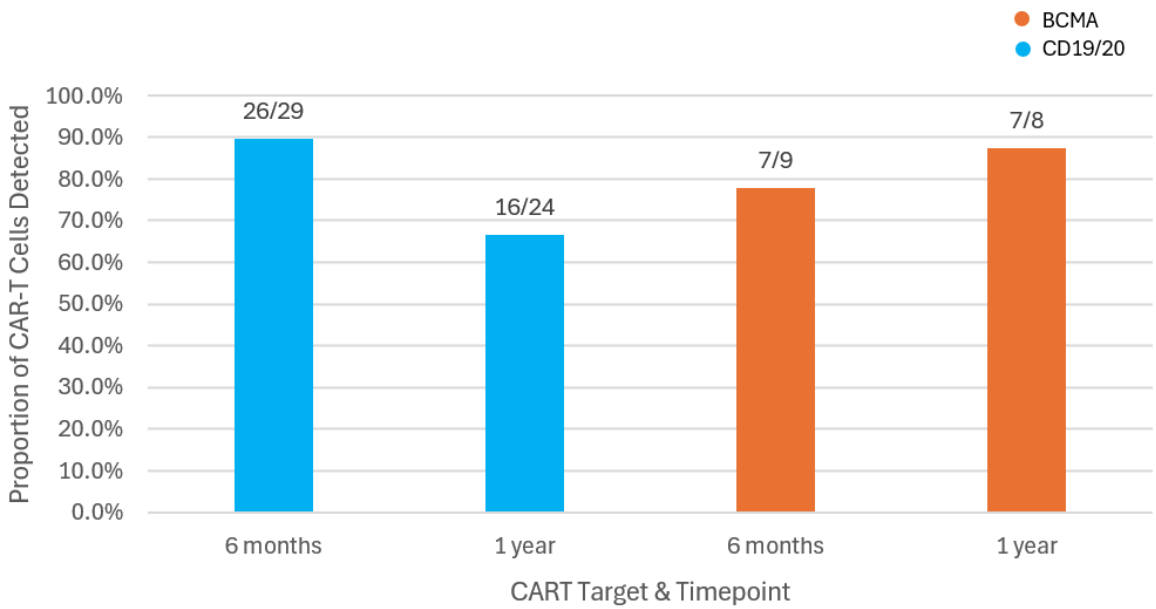


Figure S1. CAR-T cell persistence in CD19/20- and BCMA-CARTx recipients.
(A) Longitudinal quantification of CAR-T cell persistence measured by qPCR for CAR-T transgene (Flap-EF1, Hum CD19, or 28BB) copies per microgram of DNA in peripheral blood. Each line represents an individual recipient with paired measurements over time. Blue points and lines correspond to CD19/20-CARTx recipients; orange points and lines represent BCMA-CARTx recipients. The dotted horizontal line indicates the threshold above which a result is considered positive (10 copies / µg DNA). **(B)** Proportion of participants with detectable CAR-T cells at 6-months and 1-year post-infusion, stratified by CAR-T target. CD19/20-CARTx recipients are shown in blue and BCMA-CARTx recipients in orange. The number of participants with detectable CAR-T cells over the total number assessed is displayed above each bar.

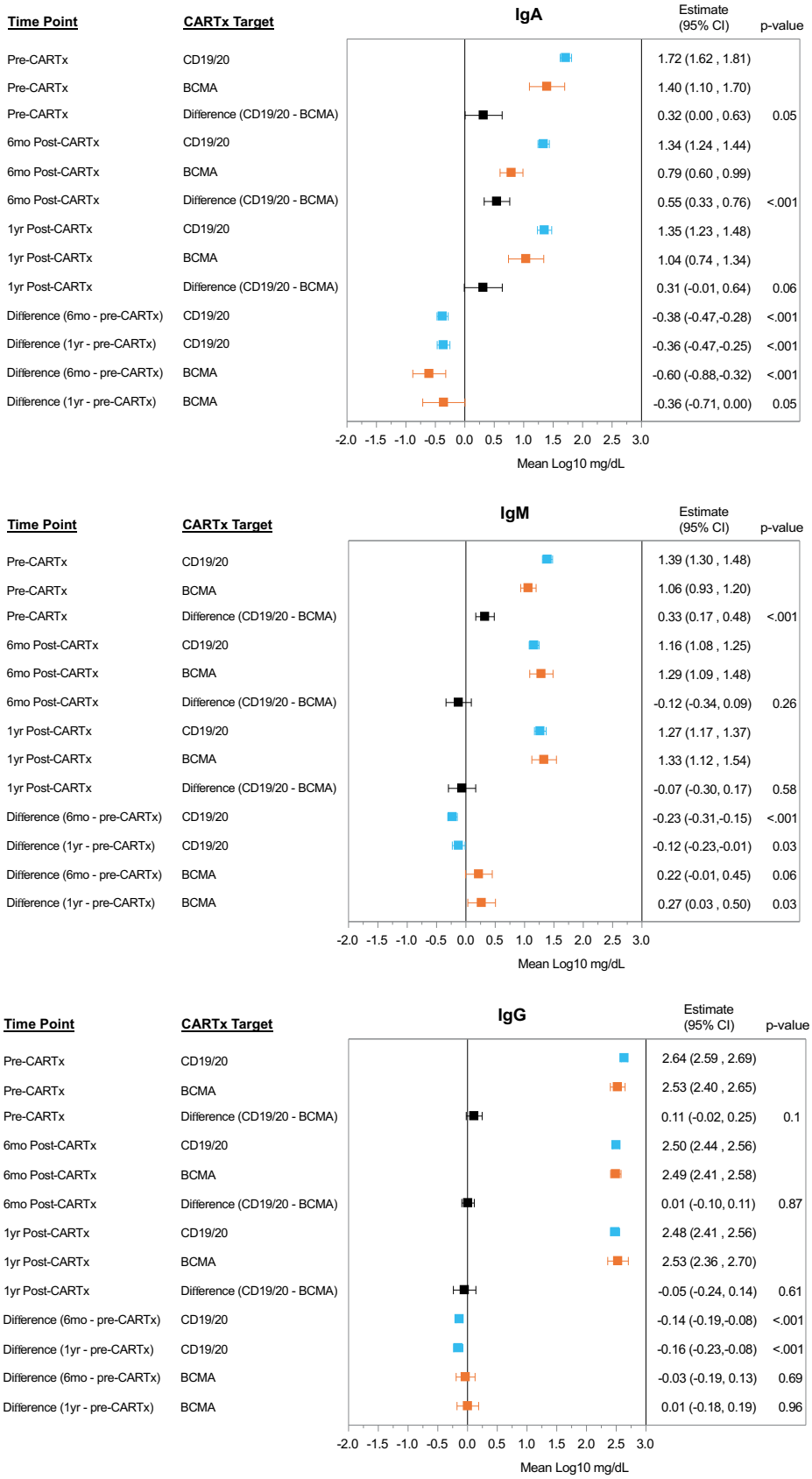
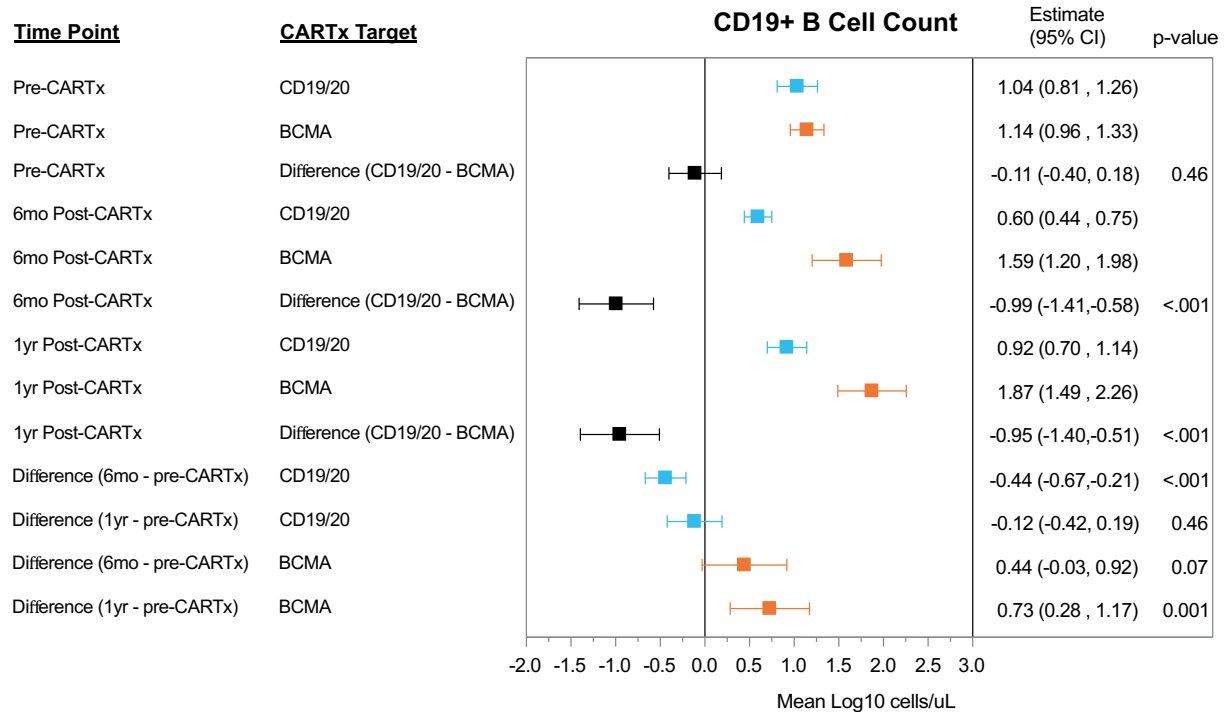


Figure S2. Unadjusted GEE model estimates of immunoglobulin subtype titers before and after BCMA- and CD19/20-CARTx.
Comparisons of immunoglobulin subtype titer by CARTx target and by time point are shown. Differences that do not cross the vertical reference line of x=0 represent statistically significant differences.



