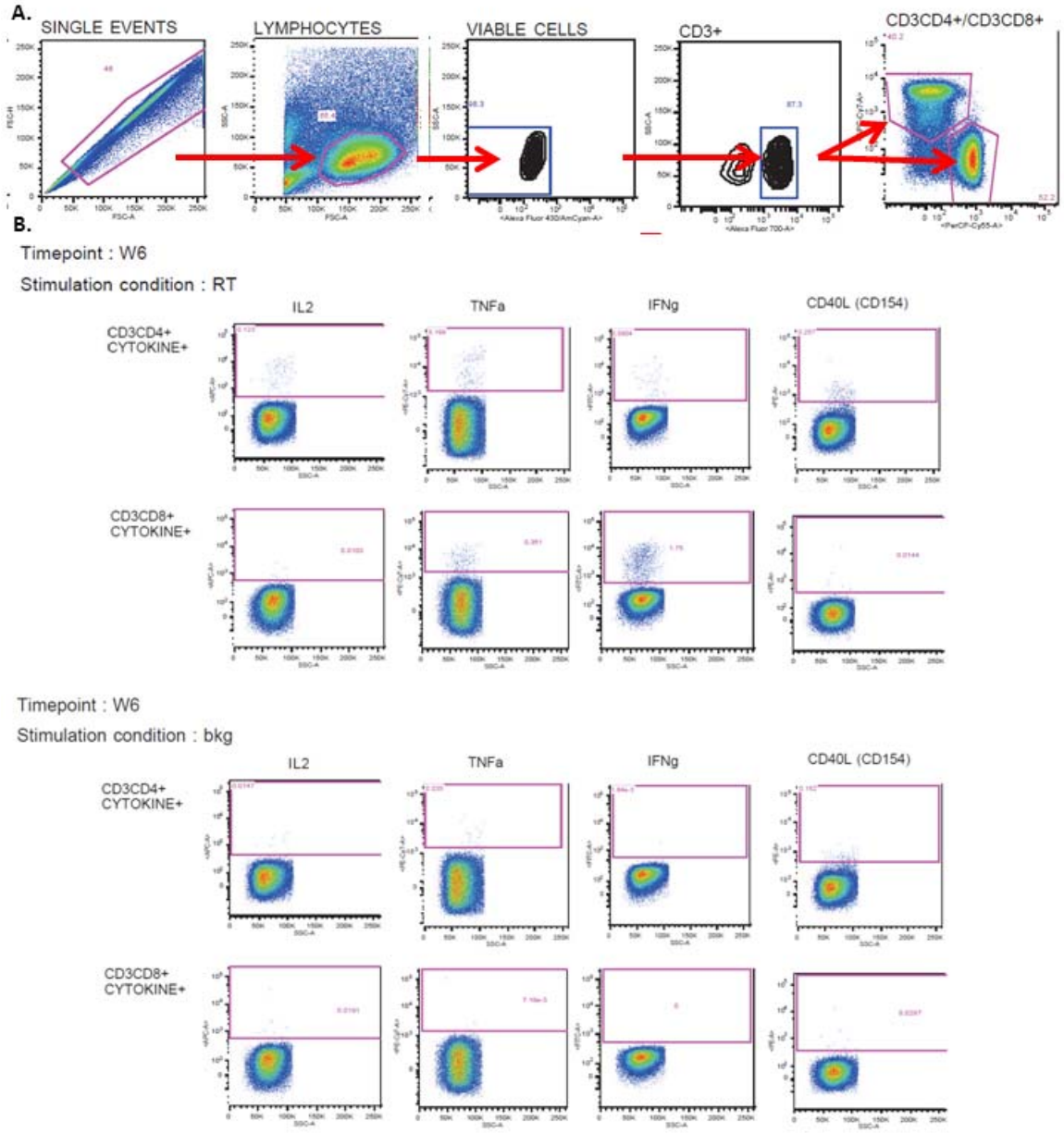
Supplement 1: The progressive gating scheme to enumerate antigen specific T cells. A. Gating strategy to identify, in succession, single cells, live lymphocytes, T-cells, and CD4 or CD8 T-cell lineages. B. Illustration of PBMC from a representative vaccinated participant following RT peptide stimulation or mock stimulation (bkg) at week 6 after vaccination. The fraction of CD40-L, TNF-a, IL-2, or IFN-γ expressing cells was defined based on individual gates; Boolean gate combinations were used to enumerate combinations of expression of cytokines.

