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Supplementary Information for

Viral Genetic Diversity and Protective Efficacy of a Tetravalent Dengue Vaccine in Two Phase 3 Trials

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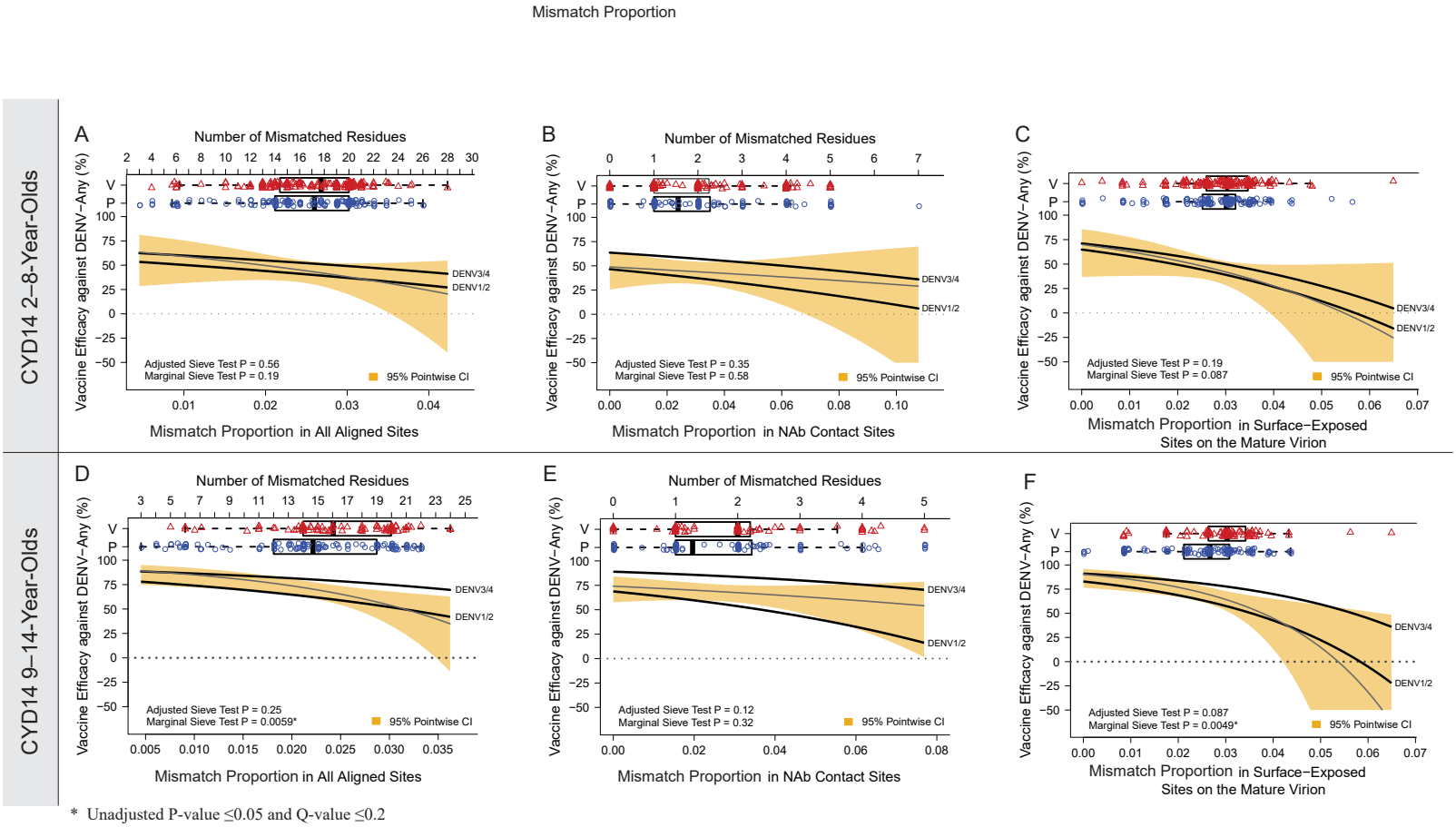
Supplementary text

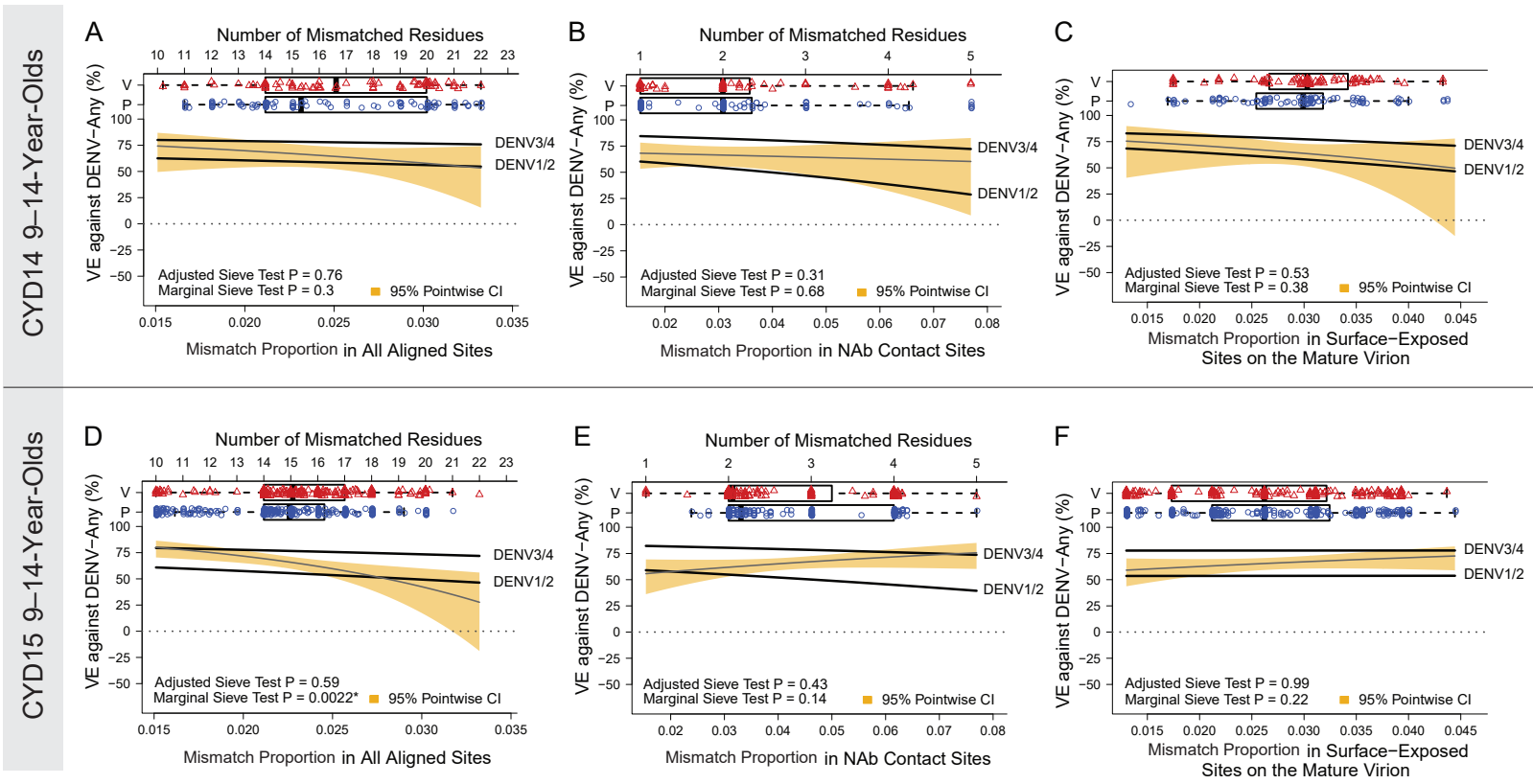
Sets of amino acid positions - solvent accessible

We analyzed sets of AA positions with a solvent accessibility surface (SAS) ≥ 0.25. The scores were assessed using atomic models built by homology modeling[1] based on the published M and E crystal structures of the mature virion. The SAS of each residue is calculated by comparison to a control tripeptide where the target residue is sandwiched between two glycine residues, defined as 100% accessibility. This calculation is performed for M/E in the mature virion state; the highest value (representing maximum accessibility) was used for analysis.