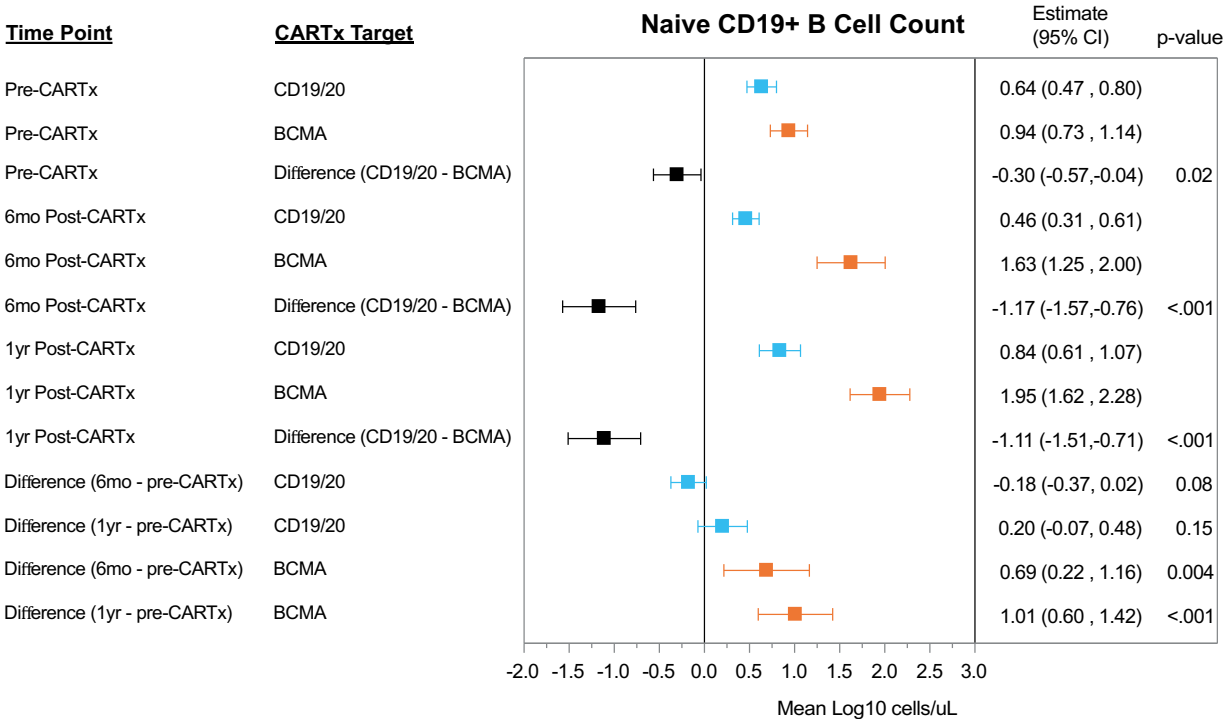
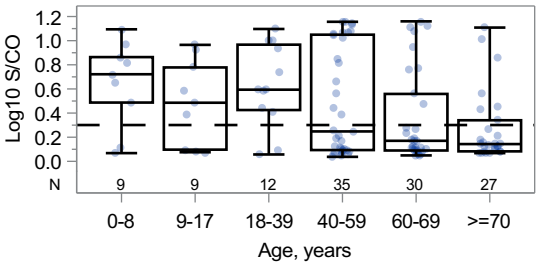
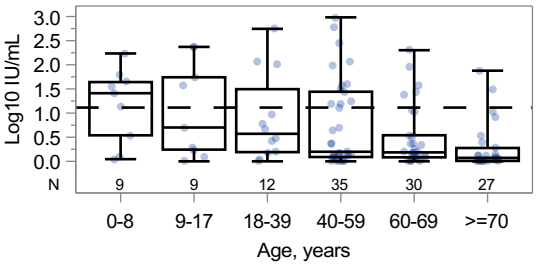


Figure S3. Unadjusted GEE model estimates of B cell counts before and after BCMA- and CD19/20-CARTx. Comparisons of B cell counts by CARTx target and by time point are shown. Differences that do not cross the vertical reference line of x=0 represent statistically significant differences.

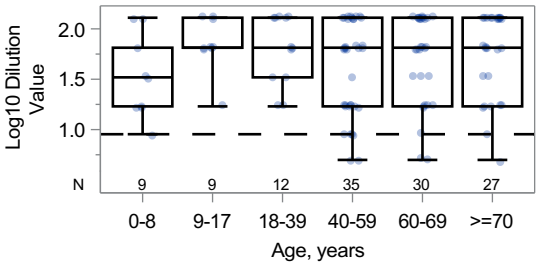
Hepatitis A Virus



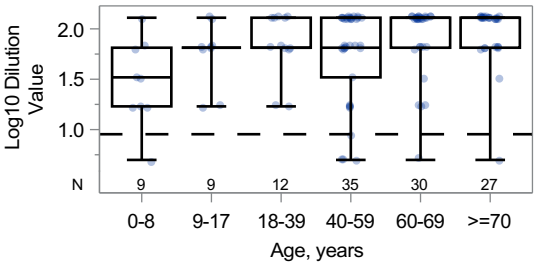
Hepatitis B Virus



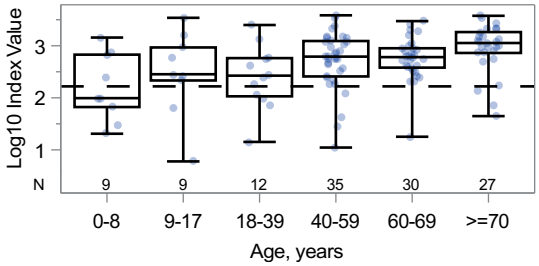
Poliovirus Type 1



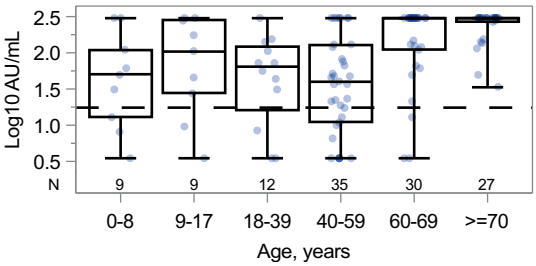
Poliovirus Type 3



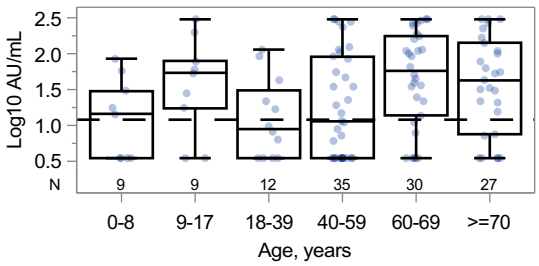
Varicella Zoster Virus



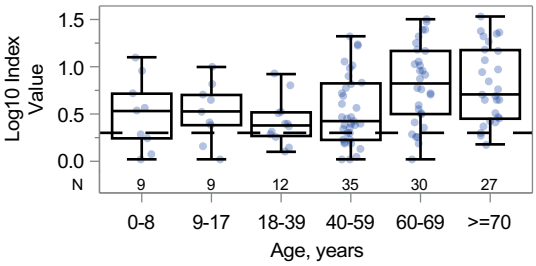
Measles



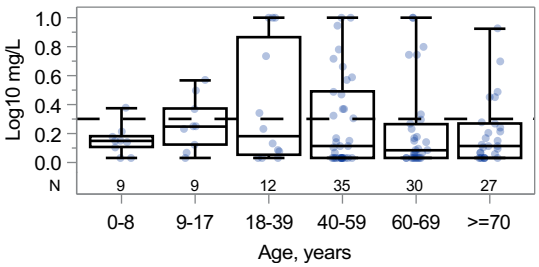
Mumps



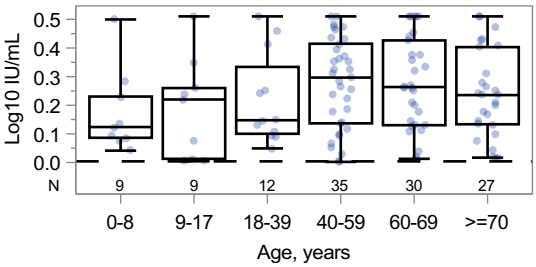
Rubella



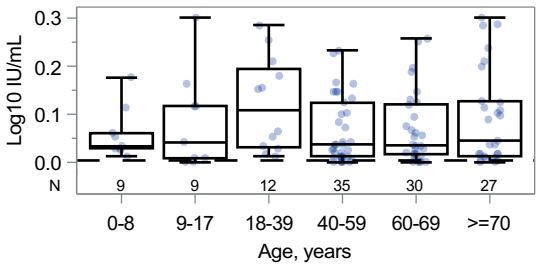
Haemophilus influenzae type B



C. tetani



C. diphtheriae



S. pneumoniae

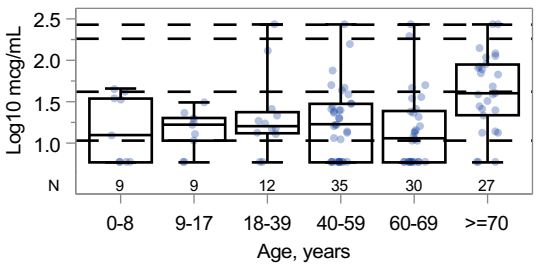


Figure S4. Pre-CARTx pathogen-specific antibody titers stratified by age among CD19, CD20, CD22, or CD19/22 CARTx recipients.

Distribution of pathogen-specific antibody titers, expressed as $\log_{10}(\text{value} + 1)$, at baseline (pre-CARTx) for CD19, CD20, CD22, or CD19/22 participants by age group. Number of participants is shown above the x-axis. The dashed reference line represents the threshold for seroprotection; for *S. pneumoniae*, the reference lines correspond to boundaries between low, modest, intermediate, moderate, and high levels.

