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QHR79135.1     MAVILNKRYYSNLLILILMISECSVGILHYEKLSKVGLVKGITRKYKIKSNPLTKDIVIK    60
QHR79216.1     MAVILNKRYYSNLLILILIISECSIGILHYEKLSKIGLVKGITRKYKIKSNPLTKDIVIK    60
AWT50993.1     MAVILNKRYYSNLLLLILMISECSVGILHYEKLSKIGLVKGITRKYKIKSNPLTKDIVIK    60
ACT32614.1     MAVILNKRYYSNLLILILIISECSVGILHYEKLSKIGLVKGITRKYKIKSNPLTKDIVIK    60
QHR79180.1     MAVILNKRYYSNLLILILMISECSVGILHYEKLSKIGLVKGITRKYKIKSNPLTKDIVIK    60
QHR79045.1     MAVILNKRYYSNLLILILMISECSVGILHYEKLSKIGLVKGITRKYKIKSNPLTKDIVIK    60
AEZ01396.1     MAVILNKRYYSNLLILILMISECSVGILHYEKLSKIGLVKGITRKYKIKSNPLTKDIVIK    60
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UZQ22166.1     MIPNVSNMSQCTGSVMENYKTRLNGILTPIKGALEIYKNNTHDLVGDVRLAGVIMAGVAI   120
QHR79135.1     MIPNVSNMSQCTGSVMENYKTRLNGILTPIKGALEIYKNNTHDLVGDVRLAGVIMAGVAI   120
QHR79216.1     MIPNVSNMSQCTGSVMENYKTRLNGILTPIKGALEIYKNNTHDLVGDVRLAGVIMAGVAI   120
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ACT32614.1     MIPNVSNMSQCTGSVMENYKTRLNGILTPIKGALEIYKNNTHDLVGDVRLAGVIMAGVAI   120
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QHR79045.1     MIPNVSNMSQCTGSVMENYKTRLNGILTPIKGALEIYKNNTHDLVGDVRLAGVIMAGVAI   120