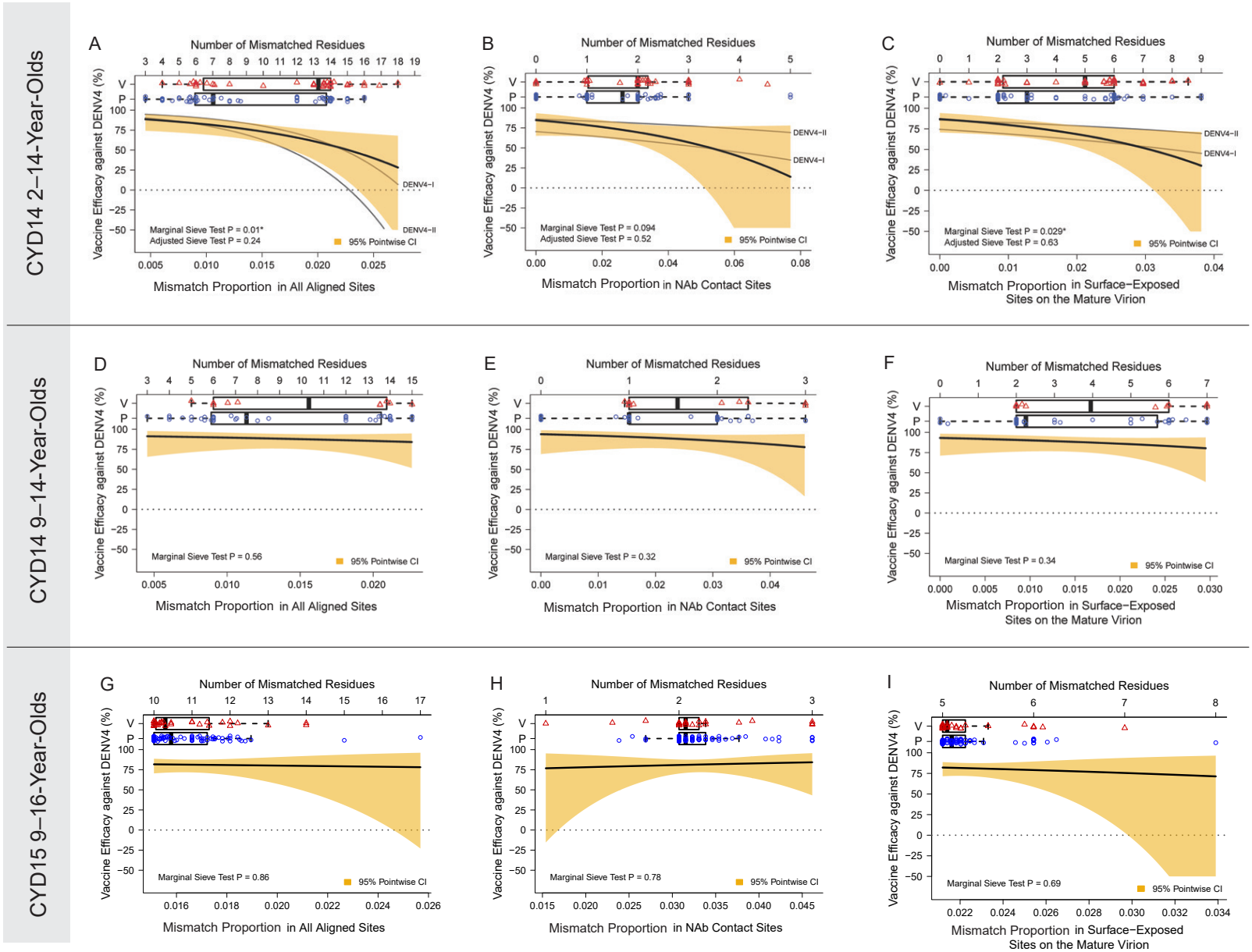
* Unadjusted P-value ≤0.05 and Q-value ≤0.2

Fig. S1. Vaccine efficacy against DENV-Any by percent residue mismatch in (A, D) all aligned sites†, (B, E) neutralizing antibody (NAb) contact sites, and (C‡, F‡) surface-exposed sites on the mature virion in the ITT cohort of CYD14 2−8-year-olds (A - C) and 9−14-year-olds (D – F). †The only alignment gap reflects that DENV3 sequences are characteristically missing AA at alignment positions 322 and 323. ‡The number of surface-exposed sites on the mature virion varies by serotype (229, 231, 225, and 236 for DENV1, DENV2, DENV3, and DENV4, respectively), hence no common top axis is displayed in panels C and F.



* Unadjusted P-value ≤0.05 and Q-value ≤0.2

Fig. S2. Vaccine efficacy against DENV-Any by percent residue mismatch, restricting to the common range of amino acid mismatches, in (A, D) all aligned sites†, (B, E) neutralizing antibody (NAb) contact sites, and (C‡, F‡) surface-exposed sites on the mature virion in the ITT cohort of CYD14 9−14-year-olds (A - C) and CYD15 9−14-year-olds (D – F). †The only alignment gap reflects that DENV3 sequences are characteristically missing AA at alignment positions 322 and 323. ‡The number of surface-exposed sites on the mature virion varies by serotype (229, 231, 225, and 236 for DENV1, DENV2, DENV3, and DENV4, respectively), hence no common top axis is displayed in panels C and F.



* Unadjusted P-value ≤0.05 and Q-value ≤0.2

Fig. S3. Vaccine efficacy against DENV4 by percent residue mismatch in: (A, D, G) all aligned sites; (B, E, H) neutralizing antibody (NAb) contact sites; and (C, F, I) surface-exposed sites on the mature virion, observed in: (A - C) the ITT cohort pooling over both age categories in CYD14; (D - F) the ITT cohort of 9−14-year-olds in CYD14; and (G - I) the ITT cohort of CYD15.

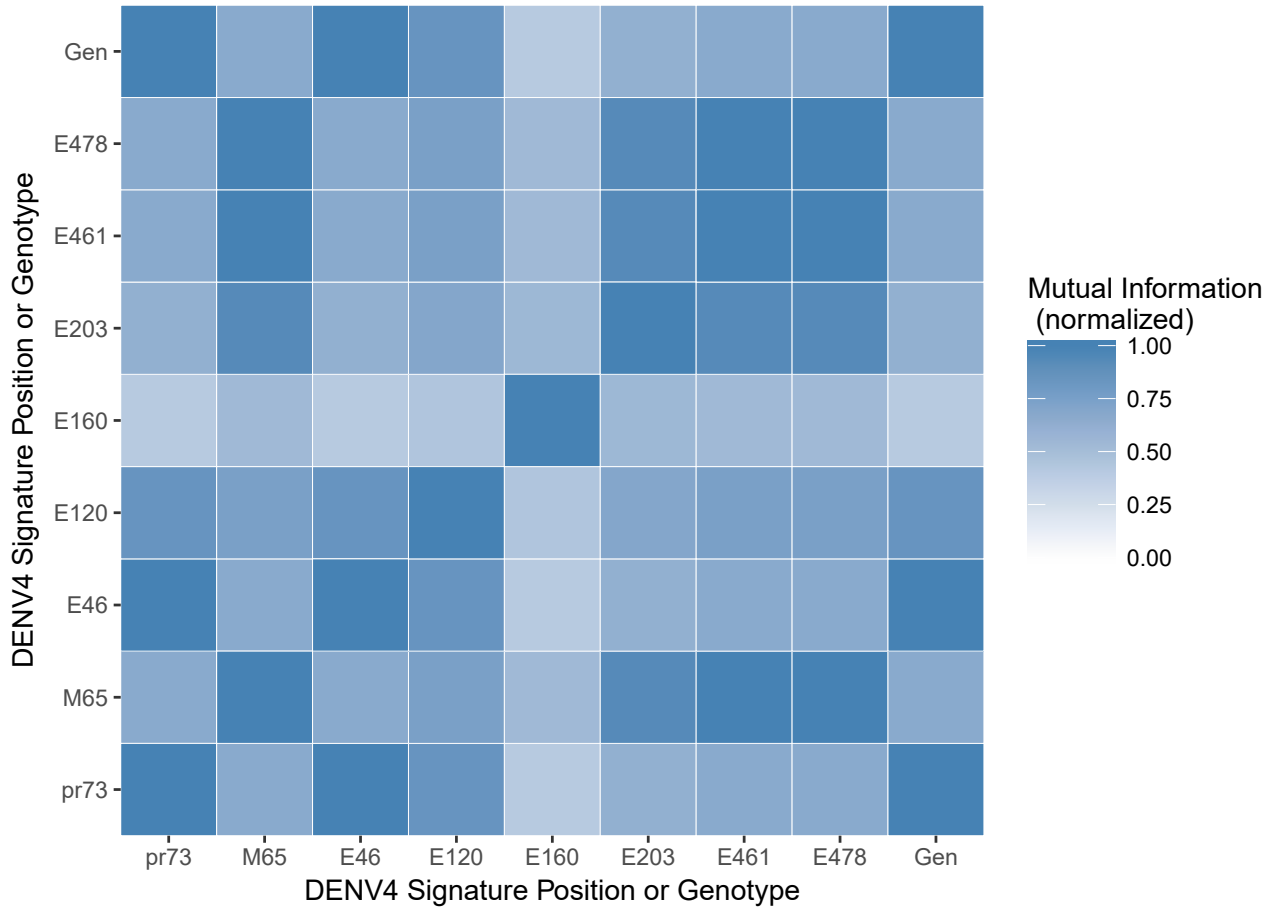
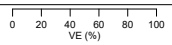