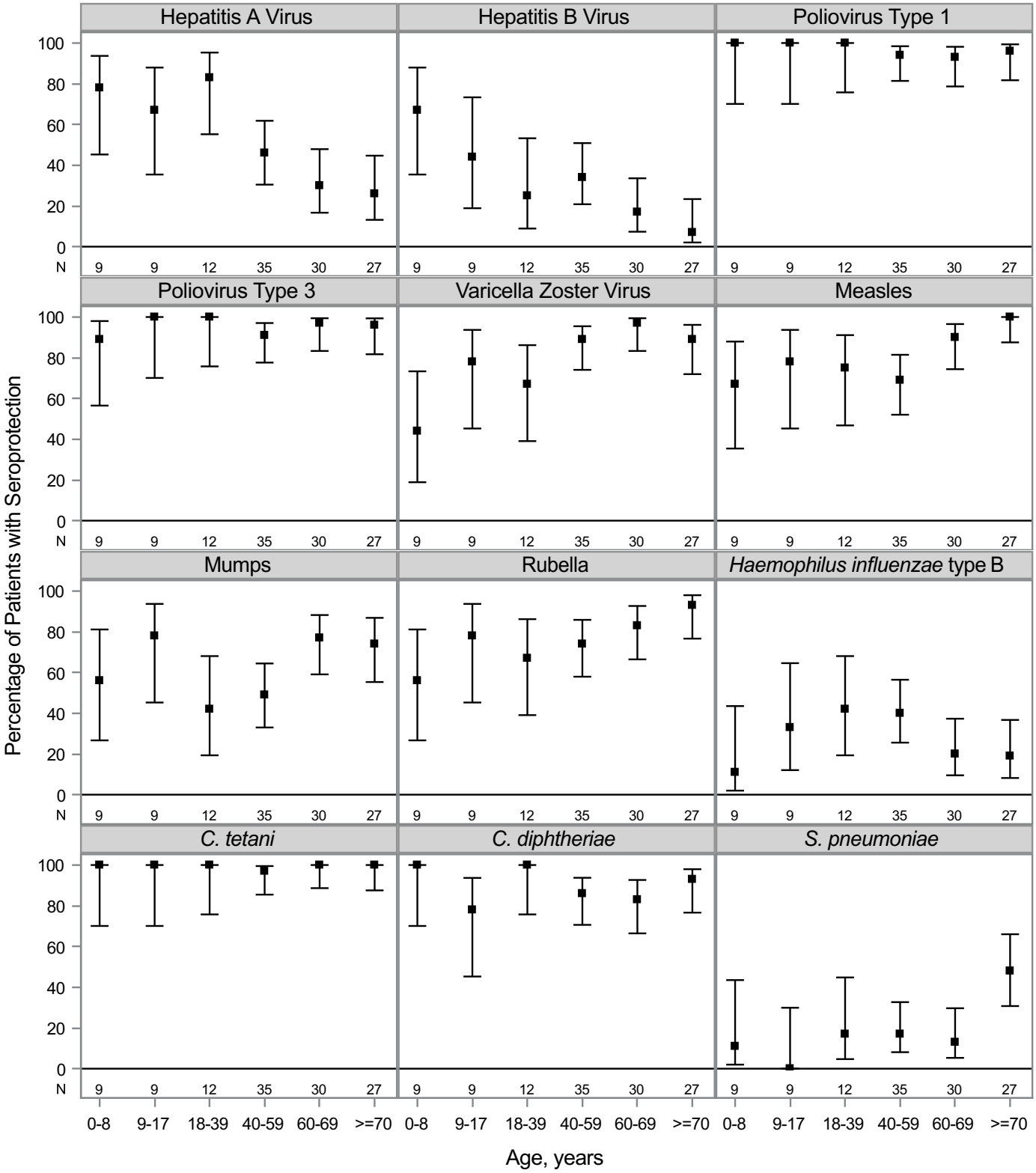


Figure S5. Percentage of CD19, CD20, CD22, or CD19/22 CARTx recipients with pre-CARTx seroprotective antibody titers stratified by age.
Number of participants is shown above the x-axis and brackets represent Wilson 95% confidence intervals.

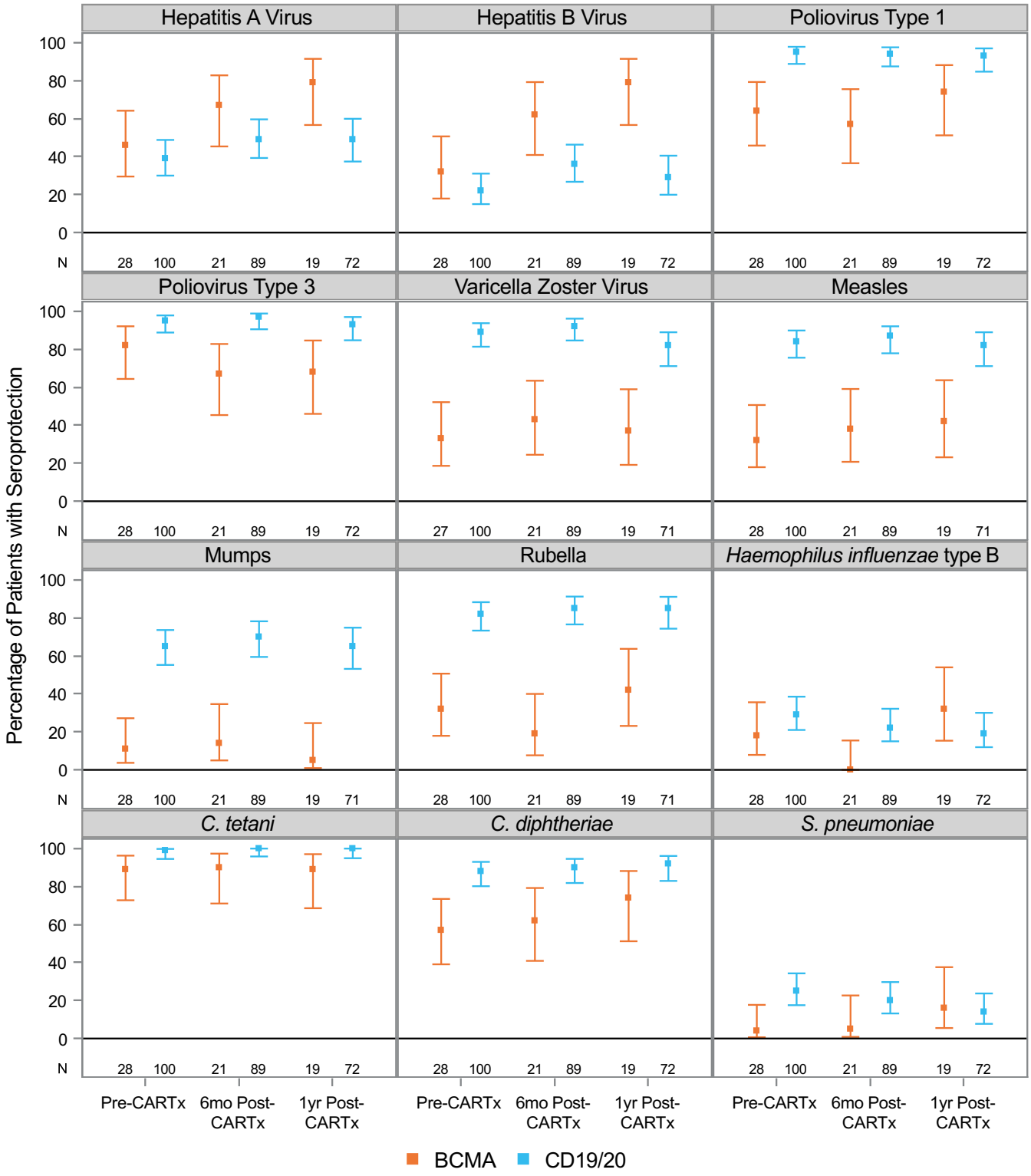


Figure S6. Percentage of participants with seroprotective pathogen-specific antibody titers before and after BCMA- and CD19/20-CARTx.

Percentage of participants with seroprotection by time points relative to CARTx and by participant groups defined by CARTx target received (BCMA-CARTx recipient data are shown in orange, CD19/20-CARTx recipient data are shown in blue). Number of participants is shown just above the x-axis and brackets represent Wilson 95% confidence intervals. For *S. pneumoniae*, seroprotection was defined as titers above the threshold for the second lowest category (>modest). Statistical comparisons from generalized estimating equation models are in **Table S5**).

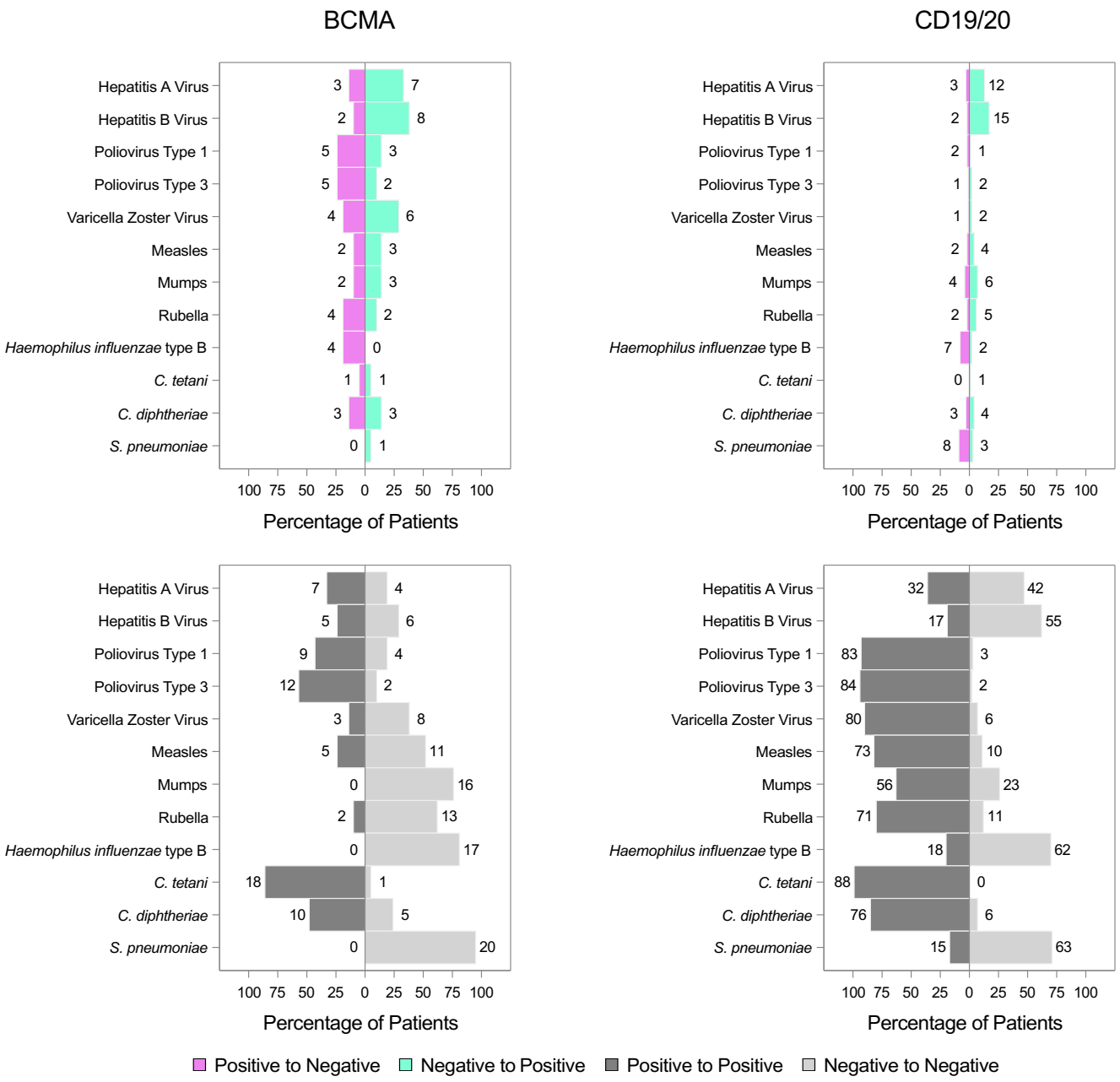


Figure S7. Change in pathogen-specific seroprotection status from pre- to six months post-BCMA- and CD19/20-CARTx.

Panels on the left show data for participants receiving BCMA-CARTx and panels on the right show data for participants receiving CD19/20-CARTx. The top row panels show the percentage of participants that changed seroprotection status, either from seroprotection pre-CARTx to no seroprotection 6 months post-CART (violet bars) or from no seroprotection pre-CARTx to seroprotection 6 months post-CART (aquamarine bars). The bottom row panels show the percentage of participants whose seroprotection status remained the same, either seroprotection at both time points (dark gray bars) or from no seroprotection at both time points (light gray bars). Percentages are computed among 21 BCMA- and 89 CD19/20-CARTx recipients contributing data, respectively. Numbers adjacent to bars represent the number of participants in each change category.