Footnote: IL-2 = interleukin-2; TNF-α= tumor necrosis factor-α; IFN-γ= interferon-γ; CD40L = CD40-ligand



Supplement 2. $CD4^+$ T-cells count using crude values and HIV-1 viral load using log_{10}-transformed values at screening (modified total vaccinated cohort)
Footnote: $F4/AS01_B$ = participants randomized to receive three doses of $F4/AS01_B$ at Weeks 0, 4 and 28 or two doses of $F4/AS01_B$ at Weeks 0 and 4, and one dose of placebo at Week 28; Control = participants randomized to receive three doses of placebo at Weeks 0, 4 and 28 ; N = total number of participants; SD = Standard deviation.

Characteristic		**$F4/AS01_B$ (N=125)**	**Control (N=60)**
$CD4^+$ T-cells count at screening	Median (Range)	646 (502, 1801)	669.5 (501, 1067)
Viral load at screening (log_{10} copies/mL)	Mean (SD)	4.13 (0.42)	4.11 (0.40)

Supplement 3. Serious adverse events reported during the entire study period (total vaccinated cohort)

Footnote: F4/AS01$_B$_3 = participants randomized to receive three doses of F4/AS01$_B$ at Weeks 0, 4 and 28; F4/AS01$_B$_2 = participants randomized to receive two doses of F4/AS01$_B$ at Weeks 0 and 4, and one dose of placebo at Week 28;
Control = participants randomized to receive three doses of placebo at Weeks 0, 4 and 28. * recovering at time of study end.

Group	Serious adverse event	Dose	Day of onset	Causality	Outcome
F4/AS01$_B$_3	Gastroenteritis due to shigella	1	1	No	Recovered
	Appendicitis	3	56	No	Recovered
F4/AS01$_B$_2	Lymph node tuberculosis	2	95	No	Recovered
	Anxiety	2	128	No	Recovered
F4/AS01$_B$_2	Angioedema	2	0	Yes	Recovered
F4/AS01$_B$_2	Hepatitis C	3	0	No	Recovering
Control	Skull fracture	1	32	No	Recovered
Control	Pre-eclampsia	2	238	No	Recovered
	Anemia	2	170	No	Recovered
	Hemorrhage intracranial	2	241	No	Recovered
	Hydrocephalus	2	241	No	Recovered
	Hydronephrosis	2	241	No	Recovered

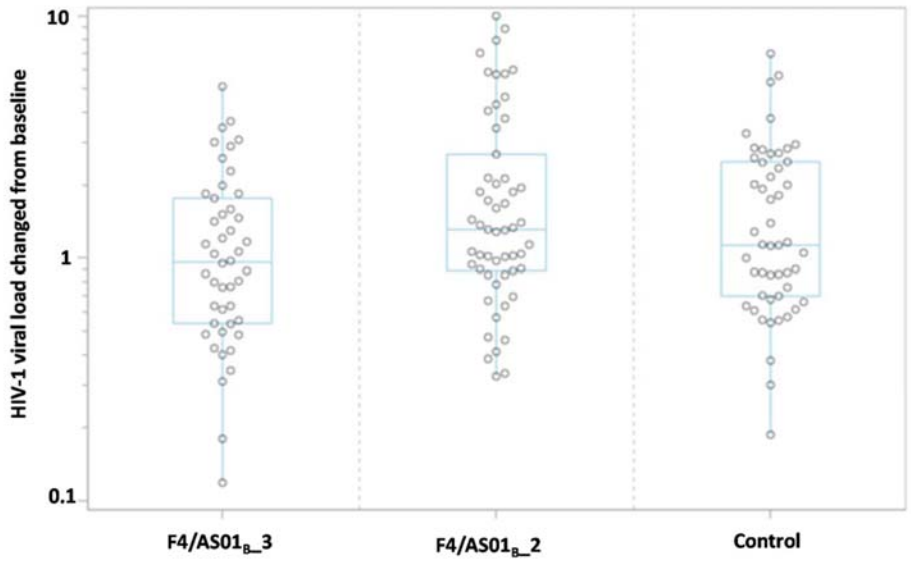
Supplement 4. Ophthalmologic results at baseline and at study conclusion (total vaccinated cohort)