Fig. S4. Covariation between pairs of DENV4 signature positions in the ITT cohort of 2−8-year-olds. All unadjusted covariation test P-values were < 0.001.

A CYD14 2–14-Year-Olds

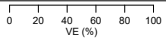
AA Position	Residue	Vaccine Residue Match/Mismatch	No. of Events (Inc. %) Vaccine	Placebo	Percent DENV4–I Events	VE (%)	95% CI	P–Value	Differential VE P–Value
pr73	T	Match	14 (0.10)	43 (0.63)	3.0	84.2	(70.6, 91.5)	<0.001	0.014 *
	A	Mismatch	26 (0.19)	29 (0.42)	100.0	55.0	(23.0, 73.7)	0.002	
M65	V	Match	16 (0.12)	44 (0.64)	9.0	81.9	(68.0, 89.8)	<0.001	0.033 *
	I	Mismatch	24 (0.17)	28 (0.41)	100.0	57.3	(26.3, 75.2)	0.001	
E46	T	Match	14 (0.10)	41 (0.60)	0.0	83.4	(69.2, 91.1)	<0.001	0.026 *
	I	Mismatch	26 (0.19)	31 (0.45)	100.0	57.8	(28.6, 75.0)	0.001	
E120	L	Match	15 (0.11)	41 (0.60)	3.0	82.3	(67.7, 90.3)	<0.001	0.042 *
	S	Mismatch	25 (0.18)	31 (0.45)	100.0	59.0	(30.4, 75.9)	0.001	
E160	V	Match	18 (0.13)	49 (0.71)	21.0	81.4	(67.5, 89.3)	<0.001	0.035 *
	E	Mismatch	22 (0.16)	23 (0.33)	97.0	53.1	(13.0, 74.7)	0.008	
E203	K	Match	16 (0.12)	44 (0.64)	9.0	81.9	(68.0, 89.8)	<0.001	0.033 *
	T,A	Mismatch	24 (0.17)	28 (0.41)	100.0	57.3	(26.3, 75.2)	0.001	
E461	F	Match	16 (0.12)	44 (0.64)	9.0	81.9	(68.0, 89.8)	<0.001	0.033 *
	L	Mismatch	24 (0.17)	28 (0.41)	100.0	57.3	(26.3, 75.2)	0.001	
E478	T	Match	16 (0.12)	44 (0.64)	9.0	81.9	(68.0, 89.8)	<0.001	0.033 *
	S	Mismatch	24 (0.17)	28 (0.41)	100.0	57.3	(26.3, 75.2)	0.001	



* Unadjusted P-value ≤0.05 and Q-value ≤0.2

B CYD14 9–14-Year-Olds

AA Position	Residue	Vaccine Residue Match/Mismatch	No. of Events (Inc. %) Vaccine	Placebo	Percent DENV4–I Events	VE (%)	95% CI	P–Value	Differential VE P–Value
pr73	T	Match	5 (0.07)	24 (0.72)	6.0	90.5	(73.0, 96.7)	<0.001	0.540
	A	Mismatch	5 (0.07)	17 (0.51)	100.0	84.0	(56.3, 94.2)	<0.001	
M65	V	Match	5 (0.07)	24 (0.72)	8.0	89.9	(73.4, 96.1)	<0.001	0.626
	I	Mismatch	5 (0.07)	17 (0.51)	100.0	84.7	(58.7, 94.4)	<0.001	
E46	T	Match	5 (0.07)	22 (0.66)	0.0	89.8	(70.9, 96.5)	<0.001	0.688
	I	Mismatch	5 (0.07)	19 (0.57)	100.0	85.5	(60.8, 94.6)	<0.001	
E120	L	Match	5 (0.07)	23 (0.69)	2.0	89.8	(71.8, 96.3)	<0.001	0.680
	S	Mismatch	5 (0.07)	18 (0.54)	100.0	85.4	(60.8, 94.5)	<0.001	
E160	V	Match	5 (0.07)	24 (0.72)	11.0	89.3	(70.2, 96.2)	<0.001	0.795
	E	Mismatch	5 (0.07)	18 (0.54)	94.0	86.0	(57.5, 95.4)	<0.001	
E203	K	Match	5 (0.07)	24 (0.72)	8.0	89.9	(73.4, 96.1)	<0.001	0.626
	T,A	Mismatch	5 (0.07)	17 (0.51)	100.0	84.7	(58.7, 94.4)	<0.001	
E461	F	Match	5 (0.07)	24 (0.72)	8.0	89.9	(73.4, 96.1)	<0.001	0.626
	L	Mismatch	5 (0.07)	17 (0.51)	100.0	84.7	(58.7, 94.4)	<0.001	
E478	T	Match	5 (0.07)	24 (0.72)	8.0	89.9	(73.4, 96.1)	<0.001	0.626
	S	Mismatch	5 (0.07)	17 (0.51)	100.0	84.7	(58.7, 94.4)	<0.001	



C CYD15 9–16-Year-Olds

AA Position	Residue	Vaccine Residue Match/Mismatch	No. of Events (Inc. %) Vaccine	Placebo	Percent DENV4–I Events	VE (%)	95% CI	P–Value	Differential VE P–Value
pr73	T	Match	29 (0.11)	82 (0.61)	0	82.9	(73.8, 88.8)	<0.001	---
	–	Mismatch	0 (0.00)	0 (0.00)	–	--	---	---	
M65	V	Match	29 (0.11)	82 (0.61)	0	82.9	(73.8, 88.8)	<0.001	---
	–	Mismatch	0 (0.00)	0 (0.00)	–	--	---	---	
E46	T	Match	29 (0.11)	82 (0.61)	0	82.9	(73.8, 88.8)	<0.001	---
	–	Mismatch	0 (0.00)	0 (0.00)	–	--	---	---	
E120	–	Match	0 (0.00)	0 (0.00)	–	--	---	---	---
	S	Mismatch	29 (0.11)	82 (0.61)	0	82.9	(73.8, 88.8)	<0.001	
E160	V	Match	26 (0.10)	77 (0.57)	0	83.8	(74.4, 89.7)	<0.001	0.121
	E	Mismatch	3 (0.01)	5 (0.04)	0	67.6	(−95.5, 94.6)	0.109	
E203	K	Match	27 (0.10)	78 (0.58)	0	83.0	(73.6, 89.0)	<0.001	0.208
	T	Mismatch	2 (0.01)	4 (0.03)	0	80.2	(−110.1, 98.1)	0.088	
E461	F	Match	29 (0.11)	82 (0.61)	0	82.9	(73.8, 88.8)	<0.001	---
	–	Mismatch	0 (0.00)	0 (0.00)	–	--	---	---	
E478	T	Match	29 (0.11)	82 (0.61)	0	82.9	(73.8, 88.8)	<0.001	---
	–	Mismatch	0 (0.00)	0 (0.00)	–	--	---	---	

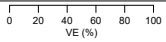


Fig. S5. Vaccine efficacy (VE) against DENV4 with a vaccine-matched and mismatched residue at signature amino acid (AA) positions in the ITT cohorts of (A) CYD14, pooling over both age categories; (B) CYD14, 9–14-year-olds; and (C) CYD15.