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AWT50993.1     GIATAAQITAGVALYEAMKNADNINKLKSSIESTNEAVVKLQETAEKTVYVLTALQDYIN   180
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QHR79045.1     GIATAAQITAGVALYEAMKNADNINKLKSSIESTNEAVVKLQETAEKTVYVLTALQDYIN   180
AEZ01396.1     GIATAAQITAGVALYEAMKNADNINKLKSSIESTNEAVVKLQETAEKTVYVLTALQDYIN   180
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UZQ22166.1     TNLVPTIDKISCKQTELSLDLALSKYLSDLLFVFGPNLQDPVSNSMTIQAISQAFGGNYE   240
QHR79135.1     TNLVPTIDKISCKQTELSLDLALSKYLSDLLFVFGPNLQDPVSNSMTIQAISQAFGGNYE   240
QHR79216.1     TNLVPTIDKISCKQTELSLDLALSKYLSDLLFVFGPNLQDPVSNSMTIQAISQAFGGNYE   240
AWT50993.1     TNLVPTIDKISCKQTELSLDLALSKYLSDLLFVFGPNLQDPVSNSMTIQAISQAFGGNYE   240
ACT32614.1     TNLVPTIDKISCKQTELSLDLALSKYLSDLLFVFGPNLQDPVSNSMTIQAISQAFGGNYE   240
QHR79180.1     TNLVPTIDKISCKQTELSLDLALSKYLSDLLFVFGPNLQDPVSNSMTIQAISQAFGGNYE   240
QHR79045.1     TNLVPTIDKISCKQTELSLDLALSKYLSDLLFVFGPNLQDPVSNSMTIQAISQAFGGNYE   240
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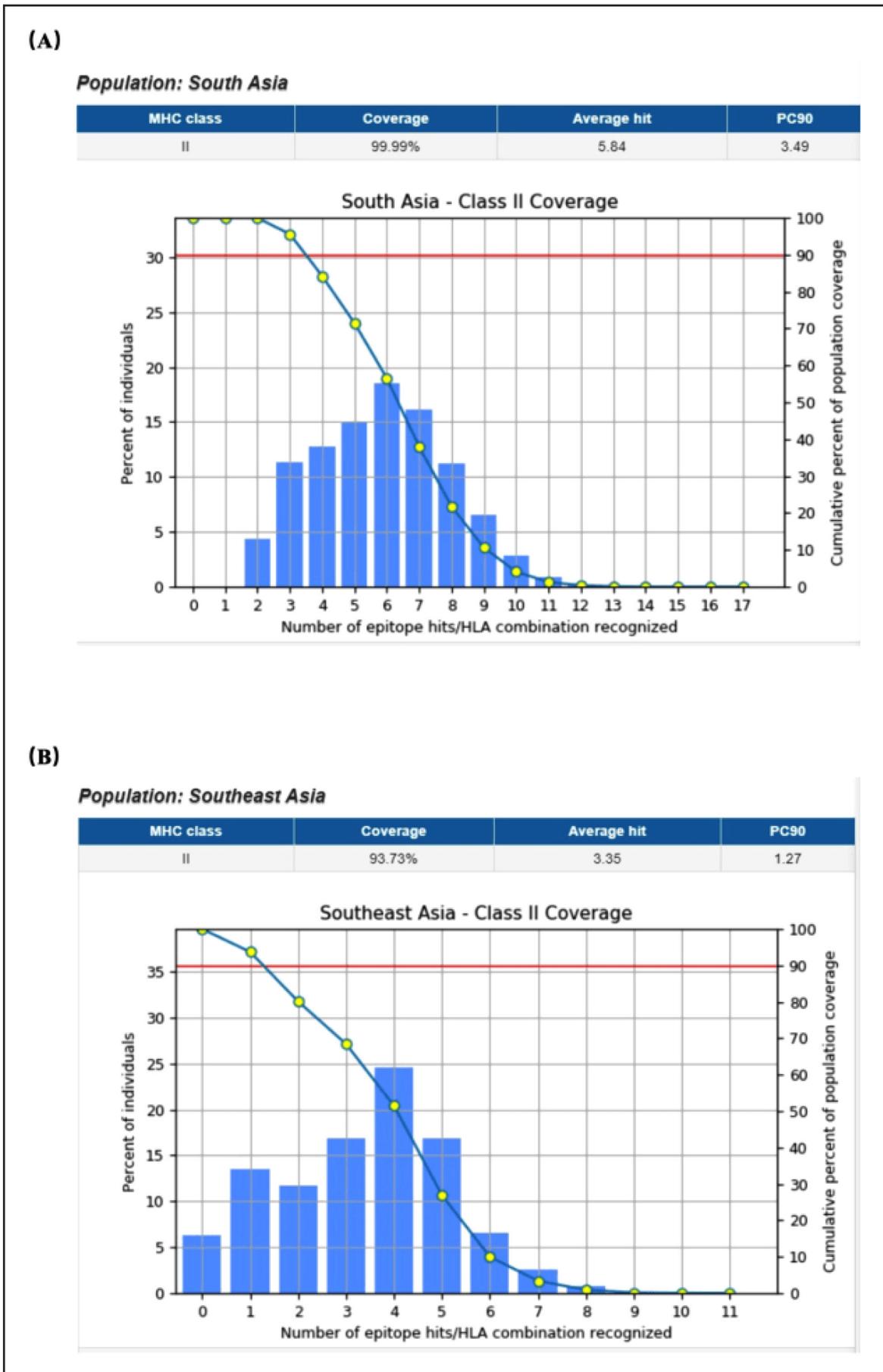
Figure S1. Multiple alignment of NiV F protein amino acid sequences from Homo sapiens originating from different countries performed on EMBL-EBI server with Clustal Omega multiple sequence alignment tool.