Footnote: F4/AS01$_B$_3 = participants randomized to receive three doses of F4/AS01$_B$ at Weeks 0, 4 and 28; F4/AS01$_B$_2 = participants randomized to receive two doses of F4/AS01$_B$ at Weeks 0 and 4, and one dose of placebo at Week 28;
Control = participants randomized to receive three doses of placebo at Weeks 0, 4 and 28; N = total number of participants in the group; n (%) = number (percentage) of participants in a given category; Normal = category < 1.0 for both eyes and all 3 eye exams (slit lamp cortical, slit lamp nuclear and slit lamp subcapsular);
Abnormal = category ≥ 1.0 for at least one eye and one exam among slit lamp cortical, slit lamp nuclear and slit lamp subcapsular.

		F4/AS01$_B$_3 N = 62	**F4/AS01$_B$_2 N = 64**	**Control N = 64**
Category at baseline	**Categories at study end**	**n (%)**	**n (%)**	**n (%)**
Abnormal	Abnormal	2 (66.7)	1 (20.0)	2 (50.0)
	Normal	1 (33.3)	4 (80.0)	2 (50.0)
	Unknown	0 (0.0)	0 (0.0)	0 (0.0)
	Missing	1 (-)	0 (-)	0 (-)
Normal	Abnormal	1 (2.0)	2 (4.0)	0 (0.0)
	Normal	50 (98.0)	47 (94.0)	48 (98.0)
	Unknown	0 (0.0)	1 (2.0)	1 (2.0)
	Missing	6 (-)	8 (-)	10 (-)
Unknown	Abnormal	0 (0.0)	0 (0.0)	0 (0.0)
	Normal	0 (0.0)	0 (0.0)	0 (0.0)
	Unknown	1 (100)	1 (100)	1 (100)

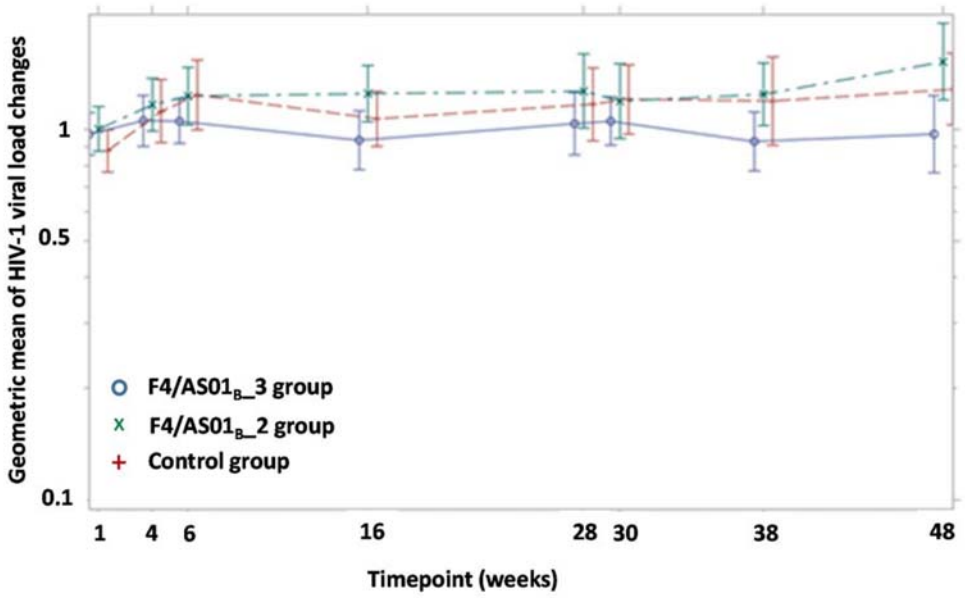
Supplement 5. Change in HIV-1 viral load from baseline to Week 48 (ratio of each crude value over baseline crude value) in ART-naive HIV-infected participants (modified total vaccinated cohort).