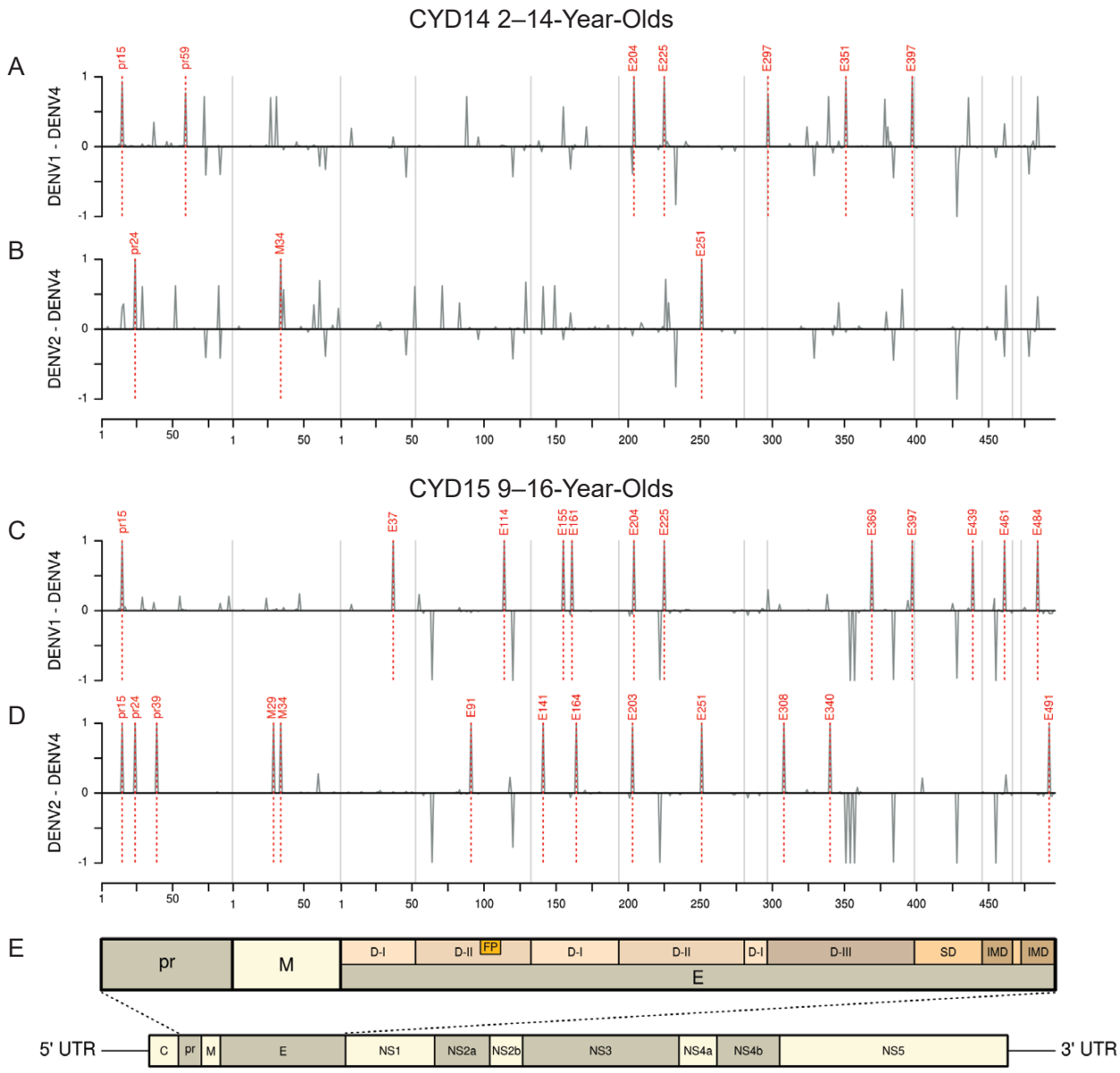


Fig. S6. Difference in the prevalence of vaccine-mismatched residues comparing (A) DENV1 to DENV4 placebo case sequences pooling over both age categories in CYD14, (B) DENV2 to DENV4 placebo case sequences in the ITT cohort pooling over both age categories in CYD14, (C) DENV1 to DENV4 placebo case sequences in the ITT cohort of CYD15, and (D) DENV2 to DENV4 placebo case sequences in the ITT cohort of CYD15. Panel (E) shows a schematic diagram of the DENV genome, with an enlarged version of the prM/E region showing the domain structure of the E protein. FP, fusion peptide. SD, stem domain. IMD, intramembrane domain. UTR, untranslated region.

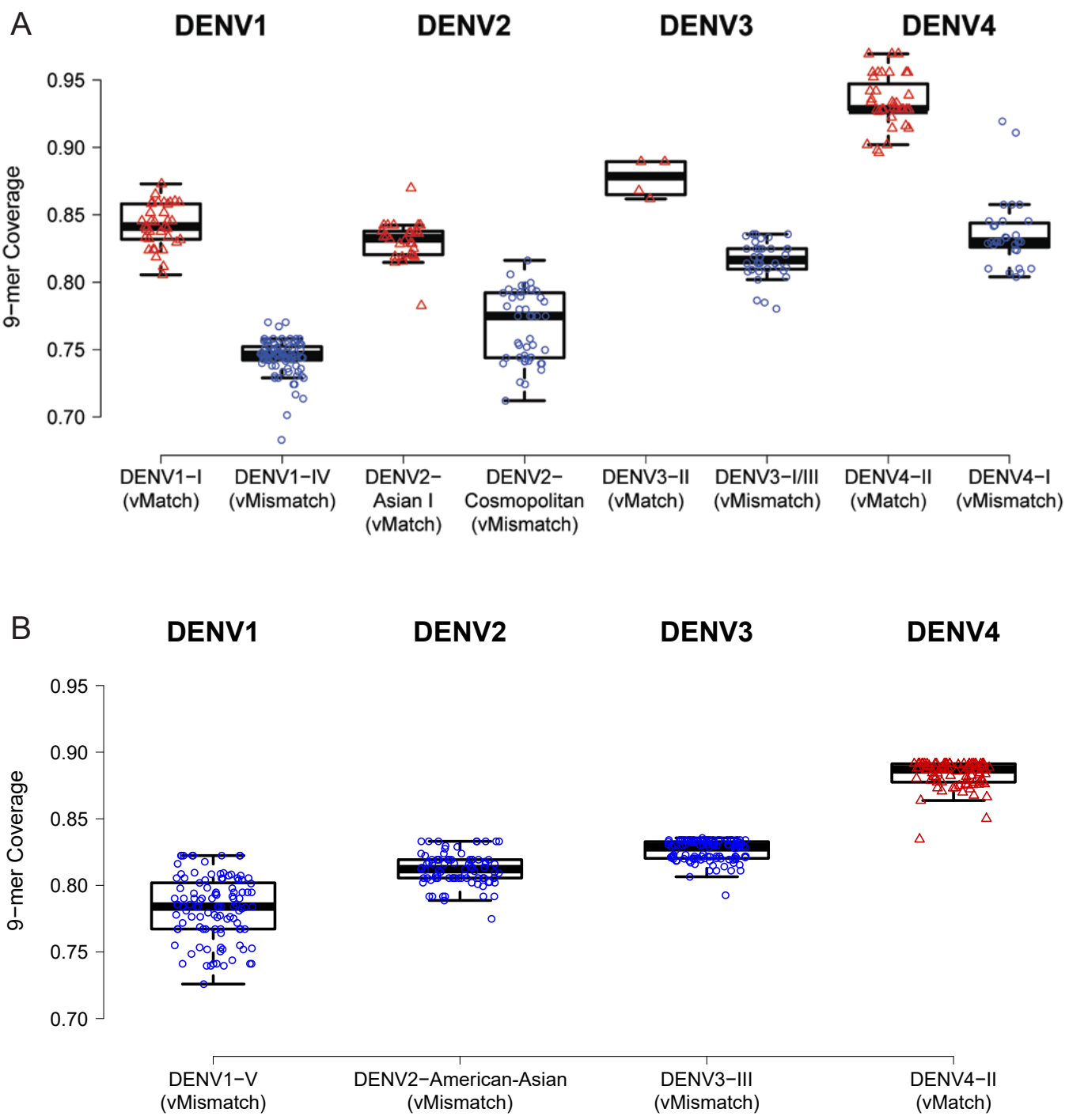


Fig. S7. 9-mer coverage of placebo case sequences by the vaccine strains in the ITT cohorts of (A) CYD14, pooling over both age categories, and (B) CYD15.