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QHR79135.1     TLLRTLGYTTEDFDDLLESDSITGQIIYVDLSGYYIIVRVYFPILTEIQQAYIQELLPVS    300
QHR79216.1     TLLRTLGYATEDFDDLLESDSITGQIIYVDLSGYYIIVRVYFPILTEIQQAYIQELLPVS    300
AWT50993.1     TLLRTLGYATEDFDDLLESDSITGQIIYVDLSGYYIIVRVYFPILTEIQQAYIQELLPVS    300
ACT32614.1     TLLRTLGYATEDFDDLLESDSITGQIIYVDLSGYYIIVRVYFPILTEIQQAYIQELLPVS    300
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QHR79045.1     TLLRTLGYATEDFDDLLESDSITGQIIYVDLSSYYIIVRVYFPILTEIQQAYIQELLPVS    300
AEZ01396.1     TLLRTLGYATEDFDDLLESDSITGQIIYVDLSGYYIIVRVYFPILTEIQQAYIQELLPVS    300
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UZQ22166.1     FNNDNSEWISIVPNFILVRNTLISNIEIGFCLITKRSVICNQDYATPMTNNMRECLTGST    360
QHR79135.1     FNNDNSEWISIVPNFILVRNTLISNIEIGFCLITKRSVICNQDYATPMTNNMRECLTGST    360
QHR79216.1     FNNDNSEWISIVPNFILVRNTLISNIEIGFCLITKRSVICNQDYATPMTNNMRECLTGST    360
AWT50993.1     FNNDNSEWISIVPNFILVRNTLISNIEIGFCLITKRSVICNQDYATPMTNNMRECLTGST    360
ACT32614.1     FNNDNSEWISIVPNFILVRNTLISNIEIGFCLITKRSVICNQDYATPMTNNMRECLTGST    360
QHR79180.1     FNNDNSEWISIVPNFILVRNTLISNIEIGFCLITKRSVICNQDYATPMTNNMRECLTGST    360
QHR79045.1     FNNDNSEWISIVPNFILVRNTLISNIEIGFCLITKRSVICNQDYATPMTNNMRECLTGST    360
AEZ01396.1     FNNDNSEWISIVPNFILVRNTLISNIEIGFCLITKRSVICNQDYATPMTNNMRECLTGST    360
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UZQ22166.1     EKCPRELVVSSHVPRFALSNGVLFANCISVTCQCQTTGRAISQSGEQTLLMIDNTTCPTA    420
QHR79135.1     EKCPRELVVSSHVPRFALSNGVLFANCISVTCQCQTTGRAISQSGEQTLLMIDNTTCPTA    420
QHR79216.1     EKCPRELVVSSHVPRFALSNGVLFANCISVTCQCQTTGRAISQSGEQTLLMIDNTTCPTA    420
AWT50993.1     EKCPRELVVSSHVPRFALSNGVLFANCISVTCQCQTTGRAISQSGEQTLLMIDNTTCPTA    420
ACT32614.1     EKCPRELVVSSHVPRFALSNGVLFANCISVTCQCQTTGRAISQSGEQTLLMIDNTTCPTA    420
QHR79180.1     EKCPRELVVSSHVPRFALSNGVLFANCISVTCQCQTTGRAISQSGEQTLLMIDNTTCPTA    420
QHR79045.1     EKCPRELVVSSHVPRFALSNGVLFANCISVTCQCQTTGRAISQSGEQTLLMIDNTTCPTA    420