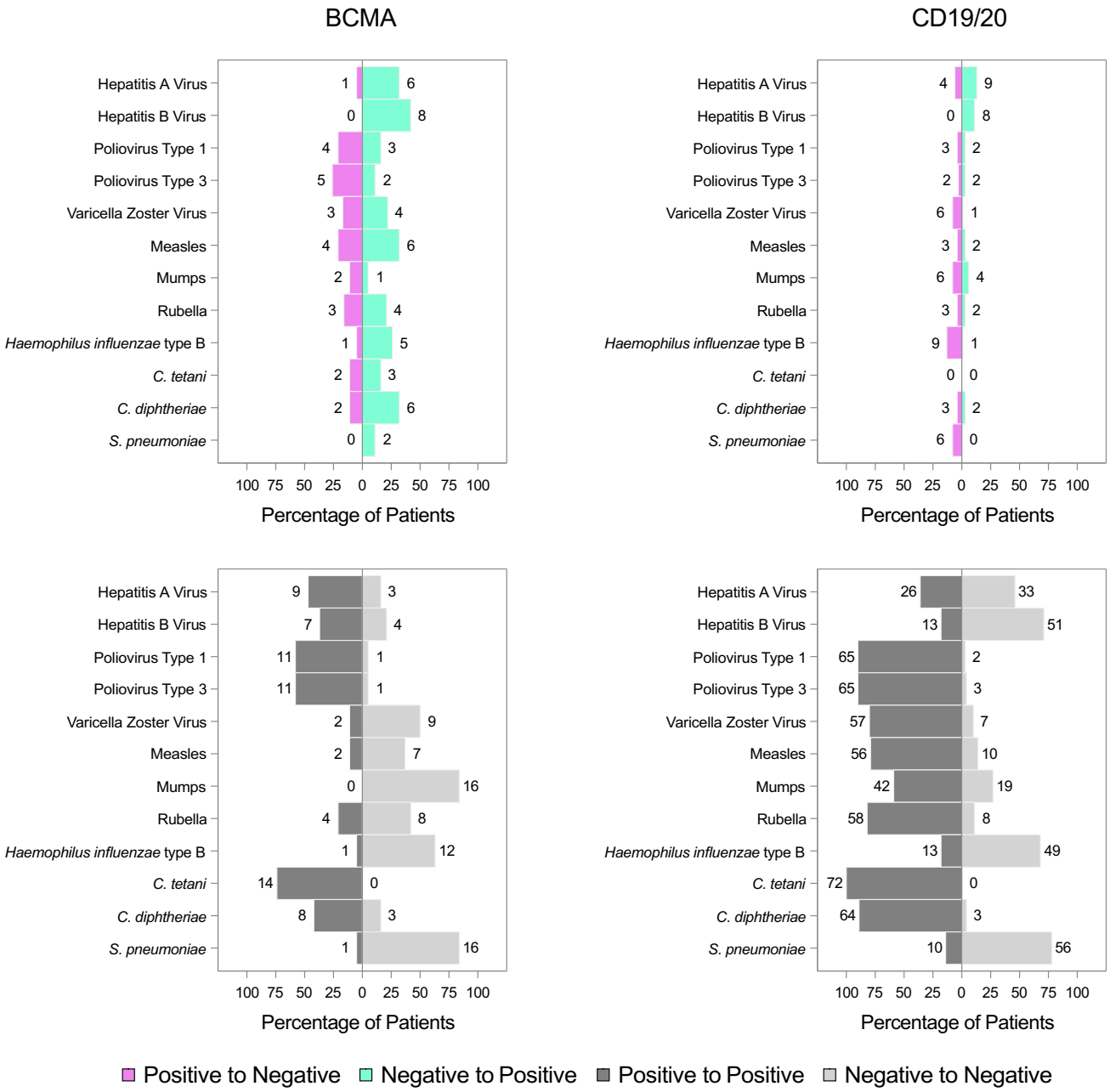


Figure S8. Change in pathogen-specific seroprotection status from pre- to one year post-BCMA- and CD19/20-CARTx.

Panels on the left show data for participants receiving BCMA-CARTx and panels on the right show data for participants receiving CD19/20-CARTx. The top row panels show the percentage of participants that changed seroprotection status, either from seroprotection pre-CARTx to no seroprotection 6 months post-CART (violet bars) or from no seroprotection pre-CARTx to seroprotection 6-months post-CART (aquamarine bars). The bottom row panels show the percentage of participants whose seroprotection status remained the same, either seroprotection at both time points (dark gray bars) or from no seroprotection at both time points (light gray bars). Percentages are computed among 19 BCMA- and 72 CD19/20-CARTx recipients. Numbers adjacent to bars represent the number of participants in each change category.

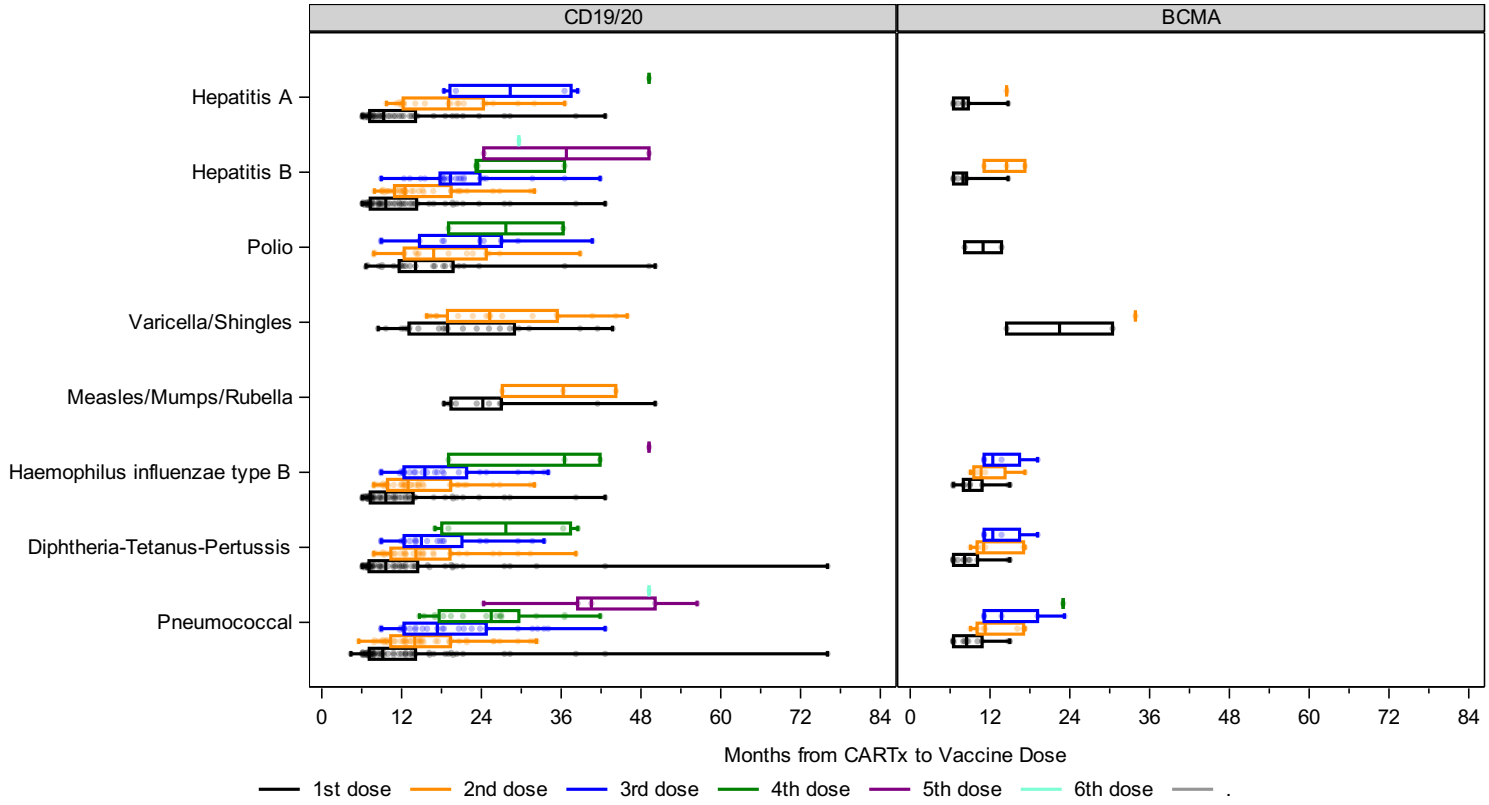


Figure S9. Timing of vaccine doses relative to BCMA- and CD19/20-CARTx.
Boxes represent interquartile range, horizontal lines within boxes represent the median, and whiskers extend to minimum and maximum values.