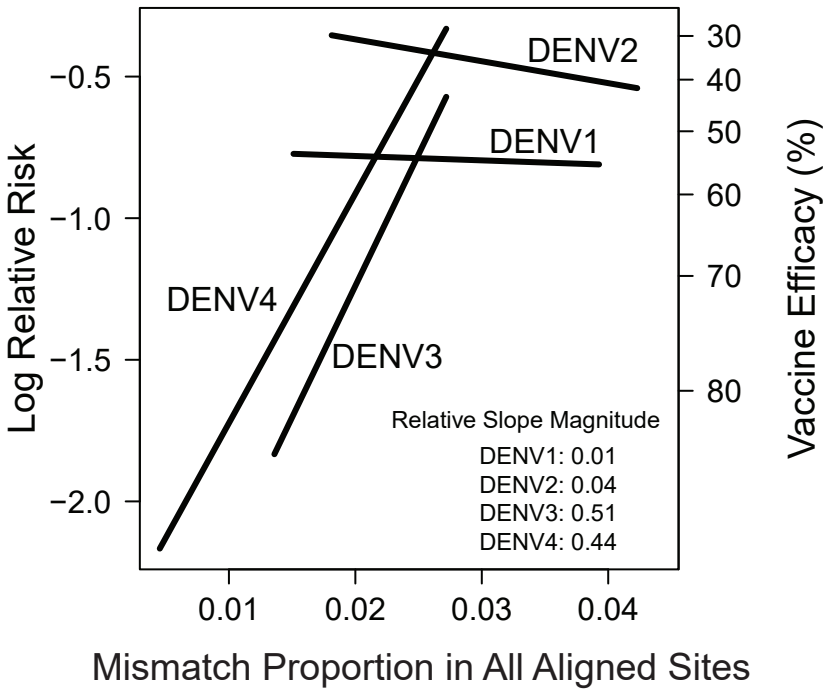


Fig. S8. Log relative risks of the serotype-specific dengue endpoints by residue mismatch proportion in all aligned AA positions in the ITT cohort of CYD14 2−14-year-olds. The relative slope magnitude for serotype i is defined as |si|/(|s1|+|s2|+|s3|+|s4|), where si is the linear slope of the curve for serotype i.

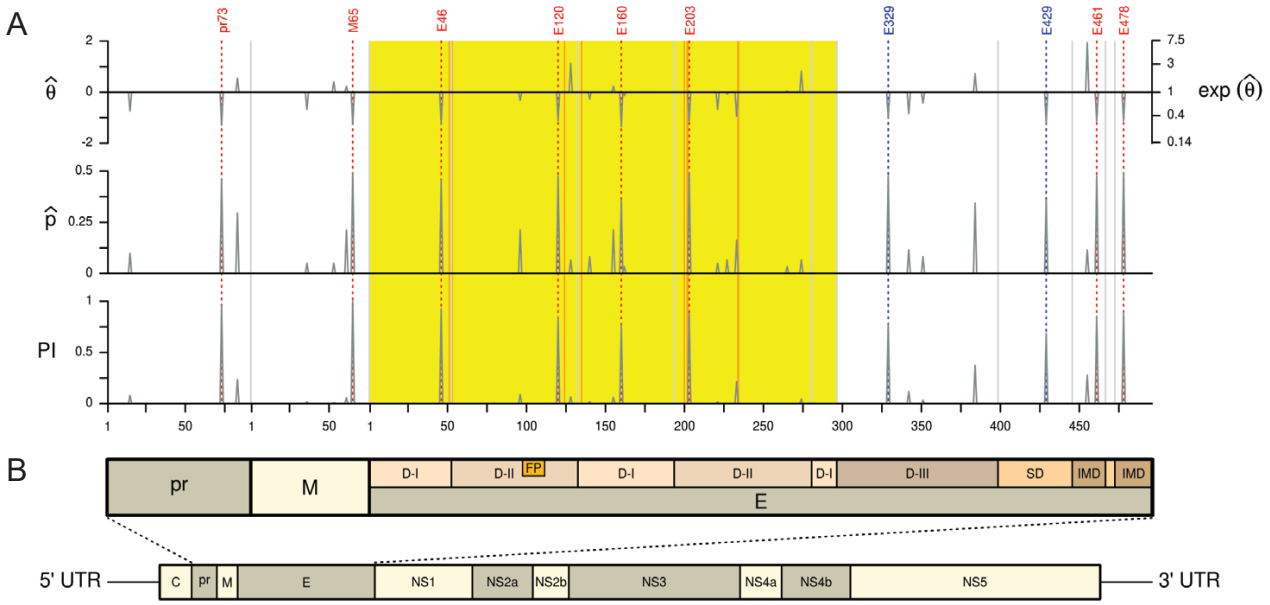


Fig. S9. Influence of amino acid positions on the DENV4 sieve effect in the ITT cohort of 2−8-year-olds. (A) Influence of amino acid positions on the DENV4 sieve effect, and (B) schematic diagram of the DENV genome. The top panel of (A) shows the degree of estimated differential vaccine efficacy (VE) against DENV4 with a residue match vs. mismatch to the vaccine at amino acid position *i* indexed on the x-axis, measured as the estimated log hazard ratio (vaccine/placebo) of matched-DENV4 minus this estimate for mismatched-DENV4 (left y-axis label), and the right y-axis label is the ratio of hazard ratios measuring how much greater VE is against matched than mismatched DENV4. The middle panel of (A) shows the estimated prevalence of the minority residue (match or mismatch) at position *i* among DENV4 placebo recipient cases, which measures the diversity of the position in circulating DENV4 sequences and hence the degree of influence that differential VE at the position can potentially have on overall VE against DENV4. The third panel of (A) shows the amino acid position influence (PI), which multiplies a 0−1 scaled version of the top panel value with a 0−1 scaled version of the middle panel value, and then is again re-scaled between 0−1. A higher value of PI indicates that the amino acid position has more influence on the overall VE against DENV4. Signature positions where VE was significantly greater against DENV4 match than mismatch are indicated by vertical red dotted lines. An additional influential position with greater VE against DENV4 match than mismatch and PI ≥ 0.25 is indicated by a vertical blue dotted line. Epitopes of two isolated DENV4-specific human monoclonal neutralizing antibodies, DV4-126 and DV4-131, were mapped to the region indicated in yellow.[2] Residues at positions indicated by vertical orange solid lines were identified as critical for DV4-126 and DV4-131 binding to DENV4. (B) shows a schematic diagram of the DENV genome, with an enlarged version of the prM/E region showing the domain structure of the E protein. FP, fusion peptide. SD, stem domain. IMD, intramembrane domain. UTR, untranslated region.