AEZ01396.1     EKCPRELVVSSHVPRFALSNGVLFANCISVTCQCQTTGRAISQSGEQTLLMIDNTTCPTA    420
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UZQ22166.1     VLGNVIISLGKYLGSVNYNSEGIAIGPPVFTDKVDISSQISSMNQSLQQSKDYIKEAQRL    480
QHR79135.1     VLGNVIISLGKYLGSVNYNSEGIAIGPPVFTDKVDISSQISSMNQSLQQSKDYIKEAQRL    480
QHR79216.1     VLGNVIISLGKYLGSVNYNSEGIAIGPPVFTDKVDISSQISSMNQSLQQSKDYIKEAQRL    480
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QHR79180.1     VLGNVIISLGKYLGSINYNSEGIAIGPPVFTDKVDISSQISSMNQSLQQSKDYIKEAQRL    480
QHR79045.1     VLGNVIISLGKYLGSVNYNSEGIAIGPPVFTDKVDISSQISSMNQSLQQSKDYIKEAQRL    480
AEZ01396.1     VLGNVIISLGKYLGSVNYNSEGIAIGPPVFTDKVDISSQISSMNQSLQQSKDYIKEAQRL    480
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QHR79216.1     LDTVNPSLISMLSMIILYVLSIASLCIGLITFISFIIVEKKRNTYSRLEDRRVRPTSSGD    540
AWT50993.1     LDTVNPSLISMLSMIILYVLSIASLCIGLITFISFIIVEKKRNTYSRLEDRRVRPTSSGD    540
ACT32614.1     LDTVNPSLISMLSMIILYVLSIASLCIGLITFISFIIVEKKRNTYSRLEDRRVRPTSSGD    540
QHR79180.1     LDTVNPSLISMLSMIILYVLSIASLCIGLITFISFIIVEKKRNTYSRLEDRRVRPTSSGD    540
QHR79045.1     LDTVNPSLISMLSMIILYVLSIASLCIGLITFISFIIVEKKRNTYSRLEDRRVRPTSSGD    540
AEZ01396.1     LDTVNPSLISMLSMIILYVLSIASLCIGLITFISFIIVEKKRNTYSRLEDRRVRPTSSGD    540
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AAY43915.1     LYYIGT    546
UZQ22166.1     LYYIGT    546
QHR79135.1     LYYIGT    546
QHR79216.1     LYYIGT    546
AWT50993.1     LYYIGT    546
ACT32614.1     LYYIGT    546
QHR79180.1     LYYIGT    546
QHR79045.1     LYYIGT    546
AEZ01396.1     LYYIGT    546
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Figure S1 *(continue)*. Multiple alignment of NiV F protein amino acid sequences from Homo sapiens originating from different countries performed on EMBL-EBI server with Clustal Omega multiple sequence alignment tool.
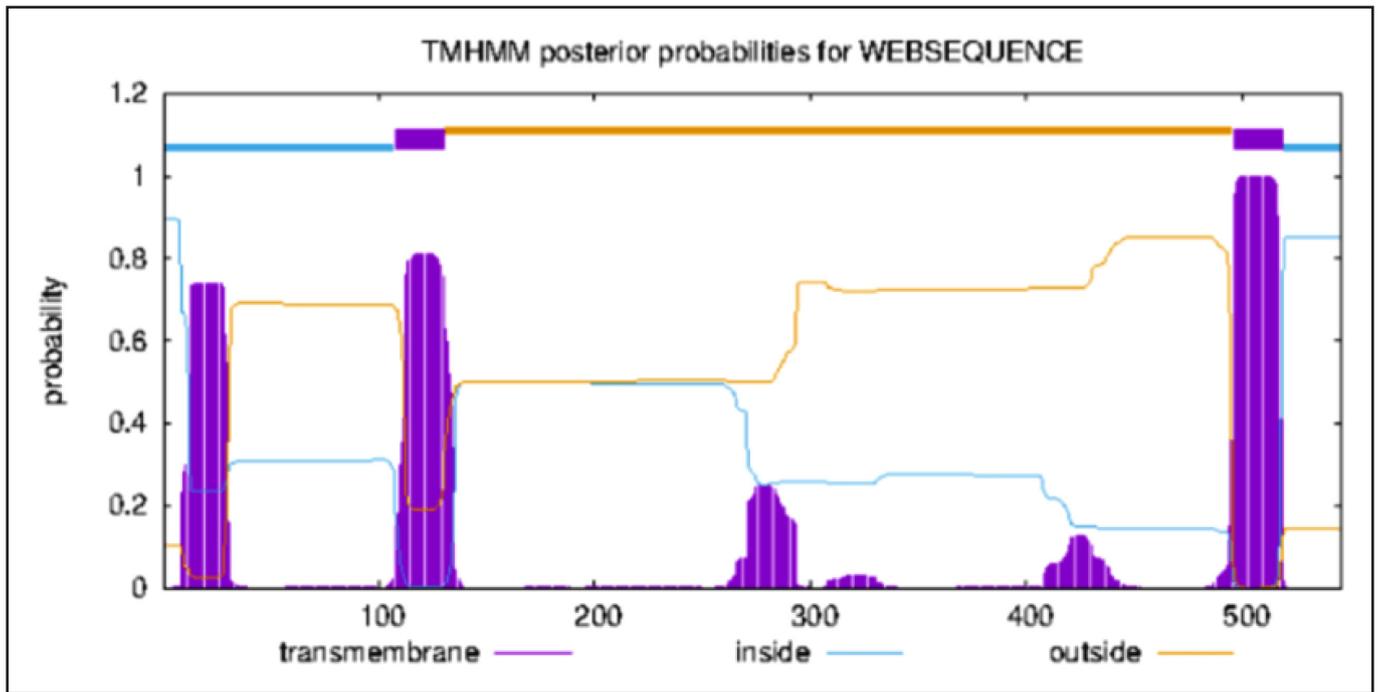
Figure S2. Result of surface availability of NiV F protein. Orange-coloured horizontal line on x-axis marks the region outside the viral membrane.