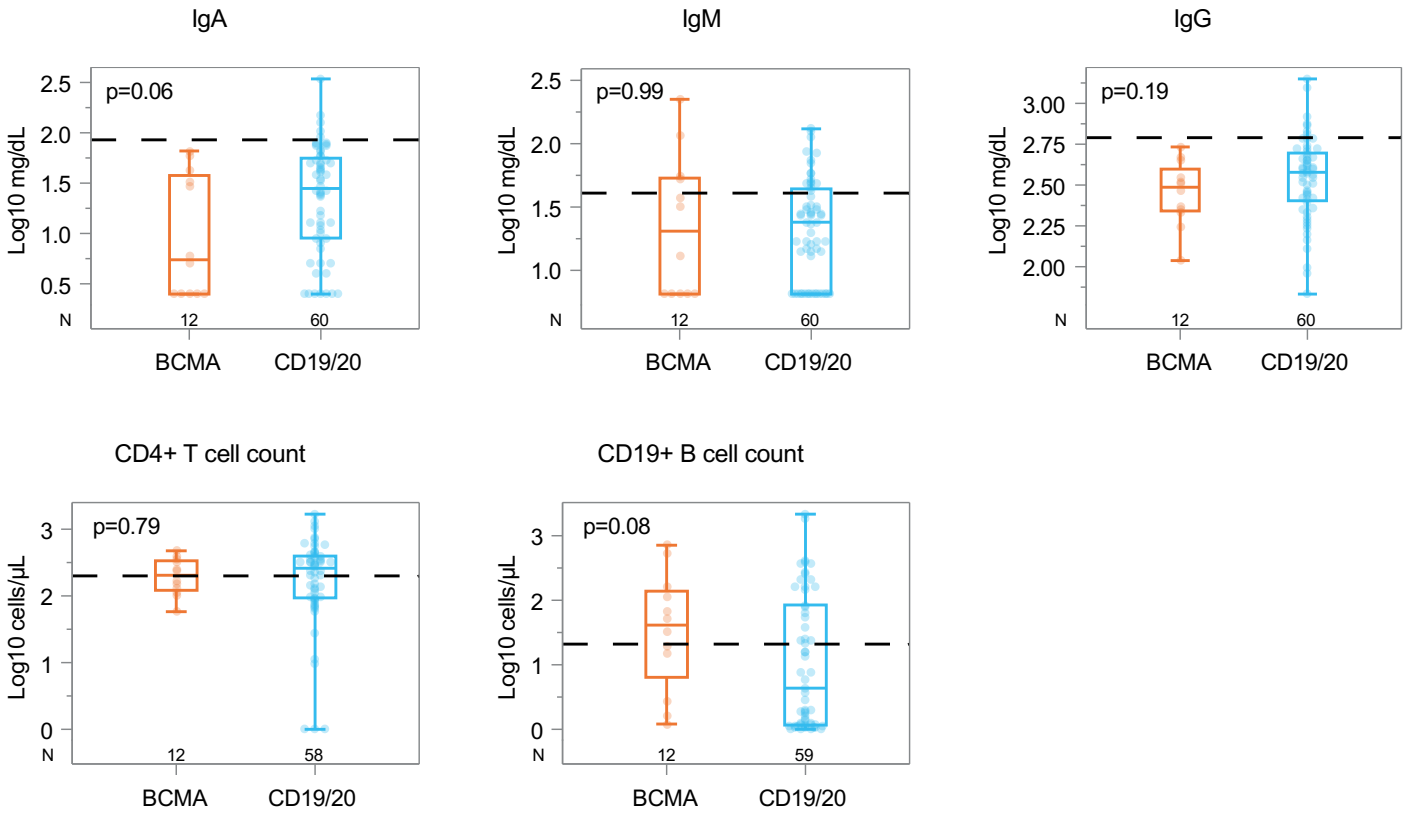


Figure S10. Immunoglobulin titers and T and B cell subsets prior to vaccination among BCMA- and CD19/20-CARTx recipients.

Distribution of IgA, IgM, IgG, and absolute CD4+ T cell and $CD19^+$ B cell counts, expressed as $\log_{10}(\text{value} + 1)$, measured within 6 months before or on date of first dose of earliest vaccine received, by participant groups defined by CARTx target (BCMA-CARTx recipient data are shown in orange, CD19/20-CARTx recipient data are shown in blue). Dashed horizontal reference lines represent the normal range for IgA, IgM, and IgG, and represent 200 µL for CD4+ T cell counts and 20 µL for $CD19^+$ B cell counts. Boxes represent interquartile range, horizontal lines within boxes represent the median, and whiskers extend to minimum and maximum values. Numbers above the x-axis show the number of participants in each group. P-values are from Wilcoxon rank-sum tests comparing values in BCMA- to CD19/20-CARTx recipients.

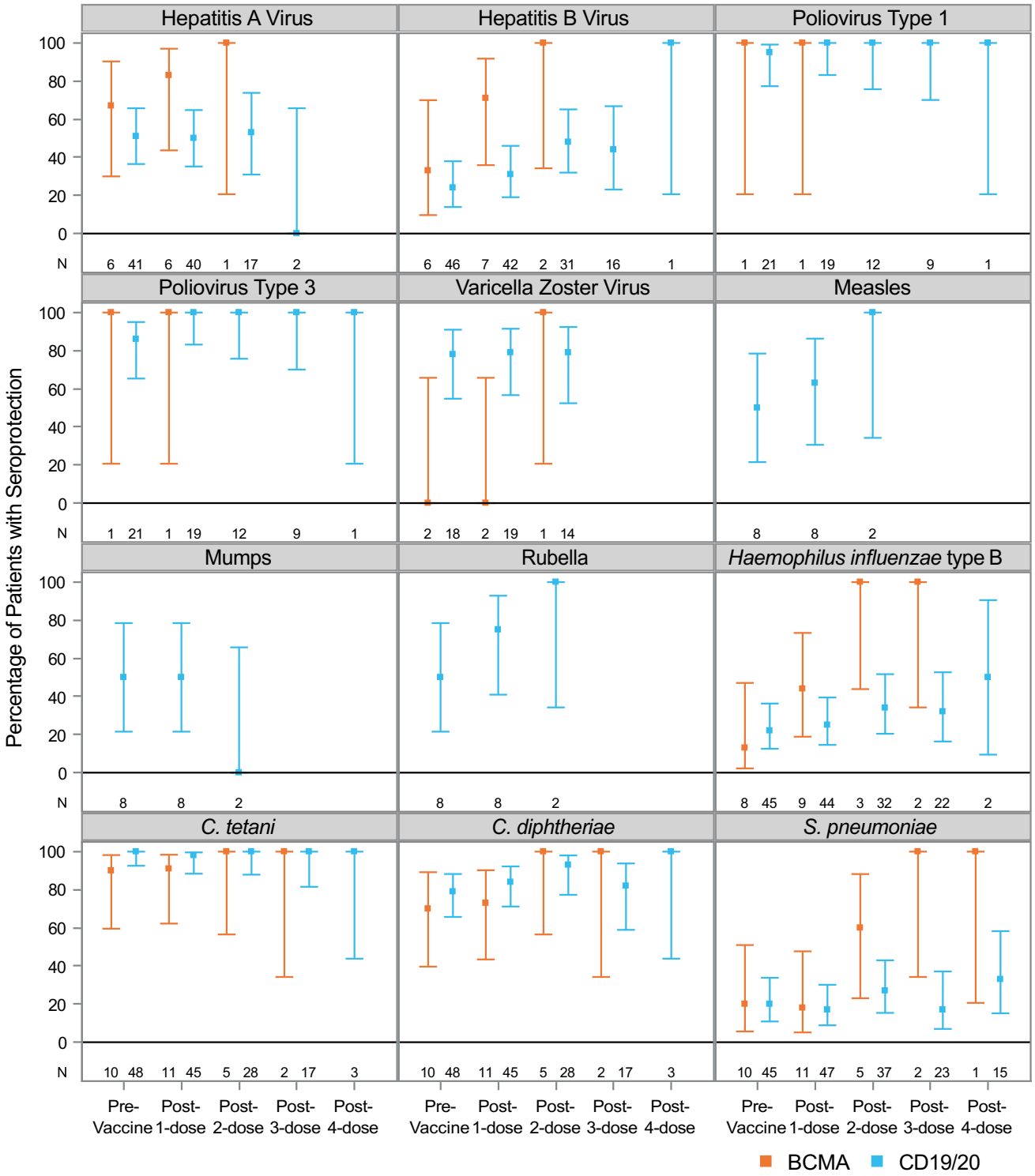


Figure S11: Percentage of BCMA- and CD19/20-CARTx recipients with seroprotective antibody levels pre/post vaccination.

Each panel displays the percentage of participants with seroprotective antibody titers to individual vaccine antigens over time, stratified by CAR-T product received: BCMA-targeted (orange) or CD19/20-targeted (blue). Seroprotection was determined using standardized, assay-specific thresholds. Data points represent the estimated proportion of participants with seroprotective titers at each timepoint, and vertical lines indicate 95% confidence intervals. X-axis labels denote vaccine timepoints. The number of participants included at each timepoint is shown directly above the x-axis. Data for three participants who received a fifth dose of pneumococcal vaccine are not shown (1 of 3 seroprotected), and data for one participant post-dose 5 and 6 of hepatitis B vaccine are excluded (seroprotected at both timepoints).

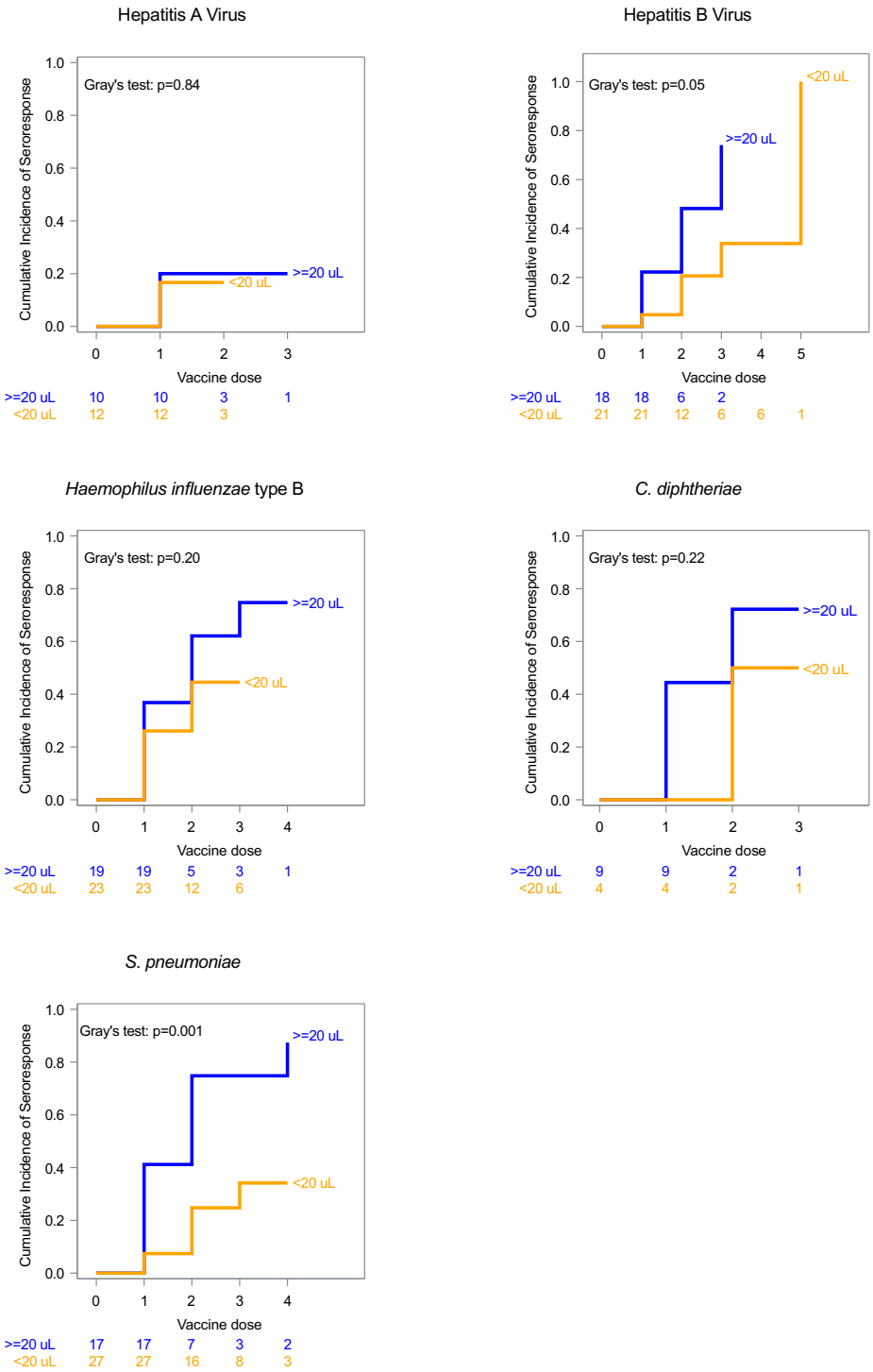


Figure S12: Cumulative incidence of seroresponse to vaccination after BCMA- and CD19/20-CARTx stratified by CD19⁺ B cell count.