Footnote: F4/AS01$_B$_3 = participants randomized to receive three doses of F4/AS01$_B$ at Weeks 0, 4 and 28; F4/AS01$_B$_2 = participants randomized to receive two doses of F4/AS01$_B$ at Weeks 0 and 4, and one dose of placebo at Week 28;
Control = participants randomized to receive three doses of placebo at Weeks 0, 4 and 28. The boxplot: the central box shows the interquartile range (Q1–Q3), with the thick horizontal line representing the median (Q2), the whiskers (above and below the box), the maximum and the minimum, and the circles, the observed individual data.



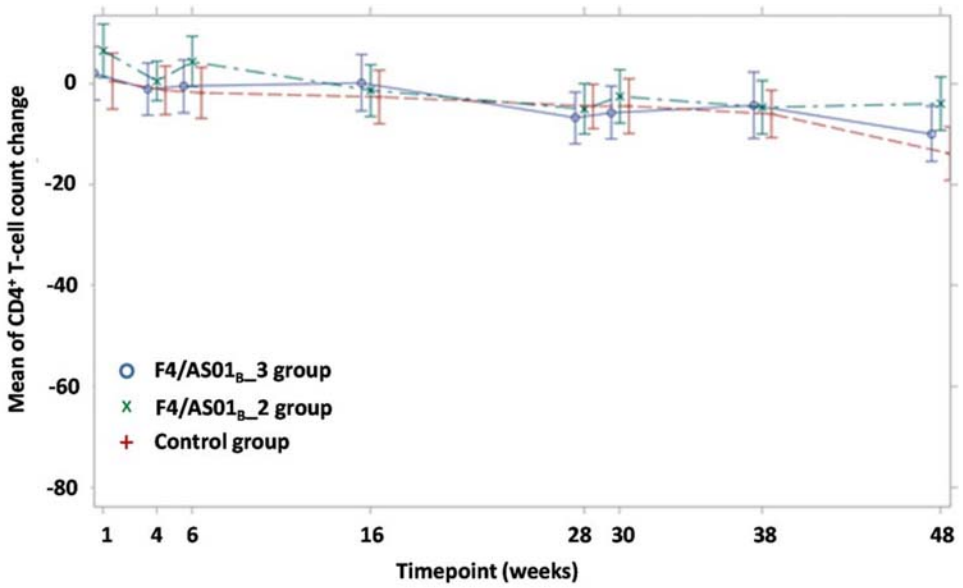
Supplement 6. Kinetics of the geometric mean of changes in HIV-1 viral load from baseline (modified total vaccinated cohort)

Footnote: F4/AS01$_B$_3 = participants randomized to receive three doses of F4/AS01$_B$ at Weeks 0, 4 and 28; F4/AS01$_B$_2 = participants randomized to receive two doses of F4/AS01$_B$ at Weeks 0 and 4, and one dose of placebo at Week 28;
Control = participants randomized to receive three doses of placebo at Weeks 0, 4 and 28. Errors bars represent 95% confidence intervals.

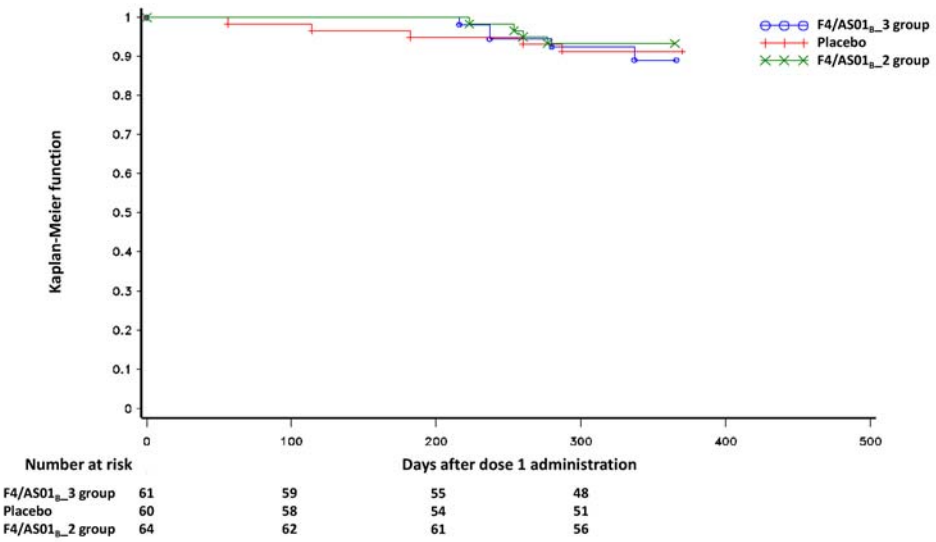


Supplement 7. Kinetics of the mean of changes in $CD4^+$ T-cells count from baseline (modified total vaccinated cohort)