(A)

Population: South Asia

MHC class	Coverage	Average hit	PC90
II	99.99%	5.84	3.49

(B)

Population: Southeast Asia

MHC class	Coverage	Average hit	PC90
II	93.73%	3.35	1.27

Figure S3. South and Southeast Asia population coverage graphs by IEDB analysis resource, evaluation based on MHC-II NiV F protein proposed epitopes. (A) Population coverage of South Asia and (B) population coverage of Southeast Asia. The line (-o-) represents the cumulative percentage of population coverage of the epitopes; the bars represent the population coverage for each epitope.
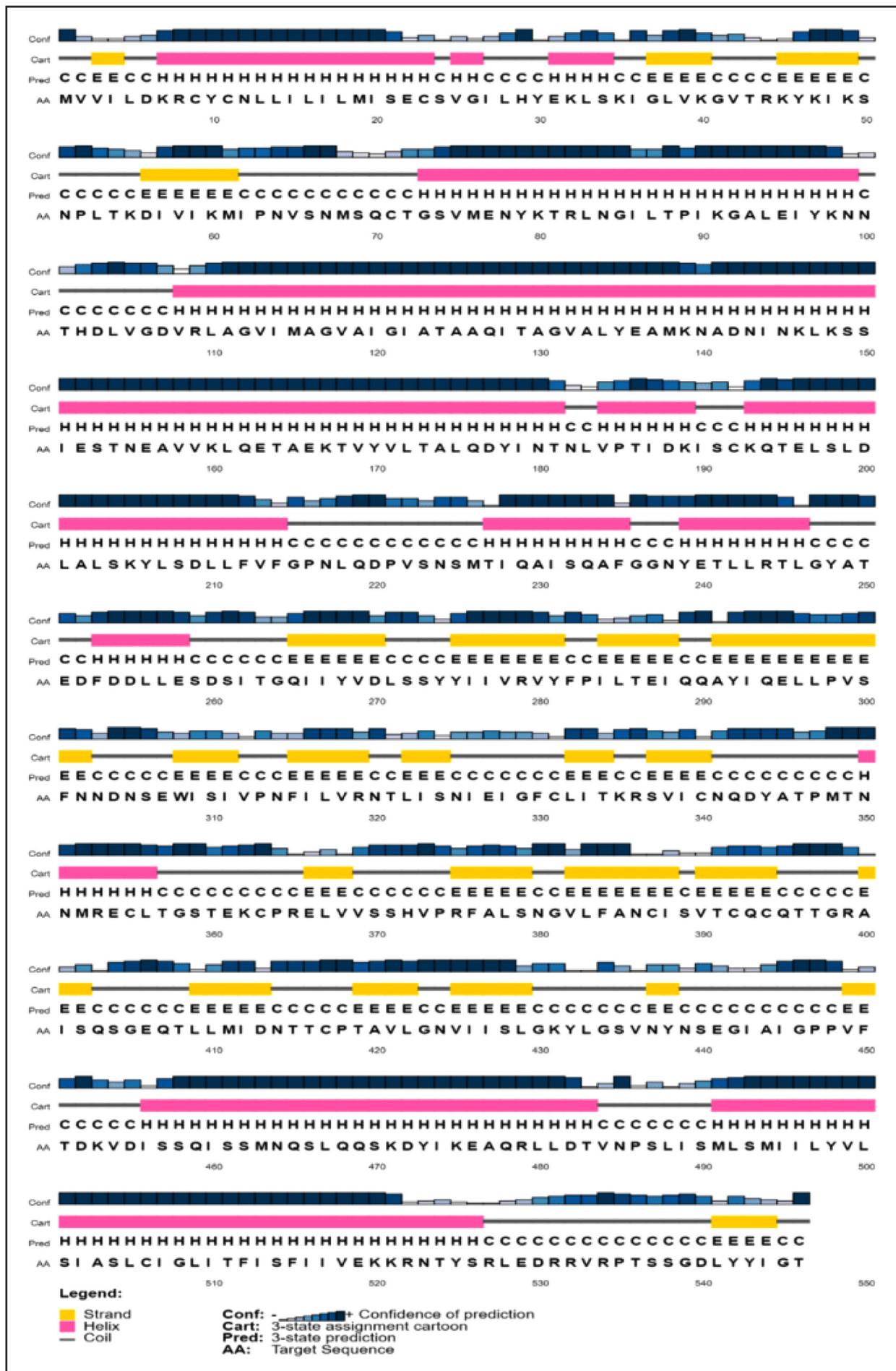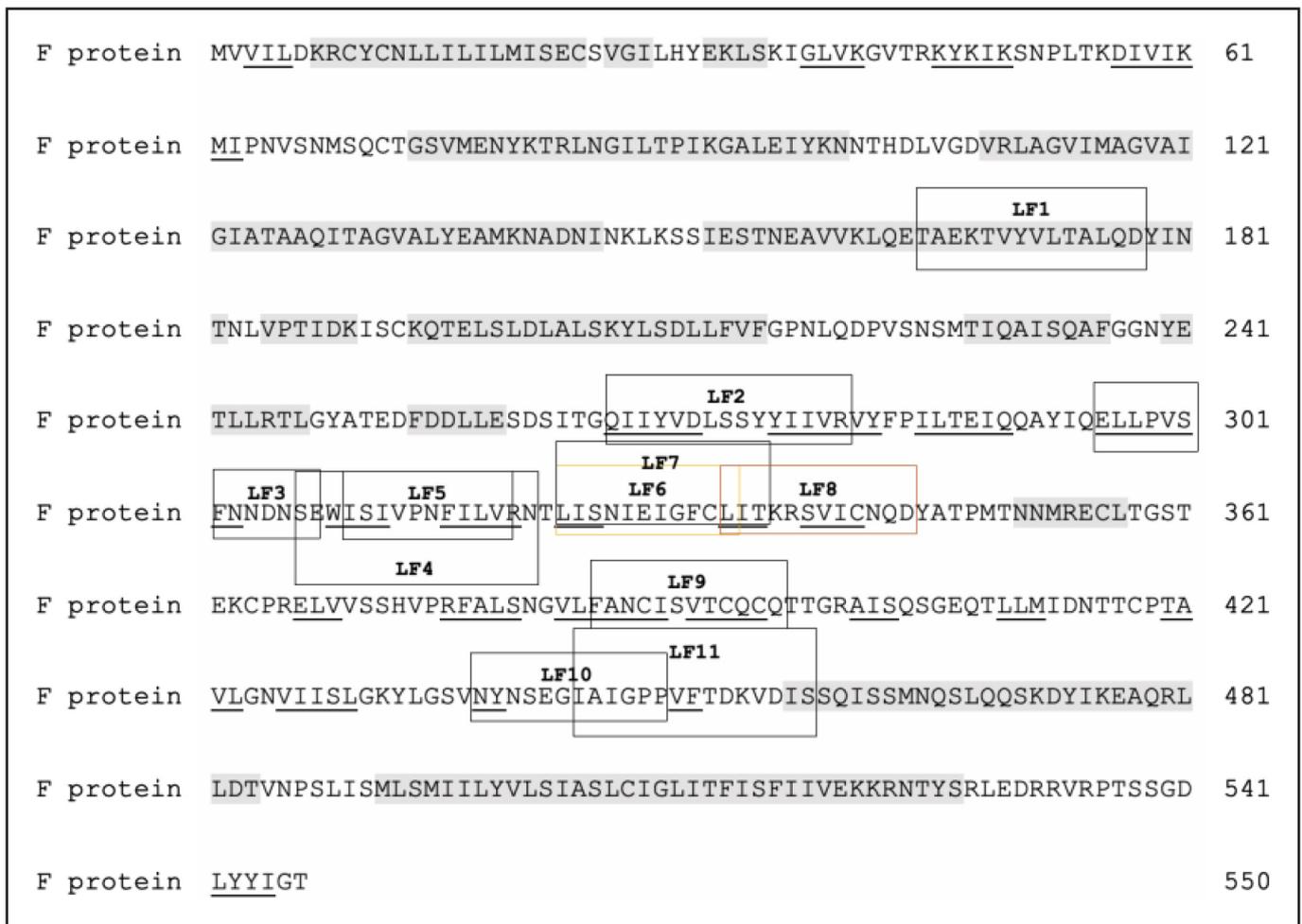
Figure S4. PSIPRED secondary structure prediction result for NiV F protein.