Cumulative incidence curves showing the proportion of participants achieving a seroresponse, defined as a ≥2-fold increase in vaccine-specific antibody titer from the pre-vaccine baseline, over the course of sequential vaccine doses. Participants are stratified by baseline CD19⁺ B cell count ≥20 cells/μL (blue) versus <20 cells/μL (orange) using the most recent value prior to the first vaccine dose. The x-axis indicates vaccine dose number, and the y-axis represents the cumulative proportion of participants achieving a seroresponse. Numbers displayed below the x-axis reflect the number of participants at risk at each dose (i.e., those without prior seroresponse who continued to receive additional doses) for each CD19⁺ cell count group. Comparisons between CD19⁺ count strata were performed using Gray's test for equality of cumulative incidence functions; corresponding p-values are shown in each panel.

SUPPLEMENTAL TABLES:

Table S1: Participant Demographics and Clinical Characteristics of Pre/Post-CARTx Cohort

Baseline Characteristic[1]	**CD19/20**[2]	**BCMA**	**All**
Number of participants, n	100	28	128
Age at CARTx in years, median (range)	62 (18 - 83)	65 (42 - 74)	62 (18 - 83)
18 years or younger at CARTx	1 (1%)	0 (0%)	1 (1%)
Female	32 (32%)	14 (50%)	46 (36%)
Race			
White	88 (88%)	22 (79%)	110 (86%)
Asian	8 (8%)	1 (4%)	9 (7%)
American Indian / Alaska Native	1 (1%)	2 (7%)	3 (2%)
Black / African American	1 (1%)	1 (4%)	2 (2%)
Native Hawaiian / Pacific Islander	1 (1%)	0 (0%)	1 (1%)
Multiple	1 (1%)	0 (0%)	1 (1%)
Unknown	0 (0%)	2 (7%)	2 (2%)
Ethnicity			
Not Hispanic or Latino	92 (92%)	23 (82%)	115 (90%)
Hispanic or Latino	7 (7%)	2 (7%)	9 (7%)
Unknown	1 (1%)	3 (11%)	4 (3%)
Diagnosis			
Lymphoma	88 (88%)	0 (0%)	88 (69%)
ALL	5 (5%)	0 (0%)	5 (4%)
CLL	7 (7%)	0 (0%)	7 (5%)
IgG Multiple Myeloma	0 (0%)	21 (75%)	21 (16%)
Other Multiple Myeloma	0 (0%)	7 (25%)	7 (5%)
Number of prior cancer treatments, median (range)	4 (1 - 9)	8 (1 - 14)	4 (1 - 14)
Prior HCT			
No prior HCT	77 (77%)	7 (25%)	84 (66%)
Prior auto HCT only	18 (18%)	19 (68%)	37 (29%)
Prior allo HCT only	5 (5%)	1 (4%)	6 (5%)
Prior auto and allo HCT	0 (0%)	1 (4%)	1 (1%)
Months from most recent HCT to CARTx, median (range)	26 (4 - 148)	55 (20 - 292)	43 (4 - 292)
Prior CARTx	5 (5%)	1 (4%)	6 (5%)
Type of CAR-T cell product			
Commercial	59 (59%)	15 (54%)	74 (58%)

Investigational	41 (41%)	13 (46%)	54 (42%)
Conditioning chemotherapy			
Cyclophosphamide & fludarabine	99 (99%)	28 (100%)	127 (99%)
Bendamustine	1 (1%)	0 (0%)	1 (1%)
B cell targeting therapy in 6 months before CARTx			
None	37 (37%)	19 (68%)	56 (44%)
Rituximab only	40 (40%)	0 (0%)	40 (31%)
Rituximab and other	13 (13%)	0 (0%)	13 (10%)
Blinatumomab only	0 (0%)	0 (0%)	0 (0%)
Blinatumomab and other	1 (1%)	0 (0%)	1 (1%)
Other	9 (9%)	9 (32%)	18 (14%)

[1]Numbers shown are n(%) unless otherwise specified. ALL indicates acute lymphoblastic leukemia; CLL, chronic lymphocytic leukemia; HCT, hematopoietic cell transplant.

[2]88 were CD19, 11 were CD20, and 1 was CD19/CD20.

Table S2. Participant Demographics and Clinical Characteristics of Child, Adolescents or Young Adult (CAYA) Cohort

Baseline Characteristic[1]	All
Number of participants, n	22
CARTx target[2]	
CD19	13 (62%)
CD22	2 (10%)
CD19,CD22	6 (29%)
Age at CARTx in years, median (range)	9 (1 - 23)
Female	12 (55%)
Race	
White	13 (59%)
Asian	1 (5%)
American Indian / Alaska Native	1 (5%)
Black / African American	1 (5%)
Native Hawaiian / Pacific Islander	0 (0%)
Multiple	1 (5%)
Unknown	5 (23%)
Ethnicity	
Not Hispanic or Latino	12 (55%)
Hispanic or Latino	6 (27%)
Unknown	4 (18%)
Diagnosis	
Lymphoma	0 (0%)
ALL	22 (100%)
CLL	0 (0%)
Multiple Myeloma	0 (0%)
Other	0 (0%)
Number of prior cancer treatments, median (range)	5 (1 - 13)
Prior HCT	
No prior HCT	13 (76%)
Prior auto HCT only	0 (0%)
Prior allo HCT only	4 (24%)
Prior auto and allo HCT	0 (0%)
Months from most recent HCT to CARTx, median (range)	17 (12 - 34)
Prior CARTx	1 (6%)
Type of CAR-T cell product	

Commercial	1 (5%)
Investigational	21 (95%)
Conditioning chemotherapy	
Cyclophosphamide and fludarabine	17 (89%)
Other	2 (11%)
B cell targeting therapy in 6 months before CARTx	
None	12 (67%)
Rituximab only	0 (0%)
Rituximab and other	0 (0%)
Blinatumomab only	4 (22%)
Blinatumomab and other	0 (0%)
Other	2 (11%)

[1]Numbers shown are n (%) unless otherwise specified. Prior chemotherapy regimens was missing for n=8, history of HCT/CART was missing for n=5, and prior B cell targeting therapy was missing for n=4. Percentages are computed among those with non-missing data for these variables. CAYA indicates child, adolescent or young adult that were treated at Seattle Children's Hospital; ALL, acute lymphoblastic leukemia; CLL, chronic lymphocytic leukemia; HCT, hematopoietic cell transplant.

[2]One B-ALL participant did not receive CARTx and is omitted from CARTx target summaries.

Table S3. Summary Statistics for $CD19^+$ B Cell Counts

Time Point	$CD19^+$ B cell count cells/μL	CD19/20[1]	BCMA[1]	All[1]
Pre-CARTx				
	0	1 (1%)	0 (0%)	1 (1%)
	≥ 0 to 20	62 (63%)	17 (63%)	79 (63%)
	≥ 20	36 (36%)	10 (37%)	46 (37%)
6mo post-CARTx				
	0	2 (2%)	1 (5%)	3 (3%)
	≥ 0 to 20	72 (81%)	5 (25%)	77 (71%)
	≥ 20	15 (17%)	14 (70%)	29 (27%)
1yr post-CARTx				
	0	1 (1%)	1 (6%)	2 (2%)
	≥ 0 to 20	43 (60%)	2 (12%)	45 (51%)
	≥ 20	28 (39%)	14 (82%)	42 (47%)

[1] Numbers shown are the number of participants in each category (%).

Table S4. P-values for Pathogen-Specific Titer Comparisons between CARTx Subgroup and Over Time

Comparison[1]	Hepatitis A Virus	Hepatitis B Virus	Poliovirus Type 1	Poliovirus Type 3	Varicella Zoster Virus	Measles	Mumps	Rubella	Haemophilus influenzae type B	C.tetani	C. diphtheriae	S. pneumoniae
Pre-CARTx CD19/20 vs BCMA	0.77	0.54	<.001	<.001	<.001	<.001	<.001	<.001	0.01	<.001	<.001	<.001
6mo Post-CARTx CD19/20 vs BCMA	0.66	0.026	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001	0.07
1yr Post-CARTx CD19/20 vs BCMA	0.03	<.001	<.001	<.001	<.001	<.001	<.001	<.001	0.04	0.005	0.63	0.17
CD19/20 6mo post-CARTx vs pre-CARTx	0.009	<.001	0.15	0.75	0.8	0.23	0.37	0.99	0.03	0.22	0.03	0.002
CD19/20 1yr post-CARTx vs pre-CARTx	0.1	0.09	0.3	0.19	0.13	0.2	0.83	0.91	0.07	0.62	0.66	<.001
BCMA 6mo post-CARTx vs pre-CARTx	0.3	0.005	0.51	0.09	0.87	0.89	0.69	0.07	0.11	0.51	0.98	0.44
BCMA 1yr post-CARTx vs pre-CARTx	0.011	<.001	0.47	0.34	0.76	0.98	0.31	0.76	0.005	0.073	0.05	<.001

[1]From GEE models with independent variables for CARTx target, time period, and the interaction between CARTx target and time period.

Color coding key: Green indicates the first group listed in the comparison has a greater estimated mean $\log_{10}$ titer than the second group listed in the comparison, and yellow indicates the first group listed in the comparison has a lower estimated mean $\log_{10}$ titer than the second group listed in the comparison. Dark shading indicates $p<0.001$ and light shading indicates $0.001 \leq p < 0.05$.

Table S5. P-values for Pathogen-Specific Seroprotection Comparisons between CARTx Subgroup and Over Time

Comparison[1]	Hepatitis A Virus	Hepatitis B Virus	Poliovirus Type 1	Poliovirus Type 3	Varicella Zoster Virus	Measles	Mumps	Rubella	Haemophilus influenzae type B[2]	C. tetani[2]	C. diphtheriae	S. pneumoniae
Pre-CARTx CD19/20 vs BCMA	0.48	0.27	<.001	0.04	<.001	<.001	<.001	<.001	0.24	0.03	<.001	0.04
6mo Post-CARTx CD19/20 vs BCMA	0.16	0.03	<.001	<.001	<.001	<.001	<.001	<.001	0.012	0.04	0.003	0.13
1yr Post-CARTx CD19/20 vs BCMA	0.02	<.001	0.03	0.007	<.001	0.001	<.001	<.001	0.35	0.04	0.04	0.83
CD19/20 6mo post-CARTx vs pre-CARTx	0.02	0.002	0.77	0.44	0.21	0.39	0.23	0.274	0.07	--	0.55	0.2
CD19/20 1yr post-CARTx vs pre-CARTx	0.06	0.1	0.50	0.42	0.05	0.5	0.97	0.528	0.04	--	0.35	0.007
BCMA 6mo post-CARTx vs pre-CARTx	0.15	0.03	0.58	0.18	0.5	0.57	0.72	0.260	--	0.87	0.68	0.84
BCMA 1yr post-CARTx vs pre-CARTx	0.02	<.001	0.5	0.28	0.8	0.51	0.53	0.438	0.26	0.99	0.22	0.06

[1]From GEE models with independent variables for CARTx target, time period, and the interaction between CARTx target and time period for all pathogens except for Haemophilus influenzae type B and C. tetani.