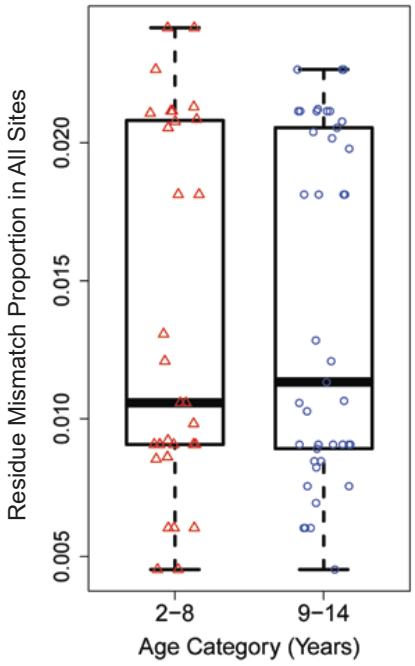


Fig. S10. Residue mismatch proportion to the DENV4 vaccine sequence for all sites in the placebo group's DENV4 ITT cases by age category in CYD14.

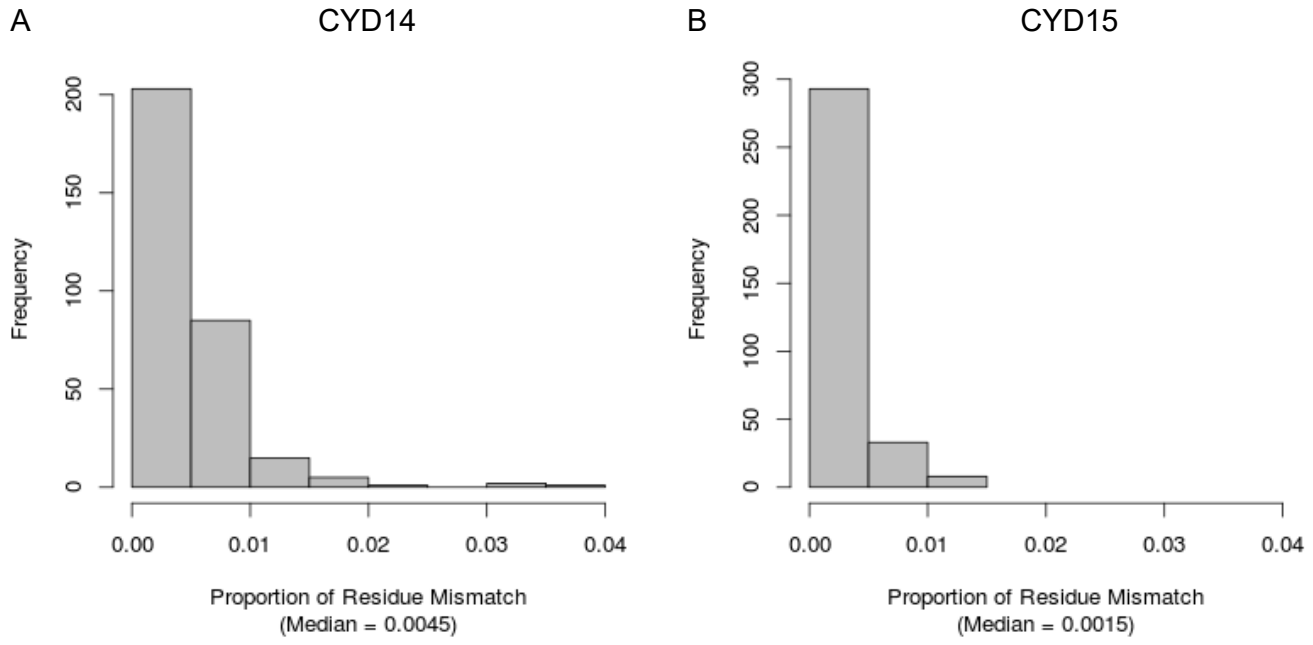


Fig. S11. Distribution of mean proportion residue mismatch between complete sequences observed in proximal virologically confirmed dengue (VCD) cases in the ITT cohorts of (A) CYD14 and (B) CYD15. Mean proportion residue mismatch between complete sequences observed in proximal VCD cases in the ITT cohorts of CYD14 and CYD15 was calculated by pooling serotype-matched observed sequences from the most-geographically constrained region (in increasing order: clinic vs. site vs. country) according to the specified sequence imputation process. The accuracy of the sequence imputation process was evaluated by assessing the mean proportion residue mismatch between each observed sequence and the respective observed proximal sequences as determined by each of the three described algorithms. In this analysis, amino acid positions with missing flanking residues were ignored.

Table S1. Numbers of virologically confirmed dengue (VCD) endpoint cases in the ITT cohorts of (A) CYD14 and (B) CYD15 with complete and imputed sequence data. (C) Amino acid positions with sufficient variability in the ITT cohort of CYD14.

A. Numbers of VCD endpoint cases in the ITT cohort of CYD14 with complete and imputed sequence data

All Study Participants

	Cases with Complete Seqs		Cases with Imputed Seqs		Imputation Level							
					Clinic		Site		Country		Excluded	
Endpoint	V	P	V	P	V	P	V	P	V	P	V	P
DENV-Any	147	165	131	134	117	120	11	9	3	5	1	3
DENV1	55	69	61	57	58	56	3	1	0	0	0	0
DENV2	56	36	41	38	34	32	7	5	0	1	0	0
DENV3	13	25	16	15	14	11	0	2	2	2	1	3
DENV4	27	42	13	30	11	27	1	1	1	2	0	0

2−8-Year-Old Participants Only

	Cases with Complete Seqs		Cases with Imputed Seqs		Imputation Level							
					Clinic		Site		Country		Excluded	
Endpoint	V	P	V	P	V	P	V	P	V	P	V	P
DENV-Any	107	89	84	78	72	70	10	6	2	2	0	1
DENV1	41	42	39	32	36	31	3	1	0	0	0	0
DENV2	39	23	25	25	19	22	6	3	0	0	0	0
DENV3	8	14	11	10	10	9	0	1	1	0	0	1
DENV4	21	16	9	15	7	12	1	1	1	2	0	0

9−14-Year-Old Participants Only

	Cases with Complete Seqs		Cases with Imputed Seqs		Imputation Level							
					Clinic		Site		Country		Excluded	
Endpoint	V	P	V	P	V	P	V	P	V	P	V	P
DENV-Any	40	76	47	56	45	50	1	3	1	3	1	2
DENV1	14	27	22	25	22	25	0	0	0	0	0	0
DENV2	17	13	16	13	15	10	1	2	0	1	0	0
DENV3	5	11	5	5	4	2	0	1	1	2	1	2
DENV4	6	26	4	15	4	15	0	0	0	0	0	0

B. Numbers of VCD endpoint cases in the ITT cohort of CYD15 with complete and imputed sequence data

All Study Participants

	Cases with Complete Seqs		Cases with Imputed Seqs		Imputation Level							
					Clinic		Site		Country		Excluded	
Endpoint	V	P	V	P	V	P	V	P	V	P	V	P
DENV-Any	134	199	125	173	125	169	0	0	0	4	3	1
DENV1	53	76	46	33	46	33	0	0	0	0	0	0
DENV2	48	50	35	33	35	33	0	0	0	0	0	0
DENV3	23	47	32	59	32	59	0	0	0	0	0	0
DENV4	11	31	18	51	18	47	0	0	0	4	3	1