Footnote: $F4/AS01_B$_3 = participants randomized to receive three doses of $F4/AS01_B$ at Weeks 0, 4 and 28; $F4/AS01_B$_2 = participants randomized to receive two doses of $F4/AS01_B$ at Weeks 0 and 4, and one dose of placebo at Week 28;
Control = participants randomized to receive three doses of placebo at Weeks 0, 4 and 28. Errors bars represent 95% confidence intervals.

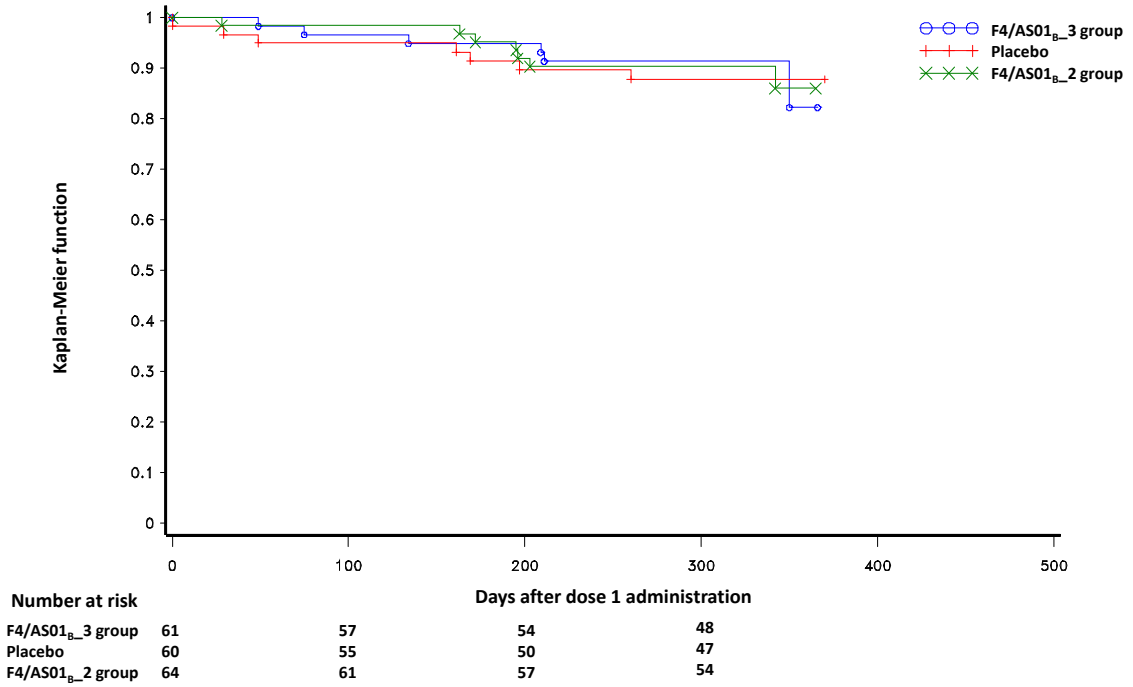


Supplement 8. Reverse cumulative curves Kaplan-Meyer graphical presentation of time to ART initiation after the first vaccination (modified total vaccinated cohort)
Footnote: F4/AS01$_B$_3 = participants randomized to receive three doses of F4/AS01$_B$ at Weeks 0, 4 and 28; F4/AS01$_B$_2 = participants randomized to receive two doses of F4/AS01$_B$ at Weeks 0 and 4, and one dose of placebo at Week 28;
Placebo = participants randomized in Control group to receive three doses of placebo at Weeks 0, 4 and 28. ART = anti-retroviral therapy



Supplement 9. Reverse cumulative curves Kaplan-Meyer graphical presentation of occurrence of any HIV-related clinical event after the first vaccination (modified total vaccinated cohort)

Footnote: F4/AS01$_B$_3 = participants randomized to receive three doses of F4/AS01$_B$ at Weeks 0, 4 and 28; F4/AS01$_B$_2 = participants randomized to receive two doses of F4/AS01$_B$ at Weeks 0 and 4, and one dose of placebo at Week 28;
Placebo = participants randomized in control group to receive three doses of placebo at Weeks 0, 4 and 28. HIV related clinical events are (a) confirmed CD4$^+$ T-cell count < 350 cells/mm^3 (b) confirmed HIV viral load > 100 000 copies/mL (c) clinical disease progression.