Figure S5. Alignment summary of secondary structure and amino acid positions of each candidate epitopes in the F protein. α-helices are underlined; β-sheets are shaded in grey; random coils are unmarked; predicted epitopes as shown within text boxes.

Table S1. The most promising helper T cell epitopes with their MHC-II alleles

F protein	MHC-II alleles
HF1	HLA-DPA1*01:03/DPB1*02:01, HLA-DPA1*02:01/DPB1*01:01, HLA-DPA1*02:02/DPB1*05:01
HF2	HLA-DQA1*05:01/DQB1*05:02, HLA-DQA1*05:01/DQB1*02:01, HLA-DPA1*02:02/DPB1*05:01, HLA-DQA1*01:01/DQB1*05:01, HLA-DQA1*04:01/DQB1*04:02, HLA-DPA1*01:03/DPB1*02:01, HLA-DPA1*02:01/DPB1*01:01, HLA-DPA1*03:01/DPB1*04:02
HF3	HLA-DPA1*02:01/DPB1*01:01, HLA-DPA1*02:02/DPB1*05:01HLA-DRB3*01:01

Table S2. Population coverage results of the South and Southeast Asia from the most promising epitopes of MHC-II according to F protein

Population	MHC class II		
	Coverage[a]	Average_hit[b]	pc90[c]
South Asia	99.99%	5.84	3.49
Southeast Asia	93.73%	3.35	1.27
Average	96.86%	4.59	2.38

[a]Population coverage.
[b]Average number of epitope hits/HLA combinations recognized by the population.
[c]Minimum number of epitope hits/HLA combinations recognized by 90% of the population.

Table S3. Results of the repeatability assay for LF6-iELISA. The intra-assay coefficient of variability (CV) was determined by calculating the standard deviation of the means divided the means of 3 replicates for each serum sample. Inter-assay variability was calculated by the standard deviation of plate means/overall mean OD from the plate means for each serum sample

Sample number	Inter-assay CV (%)		Intra-assay CV (%)	
	X ± SD	CV (%)	X ± SD	CV (%)
1	0.080 ± 0.007	9.253	0.069 ± 0.003	4.389
2	0.082 ± 0.005	5.739	0.079 ± 0.004	5.492
3	0.113 ± 0.005	4.440	0.343 ± 0.026	7.613
4	0.111 ± 0.009	7.746	0.088 ± 0.001	0.726
5	0.361 ± 0.015	4.035	0.076 ± 0.005	6.539