[2]Due to zero cells and sparse data, alternate testing methods were used. For CARTx target comparisons, chi square or Fisher's exact tests were used. For time point comparisons, some p-values come from GEE models on subset datasets and other testing was not possible.

Color coding key: Green indicates the first group listed in the comparison has a greater estimated proportion of participants with seroprotection than the second group listed in the comparison, and yellow indicates the first group listed in the comparison has a lower estimated proportion of participants with seroprotection than the second group listed in the comparison. Dark shading indicates $p<0.001$ and light shading indicates $0.001 \leq p < 0.05$.

Table S6. GEE Model Estimates for Association of Adjustment Variables with Proportion of Pathogens with Seroprotection

Variable	Adjusted OR (95% CI)[1]	p-value
Age		
60 years or younger	1.0 (Reference)	
older than 60 years	1.09 (0.89, 1.34)	0.387
Sex		
Male	1.0 (Reference)	
Female	0.99 (0.79, 1.24)	0.946
Race		
White	1.0 (Reference)	
Non-White	0.78 (0.58, 1.04)	0.088
Ethnicity		
Non-Hispanic or Latino	1.0 (Reference)	
Hispanic or Latino	1.65 (0.97, 2.78)	0.064
Number of prior cancer treatments		
4 or fewer	1.0 (Reference)	
greater than 4	0.83 (0.67, 1.02)	0.080
History of HCT or CARTx		
No	1.0 (Reference)	
Yes	0.91 (0.73, 1.15)	0.443
B cell targeting therapy in 6 months before CARTx		
No	1.0 (Reference)	
Yes	1.02 (0.83, 1.26)	0.834
Type of CAR T cell product		
Investigational	1.0 (Reference)	
Commercial	0.88 (0.72, 1.07)	0.202
Time-dependent total IgG		
Less than 400	1.0 (Reference)	
400 or higher	1.35 (1.14, 1.61)	<.001
Time-dependent total IgA		
per 1 $\log_{10}$ unit increase	1.29 (1.08, 1.53)	0.004
Time-dependent total IgM		
Less than 20	1.0 (Reference)	
20 or higher	1.11 (0.94, 1.32)	0.217
Time-dependent B cell count		
per 1 $\log_{10}$ unit increase	0.97 (0.89, 1.05)	0.467
Time-dependent CD4 T cell count		

per 1 log_{10} unit increase	0.95 (0.83, 1.09)	0.471
Time-dependent receipt of IGRT in previous 8-16 weeks		
No	1.0 (Reference)	
Yes	2.24 (1.63, 3.09)	<.001
Time-dependent number of pathogens with vaccination prior to sample		
per 1 unit increase in pathogens	1.01 (0.96, 1.05)	0.762

[1]OR indicates odds ratio; CI, confidence interval. Estimates adjusted for CARTx target, time point, and interaction between CARTx target and time point, time-dependent receipt of IGRT in 8-16 weeks prior to sample, and time-dependent number of pathogens with vaccination prior to sample. Variables meeting criteria for consideration in a multivariable model are highlighted in yellow.

Table S7. GEE Multivariable Model Estimates for Association of Primary Exposure and Adjustment Variables with Proportion of Pathogens with Seroprotection

Variable	Adjusted OR (95% CI)[1]	p-value
At Pre-CARTX, CARTx target		
BCMA	1.0 (Reference)	
CD19/20	2.71 (1.99, 3.69)	<.001
At 6mo Post-CARTX, CARTx target		
BCMA	1.0 (Reference)	
CD19/20	2.92 (1.88, 4.55)	<.001
At 1yr Post-CARTX, CARTx target		
BCMA	1.0 (Reference)	
CD19/20	1.89 (1.26, 2.83)	0.002
Among CD19/20 participants, time point		
Pre-CARTx	1.0 (Reference)	
6mo post-CARTx	1.20 (1.06, 1.35)	0.004
1yr post-CARTx	0.97 (0.76, 1.23)	0.800
Among BCMA participants, time point		
Pre-CARTx	1.0 (Reference)	
6mo post-CARTx	1.11 (0.70, 1.75)	0.654
1yr post-CARTx	1.39 (0.85, 2.26)	0.185
Race		
White	1.0 (Reference)	
Non-White	0.73 (0.55, 0.97)	0.032
Time-dependent total IgG		
Less than 400	1.0 (Reference)	
400 or higher	1.31 (1.11, 1.55)	0.002
Time-dependent total IgA		
per 1 $\log_{10}$ unit increase	1.20 (1.00, 1.45)	0.048
Time-dependent B cell count		
per 1 $\log_{10}$ unit increase	0.95 (0.87, 1.02)	0.161
Time-dependent receipt of IGRT in 8-16 weeks prior to sample		
No	1.0 (Reference)	
Yes	2.39 (1.72, 3.32)	<.001
Time-dependent number of pathogens with vaccination prior to sample		
per 1 unit increase in pathogens	1.00 (0.96, 1.05)	0.954

[1]OR indicates odds ratio; CI, confidence interval. Estimates adjusted for CARTx target, time point, and interaction between CARTx target and time point, race, time-dependent total IgG, time-dependent total IgA, time-dependent $CD19^+$

B cell count, time-dependent receipt of IGRT in previous 8-16 weeks, and time-dependent number of pathogens with vaccination prior to sample.

Table S8. GEE Multivariable Model Estimates for Association of Adjustment Variables with VirScan Total Epitope Scores

Variable	Estimate (95% CI)[1]	p-value
Age		
60 years or younger	(Reference)	
older than 60 years	1.84 (-21.56, 25.23)	0.88
Sex		
Male	(Reference)	
Female	8.52 (-16.68, 33.72)	0.51
Race		
White	(Reference)	
Non-White	15.18 (-23.83, 54.20)	0.45
Ethnicity		
Non-Hispanic or Latino	(Reference)	
Hispanic or Latino	40.42 (-15.36, 96.20)	0.16
Number of prior cancer treatments		
4 or fewer	(Reference)	
greater than 4	-18.48 (-44.33, 7.37)	0.16
History of HCT or CARTx		
No	(Reference)	
Yes	-29.70 (-54.14, -5.27)	0.017
B cell targeting therapy in 6 months before CARTx		
No	(Reference)	
Yes	18.91 (-6.50, 44.31)	0.15
Type of CAR-T cell product		
Investigational	(Reference)	
Commercial	13.37 (-10.13, 36.87)	0.27
Time-dependent total IgG		
Less than 400	(Reference)	
400 or higher	-4.00 (-25.46, 17.45)	0.72
Time-dependent total IgA		
per 1 $\log_{10}$ unit increase	14.77 (-9,38, 38.92)	0.23
Time-dependent total IgM		
Less than 20	(Reference)	
20 or higher	2.86 (-17.85, 23.58)	0.79
Time-dependent B cell count		
per 1 $\log_{10}$ unit increase	-4.44 (-16.52, 7.65)	0.47

Time-dependent CD4 T cell count		
per 1 log_{10} unit increase	0.58 (-18.65, 19.82)	0.95

OR indicates odds ratio; HCT, hematopoietic cell transplant. Estimates adjusted for CARTx target, time point, and interaction between CARTx target and time point. The variables chosen for inclusion in adjusted modelling are highlited in yellow.

Table S9. GEE Multivariable Model Estimates for Association of Primary Exposure and Adjustment Variables with VirScan Total Epitope Scores

Variable	Adjusted Estimate (95% CI)[1]	p-value
At Pre-CARTX, CARTx target		
BCMA	1.0 (Reference)	
CD19/20	46.98 (10.28, 83.67)	0.012
At 6mo Post-CARTX, CARTx target		
BCMA	1.0 (Reference)	
CD19/20	74.3 (30.96, 117.65)	0.0008
At 1yr Post-CARTX, CARTx target		
BCMA	1.0 (Reference)	
CD19/20	80.06 (33.82, 126.3)	0.0007
Among CD19/20 participants, time point		
Pre-CARTx	1.0 (Reference)	
6mo post-CARTx	15.47 (-7.89, 38.83)	0.19
1yr post-CARTx	20.23 (-7.67, 48.12)	0.16
Among BCMA , time point		
Pre-CARTx	1.0 (Reference)	
6mo post-CARTx	-11.857 (-56.99, 33.27)	0.61
1yr post-CARTx	-12.86 (-59.58, 33.86)	0.59
Ethnicity		
Non-Hispanic	1.0 (Reference)	
Hispanic	35.01 (-26, 96.02)	0.26
History of HCT or CARTx		
No	1.0 (Reference)	
Yes	-28.79 (-54, -3.58)	0.03
Time-dependent total IgG		
Less than 400	1.0 (Reference)	
400 or higher	-16.23 (-40.94, 8.49)	0.2
Time-dependent total IgA		
per 1 $\log_{10}$ unit increase	12.39 (-17.71, 41.86)	0.41

[1]OR indicates odds ratio; CI, confidence interval; HCT, hematopoietic cell transplant. Estimates adjusted for CARTx target, time point, and interaction between CARTx target and time point, race, time-dependent total IgG, time-dependent total IgA, time-dependent $CD19^+$ B cell count, time-dependent receipt of IVIG in previous 8-16 weeks, and time-dependent number of pathogens with vaccination prior to sample.