Number at risk				
F4/AS01$_B$_3 group	61	57	54	48
Placebo	60	55	50	47
F4/AS01$_B$_2 group	64	61	57	54

Supplement 10. Percentage of responders in terms of F4-specific $CD40L^+CD4^+$ T-cell vaccine expressing at least IL-2 (alone or together with other cytokines)
Footnote: $F4/AS01_B$_3 = participants randomized to receive three doses of $F4/AS01_B$ at Weeks 0, 4 and 28; $F4/AS01_B$_2 = participants randomized to receive two doses of $F4/AS01_B$ at Weeks 0 and 4, and one dose of placebo at Week 28. Participants with undetectable cytokine secretion at pre-vaccination were defined as responders if they had ≥0.08% of CD40L + CD4 + T-cells expressing at least one cytokine. In participants with detectable cytokine secretion at pre-vaccination, response was defined as a > 2-fold increase from baseline in $CD40L^+CD4^+$ T-cells expressing at least IL-2 (interleukin-2).

Time point	$F4/AS01_B$_3	$F4/AS01_B$_2	Placebo
Week 6	78	75	-
Week 28	39	33	-
Week 30	76	37	-
Week 48	51	22	-
Pooled post-vaccination timepoints	-	-	30

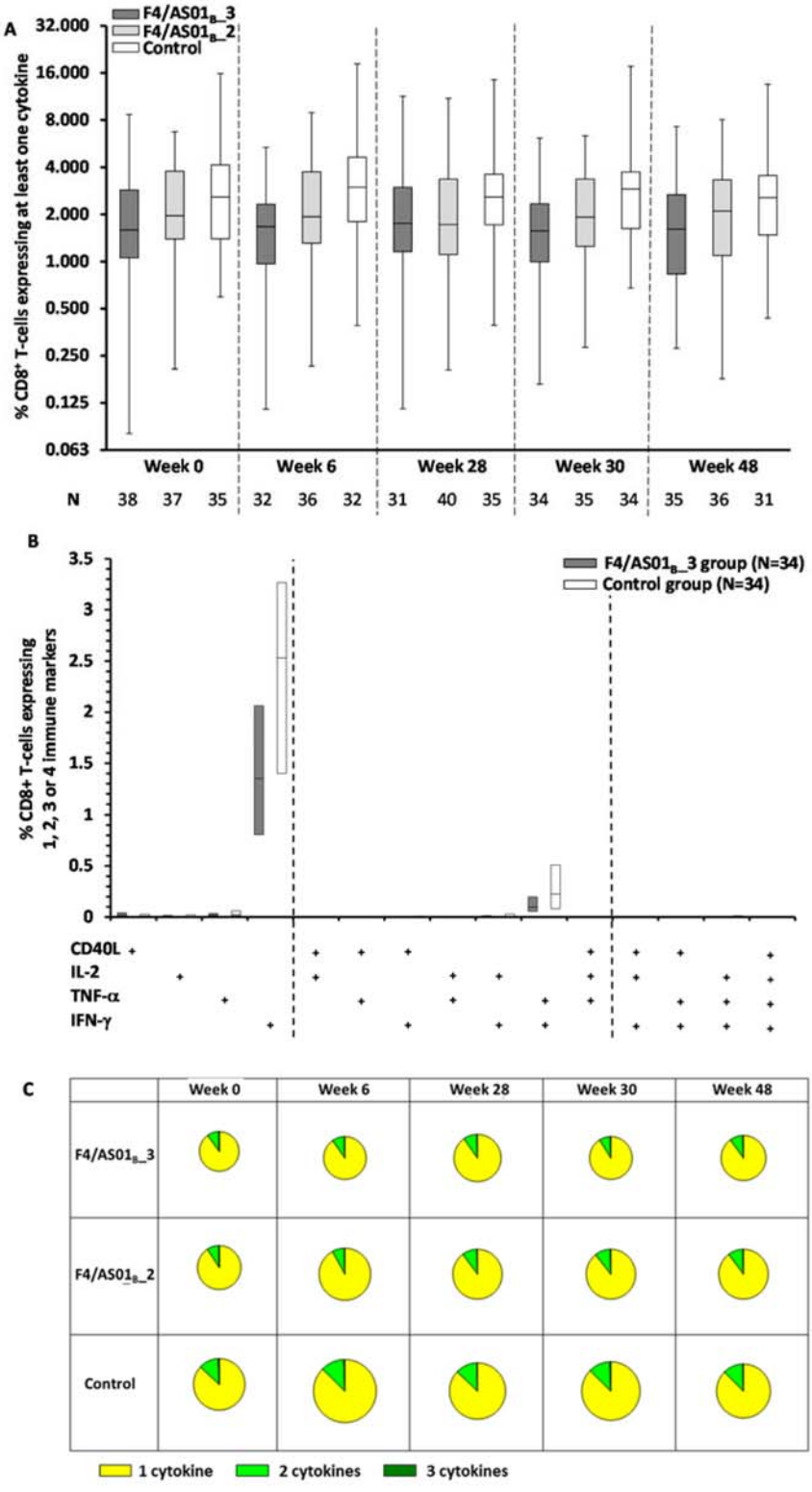
Supplement 11. Between group comparison for the number of each antigen-specific $CD40L^+CD4^+$ T-cell expressing at least IL-2 (alone or together with other cytokines): geometric mean ratio per million $CD4^+$ T-cells (according-to-protocol cohort for immunogenicity)