C. Amino acid positions with sufficient variability in the ITT cohort of CYD14

Endpoint	All	NAb Contact Sites	SAS Mat	Positions with Sufficient Variability
DENV-Any	133	22	65	pr5, pr13, pr15, pr16, pr17, pr24, pr29, pr31, pr37, pr38, pr46, pr49, pr52, pr55, pr57, pr59, pr72, pr73, pr81, pr82, pr83, M16, M24, M27, M31, M34, M36, M37, M44, M45, M48, M52, M53, M57, M60, M61, M65, M70, M74, M75, E8[2], E26[2], E27[2], E28[2], E37[2], E46[2], E52[1,2], E55[2], E68[2], E71[2], E81[2], E83[1,2], E88[2], E96[2], E108[2], E120[2], E124[1,2], E128[2], E129[2], E132[2], E138[2], E140, E141, E147[2], E149[2], E154[1,2], E155[1,2], E160[2], E162[1], E163[1,2], E171[2], E174[1,2], E177[2], E186[2], E203[1,2], E204[2], E209[2], E210[2], E221, E222[2], E224[2], E225[2], E226[1,2], E227[1,2], E228[2], E233[2], E240, E241[2], E251[2], E272[2], E273[1,2], E274[1,2], E297[2], E303[1,2], E305[1,2], E312[1], E324, E329[1,2], E331[2], E338[1,2], E339, E342[2], E343[2], E345[2], E346[2], E348[2], E351[2], E359[2], E360[1,2], E378, E379, E380, E382[2], E384[1,2], E385[1,2], E390[1,2], E397, E425, E428, E429, E432, E436, E454, E455, E461, E462, E475, E478, E480, E481, E484, E492, E495
DENV1	48	4	14	pr13, pr17, pr29, pr37, pr38, pr46, pr49, pr59, pr72, pr83, M24, M27, M31, M45, M52, M57, M65, E8[2], E37[2], E88[2], E96[2], E112, E113, E132, E138, E155[1,2], E171, E227[1,2], E228[2], E240, E241[2], E297[2], E312[1], E324, E331[2], E338[1,2], E339, E346[2], E359[2], E378, E380, E382[2], E436, E461, E475, E480, E481, E484
DENV2	42	6	23	pr5, pr15, pr16, pr29, pr31, pr52, pr82, M36, M48, M57, M60, M70, M74, E26[2], E27, E28[2], E46[2], E52[1,2], E71[2], E83[1,2], E108[2], E129[2], E141, E149[2], E160[2], E177[2], E186[2], E203[1,2], E209[2], E210[2], E224[2], E226[1,2], E228[2], E343[2], E345[2], E346[2], E360[1,2], E379, E390[1,2], E425, E462, E484
DENV3	25	8	11	pr55, pr57, M16, M37, M44, E68[2], E81[2], E124[1,2], E132, E140, E154[1,2], E162[1], E171[2], E174[1,2], E233[2], E272, E273[1,2], E303[1,2], E305[1,2], E379, E385[1,2], E454, E461, E481, E492
DENV4	28	7	11	pr15, pr73, pr83, M36, M53, M61, M65, E46[2], E96[2], E120[2], E128, E140, E155[1,2], E160, E162[1], E203[1,2], E221, E227[1,2], E233[2], E274[1,2], E329[1,2], E342[2], E351, E384[1,2], E429, E455, E461, E478

D. Amino acid positions with sufficient variability in the ITT cohort of CYD15

Endpoint	All	NAb Contact Sites	SAS Mat	Positions with Sufficient Variability
DENV-Any	89	16	44	pr13, pr15, pr17, pr24, pr29, pr37, pr39, pr55, pr57, pr58, pr83, pr89, M24, M29, M31, M34, M36, M47, M60, M69, M75, E8[2], E28[2], E37[2], E55[2], E58[1,2], E64[1,2], E81[2], E83[1,2], E91, E114, E118[2], E120[2], E124[1,2], E132[2], E139, E141, E147[2], E154[1,2], E155[1,2], E160[2], E161[1,2], E164, E170[2], E171[2], E201, E203[1,2], E204[2], E222[2], E225[2], E229[2], E251[2], E283, E291[2], E293[1,2], E297[2], E303[1,2], E304[2], E305[1,2], E308[1,2], E324, E331[2], E338[1,2], E340[2], E347[2], E351[2], E354[2], E357[2], E359[2], E360[1,2], E369, E384[1,2], E385[1,2], E394[2], E397, E404, E425, E428, E436, E439, E454, E455, E461, E462, E475, E480, E484, E492, E495
DENV1	25	4	7	pr13, pr17, pr29, pr37, pr55, pr83, pr89, M24, M31, M36, M47, E8[2], E55[2], E83[1,2], E293[1,2], E297[2], E305[1], E338[1,2], E394[2], E425, E428, E436, E439, E454, E475
DENV2	11	1	6	M60, E28[2], E83[1,2], E118, E120[2], E170[2], E324, E347[2], E359[2], E404, E462
DENV3	6	0	2	pr58, M69, E132, E139, E304[2], E331[2]
DENV4	8	2	3	pr15, E58[1], E160, E201, E203[1,2], E229[2], E283, E291[2]

*V = Vaccine; P = Placebo.

[1] Neutralizing antibody (NAb) contact site

[2] Surface-accessible (SAS ≥ 0.25) position on the mature virion

Table S2. Vaccine efficacy against DENV2 with a vaccine-matched and mismatched residue at amino acid position E226 in the ITT cohorts of (A) CYD14, 2−8-year-olds; (B) CYD14, 9−14-year-olds; (C) CYD14, pooling over both age categories; and (D) CYD15.

A. CYD14 2−8-Year-Old Participants Only				
Residue	**Vaccine Residue Match/Mismatch**	**VE (%)**	**95% CI**	**Differential VE P-Value**
T	Match	66.0	(34.5, 82.4)	0.015
K	Mismatch	1.7	(-60.5, 39.7)	
B. CYD14 9−14-Year-Old Participants Only				
Residue	**Vaccine Residue Match/Mismatch**	**VE (%)**	**95% CI**	**Differential VE P-Value**
T	Match	-35.5	(-763.1, 78.7)	0.40
K	Mismatch	43.5	(1.7, 67.5)	
C. CYD14 Pooled Over Both Age Categories				
Residue	**Vaccine Residue Match/Mismatch**	**VE (%)**	**95% CI**	**Differential VE P-Value**
T	Match	57.0	(17.8, 77.5)	0.15
K	Mismatch	25.6	(-5.8, 47.7)	
D. CYD15				
Residue	**Vaccine Residue Match/Mismatch**	**VE (%)**	**95% CI**	**Differential VE P-Value**
T	Match	50.7	(32.9, 63.7)	-
K	Mismatch	33.1	(-1109.0, 96.3)	

Table S3. Estimated ratios between (A) serotypes of the mean percent residue mismatch[1] in the ITT cohort of CYD14 (left) and the ITT cohort of CYD15 (right), (B) vaccine-matched vs. mismatched genotypes of the mean percent residue mismatch[1] in CYD14, and (C) serotypes of the vaccine strains' mean 9-mer coverage[2] of placebo recipient dengue sequences in the ITT cohort of CYD14 (left) and CYD15 (right)

A. Estimated ratios between serotypes of the mean percent residue mismatch

Percent residue mismatch in all aligned sites*

CYD14			CYD15		
Comparison	**Ratio of Means (%)**	**95% CI**	**Comparison**	**Ratio of Means (%)**	**95% CI**
DENV1 / DENV2	1.01	(0.96, 1.06)	DENV1 / DENV2	1.09	(1.06, 1.11)
DENV1 / DENV3	1.27	(1.21, 1.34)	DENV1 / DENV3	1.20	(1.17, 1.22)
DENV1 / DENV4	2.01	(1.80, 2.26)	DENV1 / DENV4	1.58	(1.53, 1.62)
DENV2 / DENV3	1.26	(1.19, 1.33)	DENV2 / DENV3	1.10	(1.08, 1.12)
DENV2 / DENV4	1.99	(1.78, 2.24)	DENV2 / DENV4	1.45	(1.41, 1.49)
DENV3 / DENV4	1.58	(1.41, 1.78)	DENV3 / DENV4	1.32	(1.29, 1.35)

Percent residue mismatch in NAb contact sites

CYD14			CYD15		
Comparison	**Ratio of Means (%)**	**95% CI**	**Comparison**	**Ratio of Means (%)**	**95% CI**
DENV1 / DENV2	0.35	(0.31, 0.41)	DENV1 / DENV2	1.16	(1.10, 1.22)
DENV1 / DENV3	0.29	(0.25, 0.34)	DENV1 / DENV3	0.60	(0.57, 0.63)
DENV1 / DENV4	0.64	(0.53, 0.78)	DENV1 / DENV4	1.13	(1.07, 1.19)
DENV2 / DENV3	0.83	(0.71, 0.96)	DENV2 / DENV3	0.52	(0.50, 0.53)
DENV2 / DENV4	1.80	(1.50, 2.21)	DENV2 / DENV4	0.97	0.94, 1.01)
DENV3 / DENV4	2.18	(1.79, 2.68)	DENV3 / DENV4	1.87	(1.82, 1.93)