Table S10. Participant Demographics and Clinical Characteristics of Post-CARTx Vaccine Cohort

Baseline Characteristic[1]	CD19/20[2]	BCMA	All
Number of participants, n	60	12	72
Age at CARTx in years, median (range)	58 (15 - 79)	64 (44 - 74)	59 (15 - 79)
18 years or younger at CARTx	1 (2%)	0 (0%)	1 (1%)
Female	16 (27%)	6 (50%)	22 (31%)
Race			
White	52 (87%)	11 (92%)	63 (88%)
Asian	7 (12%)	1 (8%)	8 (11%)
American Indian / Alaska Native	0 (0%)	0 (0%)	0 (0%)
Black / African American	0 (0%)	0 (0%)	0 (0%)
Native Hawaiian / Pacific Islander	0 (0%)	0 (0%)	0 (0%)
Multiple	1 (2%)	0 (0%)	1 (1%)
Ethnicity			
Not Hispanic or Latino	57 (95%)	11 (92%)	68 (94%)
Hispanic or Latino	3 (5%)	0 (0%)	3 (4%)
Unknown	0 (0%)	1 (8%)	1 (1%)
Diagnosis			
Lymphoma	52 (87%)	0 (0%)	52 (72%)
ALL	5 (8%)	0 (0%)	5 (7%)
CLL	2 (3%)	0 (0%)	2 (3%)
IgG Multiple Myeloma	0 (0%)	8 (67%)	8 (11%)
Other Multiple Myeloma	0 (0%)	4 (33%)	4 (6%)
Number of prior cancer treatments, median (range)	3 (1 - 12)	7 (1 - 14)	4 (1 - 14)
Prior HCT			
No prior HCT	42 (70%)	3 (25%)	45 (63%)
Prior auto HCT only	14 (23%)	8 (67%)	22 (31%)
Prior allo HCT only	4 (7%)	1 (8%)	5 (7%)
Prior auto and allo HCT	0 (0%)	0 (0%)	0 (0%)
Months from most recent HCT to CARTx, median (range)	30 (4 - 148)	55 (20 - 292)	35 (4 - 292)
Prior CARTx	3 (5%)	0 (0%)	3 (4%)
Type of CAR-T cell product			
Commercial	31 (52%)	5 (42%)	36 (50%)
Investigational	29 (48%)	7 (58%)	36 (50%)
Conditioning chemotherapy			
Cyclophosphamide and fludarabine	60 (100%)	12 (100%)	72 (100%)

B cell targeting therapy in 6 months before CARTx			
None	23 (38%)	10 (83%)	33 (46%)
Rituximab only	26 (43%)	0 (0%)	26 (36%)
Rituximab and other	5 (8%)	0 (0%)	5 (7%)
Blinatumomab only	0 (0%)	0 (0%)	0 (0%)
Blinatumomab and other	1 (2%)	0 (0%)	1 (1%)
Other	5 (8%)	2 (17%)	7 (10%)

ALL indicates acute lymphoblastic leukemia; CLL, chronic lymphocytic leukemia; HCT, hematopoietic cell transplant.

[1]Numbers shown are n (%) unless otherwise specified.

[2]63 were CD19, 6 were CD20, and 1 was CD19/CD20.

Table S11. Timing of Subsequent Vaccine Doses after CARTx

Vaccine	Summary	1st Dose	2nd Dose	3rd Dose	4th Dose	5th Dose	6th Dose
Hepatitis A	Number of participants	54	22	4	1		
	Median months from CARTx to vaccine dose (IQR)	8.8 (7.1, 13.9)	17.3 (12.2, 24.3)	28.3 (19.3, 37.5)	49.2		
Hepatitis B	Number of participants	59	39	23	3	2	1
	Median months from CARTx to vaccine dose (IQR)	9.0 (7.1, 14.1)	12.6 (11.0, 19.4)	19.4 (17.8, 23.8)	23.4 (23.2, 36.5)	36.8 (24.3, 49.2)	29.6
Polio	Number of participants	27	14	11	2		
	Median months from CARTx to vaccine dose (IQR)	13.7 (10.8, 19.7)	16.8 (12.4, 24.7)	23.8 (14.7, 27.0)	27.7 (19.1, 36.3)		
VZV	Number of participants	25	17				
	Median months from CARTx to vaccine dose (IQR)	18.9 (13.2, 29.0)	25.4 (20.5, 35.3)				
MMR	Number of participants	10	3				
	Median months from CARTx to vaccine dose (IQR)	24.2 (19.4, 27.0)	36.3 (27.1, 44.2)				
Hib	Number of participants	61	40	30	3	1	
	Median months from CARTx to vaccine dose (IQR)	9.0 (7.4, 13.5)	12.6 (9.9, 18.3)	14.6 (12.3, 20.6)	36.5 (19.1, 41.9)	49.2	
DTaP	Number of participants	65	40	24	4		
	Median months from CARTx to vaccine dose (IQR)	8.9 (7.1, 14.1)	13.2 (10.2, 17.1)	14.1 (12.3, 18.8)	27.7 (18.0, 37.4)		
Pneumococcal	Number of participants	69	51	32	17	5	1
	Median months from CARTx to vaccine dose (IQR)	8.8 (7.1, 13.9)	13.5 (10.1, 19.1)	16.6 (12.4, 23.5)	24.7 (18.3, 27.0)	40.5 (38.5, 50.1)	49.2

IQR indicates Interquartile range; MMR, measles, mumps, rubella vaccine; Hib, *Haemophilus influenzae* type B vaccine; DTaP, Diptheria,tetanus, acellular pertussis vaccine

Table S12. Number of Seroresponse Events and Cumulative Incidence Estimates Following Vaccine Doses

	Vaccine Dose							
	1		2		3		4	
Participant Category	**Number of Events[1]**	**Cumulative Incidence, % (95% CI)**	**Number of Events[1]**	**Cumulative Incidence, % (95% CI)**	**Number of Events[1]**	**Cumulative Incidence, % (95% CI)**	**Number of Events[1]**	**Cumulative Incidence, % (95% CI)**
Pathogen								
Hepatitis A Virus	4	18.2 (5.5 - 36.7)	0	18.2 (5.5 - 36.7)	0	18.2 (5.5 - 36.7)		
Hepatitis B Virus	5	12.8 (4.6 - 25.4)	4	32.2 (14.8 - 51.1)	2	49.1 (22.4 - 71.4)		49.1 (22.4 - 71.4)
Haemophilus influenzae type B	13	31.0 (17.7 - 45.2)	5	51.3 (31.9 - 67.6)	1	56.7 (35.3 - 73.3)	0	56.7 (35.3 - 73.3)
C. diphtheriae	4	30.8 (8.9 - 56.3)	2	65.4 (15.3 - 90.8)	0	65.4 (15.3 - 90.8)		
S. pneumoniae	9	20.5 (10.0 - 33.5)	7	44.7 (27.0 - 60.9)	1	49.7 (30.2 - 66.5)	1	59.8 (31.6 - 79.4)

[1]CI indicates confidence interval. Seroresponse event defined as a 2-fold or greater increase in titers from pre-first vaccine dose. Yellow shading indicates cumulative incidence of seroresponse >50% for relevant participants and vaccine

Table S13. Number of Seroresponse Events and Cumulative Incidence Estimates Following Vaccine Doses Among Participants with Pre-vaccination B cell Counts <20 cells/μL

	Vaccine Dose									
	1		2		3		4		5	
Participant Category	**Number of Events[1]**	**Cumulative Incidence, % (95% CI)**	**Number of Events[1]**	**Cumulative Incidence, % (95% CI)**	**Number of Events[1]**	**Cumulative Incidence, % (95% CI)**	**Number of Events[1]**	**Cumulative Incidence, % (95% CI)**	**Number of Events[1]**	**Cumulative Incidence, % (95% CI)**
Pathogen										
Hepatitis A Virus (n=12)	2	16.7 (2.4 - 42.4)	0	16.7 (2.4 - 42.4)						
Hepatitis B Virus (n=21)	1	4.8 (0.3 - 20.2)	2	20.6 (4.3 - 45.2)	1	33.9 (7.5 - 63.6)		33.9 (7.5 - 63.6)	1	100.0
Haemophilus influenzae type B (n=23)	6	26.1 (10.4 - 45.1)	3	44.6 (21.3 - 65.6)	0	44.6 (21.3 - 65.6)				
C. diphtheriae (n=4)	0	0.0	1	50.0 (0.0 - 96.0)	0	50.0 (0.0 - 96.0)				
S. pneumoniae (n=27)	2	7.4 (1.2 - 21.4)	3	24.8 (8.3 - 45.8)	1	34.2 (11.7 - 58.5)	0	34.2 (11.7 - 58.5)		

[1]CI indicates confidence interval. Seroresponse event defined as a 2-fold or greater increase in titers from pre-first vaccine dose.

Table S14. Unadjusted and Adjusted Cox Model Estimates for Association of Post-CARTx, Pre-Vaccine Covariates with Seroresponse

Baseline Variable	Unadjusted HR (95% CI)	p-value	Adjusted HR[2] (95% CI)	p-value
CARTx target				
CD19/20	1.0 (Reference)		1.0 (Reference)	
BCMA	2.82 (1.05, 7.58)	0.040	2.46 (0.87, 6.95)	0.09
Time from CARTx to first vaccine dose				
per 1 month increase	1.01 (0.97, 1.04)	0.799		
$CD19^+$ B cell count				
<20 cells/μL	1.0 (Reference)	-	1.0 (Reference)	
≥20 cells/μL	3.54 (1.62, 7.75)	0.002	3.72 (1.72, 8.01)	<.001

HR indicates hazard ratio; CI, confidence interval. Estimates averaged across pathogens.

[2]Estimates adjusted for CARTx target, $CD19^+$ B cell count.

Table S15. Antibody Test Details and Interpretation of Antibody Titers to Twelve Evaluated Pathogens

Pathogen	Laboratory	Method	Test name, manufacturer	Threshold for positive result[1]
Hepatitis A virus (HAV)	University of Washington	Chemi-luminescence	Architect HAVAb-IgG, Abbott Diagnostics	≥1.0 signal/cutoff ratio
Hepatitis B virus (HBV), surface antibody	University of Washington	Chemi-luminescence	Architect AUSAB, Abbott Diagnostics	≥12 mIU/mL
Varicella zoster virus (VZV)	University of Washington	Chemi-luminescence	LIAISON VZV IgG HT, Diasorin	≥165.0 index value
Measles (Rubeola)	University of Washington	Chemi-luminescence	LIAISON RBIS IgG, Diasorin	≥16.5 AU/mL
Mumps	University of Washington	Chemi-luminescence	LIAISON MPIS IgG, Diasorin	≥11.0 AU/mL
Rubella	University of Washington	Chemi-luminescence	LIAISON VZV IgG, Diasorin	≥1.00 index value
Haemophilus influenzae type b	University of Washington	ELISA	VaccZyme Haemophilus influenzae type b IgG kit, Binding Site	>1.00 mg/L
Clostridium tetani	Mayo Clinic Laboratories	ELISA	Anti-Tetanus Toxoid ELISA (IgG), EuroImmun US	≥0.01 IU/mL
Corynebacterium diphtheriae	Mayo Clinic Laboratories	ELISA	Anti-Diphtheria Toxoid ELISA (IgG), EuroImmun US	≥0.01 IU/mL
Bordetella pertussis	Mayo Clinic Laboratories	ELISA	Anti-Bordetella pertussis toxin ELISA (IgG), EuroImmun US	≥100 IU/mL
Streptococcus pneumoniae	Mayo Clinic Laboratories	ELISA	Streptococcus pneumoniae IgG Antibodies, Total, with Reflex, Serum	> or =9.7 mcg/mL
Poliovirus (Types 1, 3)	Quest Diagnostics	Neutralization	Poliovirus (Types 1, 3) Antibodies, Quest Diagnostics	≥1:8 dilution

[1] Positive results are referred to as seroprotective for the purposes of this study.
ELISA indicates enzyme-linked immunosorbent assay; IFA, indirect fluorescence antibody.