Footnote: $F4/AS01_B$_3 = participants randomized to receive three doses of $F4/AS01_B$ at Weeks 0, 4 and 28; $F4/AS01_B$_2 = participants randomized to receive two doses of $F4/AS01_B$ at Weeks 0 and 4, and one dose of placebo at Week 28;
Control = participants randomized to receive three doses of placebo at Weeks 0, 4 and 28; GMT = geometric mean titer; 95% CI = 95% confidence intervals.

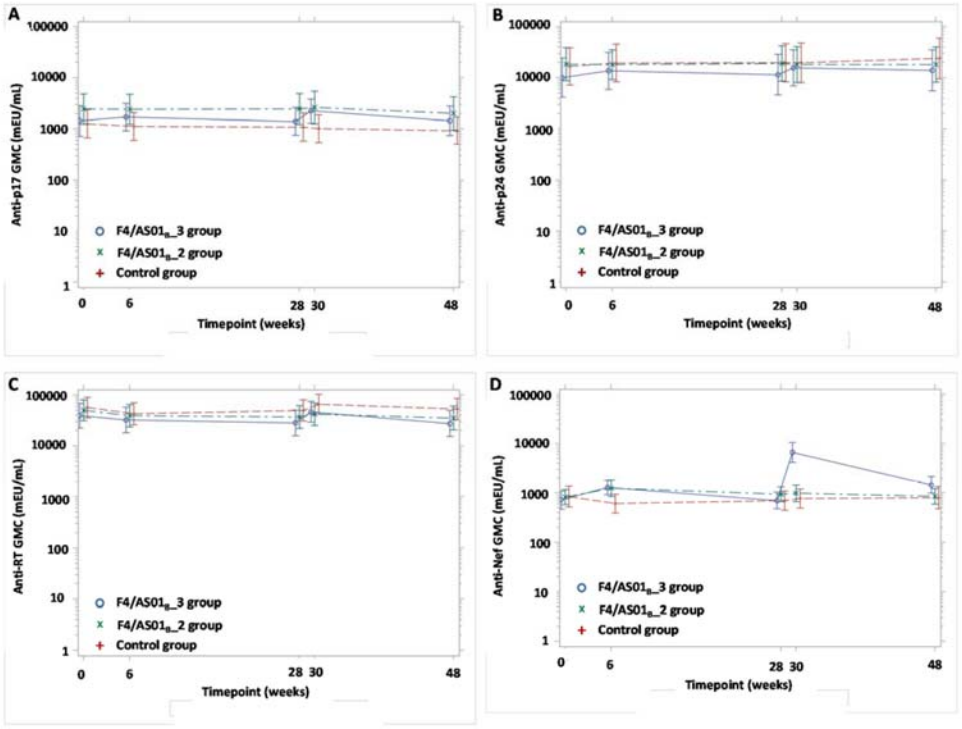
Timepoint	antigen	$F4/AS01_B$ group	GMT ratio ($F4/AS01_B$ over Control group) Value (95% CI)
Week 0	Nef	pooled $F4/AS01_B$_3 and $F4/AS01_B$_2	1.44 (0.85, 2.46)
	p17	pooled $F4/AS01_B$_3 and $F4/AS01_B$_2	0.86 (0.53, 1.39)
	p24	pooled $F4/AS01_B$_3 and $F4/AS01_B$_2	0.94 (0.63, 1.42)
	RT	pooled $F4/AS01_B$_3 and $F4/AS01_B$_2	1.66 (1.09, 2.55)
Week 6	Nef	pooled $F4/AS01_B$_3 and $F4/AS01_B$_2	9.15 (5.13, 16.30)
	p17	pooled $F4/AS01_B$_3 and $F4/AS01_B$_2	2.43 (1.53, 3.85)
	p24	pooled $F4/AS01_B$_3 and $F4/AS01_B$_2	1.71 (1.03, 2.83)
	RT	pooled $F4/AS01_B$_3 and $F4/AS01_B$_2	6.57 (3.98, 10.84)
Week 30	Nef	$F4/AS01_B$_3	23.46 (12.39, 44.44)
	p17	$F4/AS01_B$_3	2.40 (1.40, 4.11)
	p24	$F4/AS01_B$_3	2.41 (1.39, 4.16)
	RT	$F4/AS01_B$_3	6.33 (3.18, 12.60)
Week 48	Nef	$F4/AS01_B$_3	8.58 (3.98, 18.50)
	p17	$F4/AS01_B$_3	2.25 (1.19, 4.25)
	p24	$F4/AS01_B$_3	1.81 (1.00, 3.72)
	RT	$F4/AS01_B$_3	4.21 (2.11, 8.40)

Supplement 12. (A) Percentage of F4-specific $CD8^+$ T-cells expressing at least one cytokine, (B) Cytokine co-expression profile of F4-specific $CD8^+$ T-cells in the $F4/AS01_B$_3 group and the Control group at Week 30 and (C) pie charts of the cytokine co-expression of F4-specific $CD8^+$ T-cells at each timepoint in the three groups (according-to-protocol cohort for immunogenicity)