Percent residue mismatch in surface-exposed sites on the mature virion

CYD14			CYD15		
Comparison	**Ratio of Means (%)**	**95% CI**	**Comparison**	**Ratio of Means (%)**	**95% CI**
DENV1 / DENV2	0.85	(0.80, 0.90)	DENV1 / DENV2	1.99	(1.89, 2.10)
DENV1 / DENV3	0.94	(0.88, 1.01)	DENV1 / DENV3	0.91	(0.87, 0.94)
DENV1 / DENV4	1.90	(1.63, 2.27)	DENV1 / DENV4	1.40	(1.35, 1.45)
DENV2 / DENV3	1.11	(1.04, 1.19)	DENV2 / DENV3	0.45	(0.43, 0.48)
DENV2 / DENV4	2.24	(1.91, 2.69)	DENV2 / DENV4	0.70	(0.67, 0.74)
DENV3 / DENV4	2.02	(1.72, 2.42)	DENV3 / DENV4	1.55	(1.51, 1.59)

B. Estimated ratios between vaccine-matched vs. mismatched genotypes of the mean percent residue mismatch in CYD14

Percent residue mismatch in all aligned sites*

Comparison	**Ratio of Means (%)**	**95% CI**
DENV1-I / DENV1-IV	0.64	(0.61, 0.67)
DENV2-Asian I / DENV2-Cosmopolitan	0.79	(0.74, 0.83)
DENV3-II / DENV3-I/III	0.68	(0.56, 0.80)
DENV4-II / DENV4-I	0.43	(0.39, 0.47)

Percent residue mismatch in NAb contact sites

Comparison	**Ratio of Means (%)**	**95% CI**
DENV1-I / DENV1-IV	0.47	(0.33, 0.62)
DENV2-Asian I / DENV2-Cosmopolitan	0.7	(0.61, 0.81)
DENV3-II / DENV3-I/III	–†	–
DENV4-II / DENV4-I	0.43	(0.39, 0.48)

Percent residue mismatch in surface-exposed sites on the mature virion

Comparison	**Ratio of Means (%)**	**95% CI**
DENV1-I / DENV1-IV	0.73	(0.67, 0.80)
DENV2-Asian I / DENV2-Cosmopolitan	0.98	(0.90, 1.07)
DENV3-II / DENV3-I/III	0.64	(0.52, 0.77)
DENV4-II / DENV4-I	0.29	(0.23, 0.36)

C. Estimated ratios between serotypes of the mean 9-mer coverage

CYD14			CYD15		
Comparison	**Ratio of Means (%)**	**95% CI**	**Comparison**	**Ratio of Means (%)**	**95% CI**
DENV2 / DENV1	1.02	(1.01, 1.04)	DENV2 / DENV1	1.04	(1.03, 1.04)
DENV3 / DENV1	1.07	(1.05, 1.08)	DENV3 / DENV1	1.05	(1.05, 1.06)
DENV4 / DENV1	1.15	(1.13, 1.17)	DENV4 / DENV1	1.13	(1.12, 1.13)
DENV3 / DENV2	1.04	(1.03, 1.05)	DENV3 / DENV2	1.02	(1.01, 1.02)
DENV4 / DENV2	1.13	(1.11, 1.15)	DENV4 / DENV2	1.09	(1.08, 1.09)
DENV4 / DENV3	1.08	(1.06, 1.10)	DENV4 / DENV3	1.07	(1.07, 1.07)

[1] The mean percent residue mismatch is estimated based on placebo recipient dengue AA sequences to the amino acid sequence encoded by the corresponding serotype vaccine insert.
[2] The mean 9-mer coverage refers to the mean proportion of 9-mers in placebo recipient dengue sequences with a match in the vaccine sequence of the same serotype.
* The only alignment gap reflects that DENV3 sequences are characteristically missing AA at alignment positions 322 and 323.
† A constant percent residue mismatch in DENV3-II placebo cases
AA, amino acid.

Table S4. Numbers of VCD endpoint cases in (A) 2-8 year-olds and (B) 9-14 year-olds in the ITT cohort of CYD14.

A. 2-8 year-old participants in CYD14					
		DENV-Any Endpoint		Serotype-Specific Endpoint	
		Vaccine	Placebo	Vaccine	Placebo
Serotype	Genotype	N (%)	N (%)	N (%)	N (%)
Total		196	173	-	-
DENV1		80 (40.8)	70 (40.5)	80	74
	I*	9 (11.2)	9 (12.9)	9 (11.2)	10 (13.5)
	IV	32 (40.0)	31 (44.3)	32 (40.0)	32 (43.2)
	Missing†	39 (48.8)	30 (42.9)	39 (48.8)	32 (43.2)
DENV2		63 (32.1)	44 (25.4)	64	48
	Asian I*	15 (23.8)	6 (13.6)	16 (25.0)	6 (12.5)
	Cosmopolitan	23 (36.5)	14 (31.8)	23 (35.9)	17 (35.4)
	Missing†	25 (39.7)	24 (54.5)	25 (39.1)	25 (52.1)
DENV3		19 (9.7)	25 (14.5)	19	25
	I	5 (26.3)	8 (32.0)	5 (26.3)	8 (32.0)
	II*	0 (0.0)	1 (4.0)	0 (0.0)	1 (4.0)
	III	3 (15.8)	5 (20.0)	3 (15.8)	5 (20.0)
	Missing†	11 (57.9)	11 (44.0)	11 (57.9)	11 (44.0)
DENV4		29 (14.8)	29 (16.8)	30	31
	I	16 (55.2)	6 (20.7)	16 (53.3)	6 (19.4)
	II*	4 (13.8)	9 (31.0)	5 (16.7)	10 (32.3)
	Missing†	9 (31.0)	14 (48.3)	9 (30.0)	15 (48.4)
Missing‡		5 (2.6)	5 (2.9)	-	-

B. 9-14 year-old participants in CYD14					
		DENV-Any Endpoint		Serotype-Specific Endpoint	
		Vaccine	Placebo	Vaccine	Placebo
Serotype	Genotype	N (%)	N (%)	N (%)	(%)
Total		90	136	-	-
DENV1		36 (40.0)	49 (36.0)	36	52
	I*	6 (16.7)	7 (14.3)	6 (16.7)	8 (15.4)
	IV	8 (22.2)	19 (38.8)	8 (22.2)	19 (36.5)
	Missing†	22 (61.1)	23 (46.9)	22 (61.1)	25 (48.1)
DENV2		31 (34.4)	26 (19.1)	33	26
	Asian I*	11 (35.5)	9 (34.6)	12 (36.4)	9 (34.6)
	Cosmopolitan	4 (12.9)	4 (15.4)	5 (15.2)	4 (15.4)
	Missing†	16 (51.6)	13 (50.0)	16 (48.5)	13 (50.0)
DENV3		11 (12.2)	18 (13.2)	11	18
	I	4 (36.4)	6 (33.3)	4 (36.4)	6 (33.3)
	II*	0 (0.0)	3 (16.7)	0 (0.0)	3 (16.7)
	III	1 (9.1)	2 (11.1)	1 (9.1)	2 (11.1)
	Missing†	6 (54.5)	7 (38.9)	6 (54.5)	7 (38.9)
DENV4		10 (11.1)	41 (30.1)	10	41
	I	3 (30.0)	12 (29.3)	3 (30.0)	12 (29.3)
	II*	3 (30.0)	14 (34.1)	3 (30.0)	14 (34.1)
	Missing†	4 (40.0)	15 (36.6)	4 (40.0)	15 (36.6)
Missing‡		2 (2.2)	2 (1.5)	-	-

* This genotype is contained in the vaccine.

† Sequences were unobtainable due to either a DENV titer below the lower limit of quantification, insufficient volume of unthawed serum, or a lack of participant consent. Some participants with VCD were found to be positive for more than one serotype. These participants were included in the analysis for each appropriate endpoint. For the DENV-Any endpoint, the sequence from the earliest-documented VCD event was included for analysis.

‡ Participants were confirmed as VCD cases, but the infecting strains could not be positively serotyped. These participants were not included in the analysis, as their sequences could not be imputed without serotype information.

References

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