Footnote: $F4/AS01_B$_3 = participants randomized to receive three doses of $F4/AS01_B$ at Weeks 0, 4 and 28; $F4/AS01_B$_2 = participants randomized to receive two doses of $F4/AS01_B$ at Weeks 0 and 4, and one dose of placebo at Week 28; Control = participants randomized to receive three doses of placebo at Weeks 0, 4 and 28; IL-2 = interleukin-2; TNF-α = tumor necrosis factor-α; IFN-γ = interferon-γ; CD40L = CD40-ligand. The boxplot: the central box shows the interquartile range (Q1–Q3), with the thick horizontal line representing the median (Q2), and the whiskers (above and below the box), the maximum and the minimum. The percentage of $CD8^+$ T-cells expressing cytokines in response to the fusion protein F4 was determined by adding the individual frequencies of the $CD8^+$ T-cell response to each of the four individual antigens. Whiskers were not added to Supplement 10B for clarity. The sizes of the pie charts represent the proportions of total $CD8^+$ T-cells producing at least one cytokine.


A
F4/AS01B_3
F4/AS01B_2
Control
% CD8+ T-cells expressing at least one cytokine
32.000
16.000
8.000
4.000
2.000
1.000
0.500
0.250
0.125
0.063
Week 0
Week 6
Week 28
Week 30
Week 48
N
38 37 35 32 36 32 31 40 35 34 35 34 35 36 31
B
F4/AS01B_3 group (N=34)
Control group (N=34)
% CD8+ T-cells expressing
1, 2, 3 or 4 immune markers
3.5
3
2.5
2
1.5
1
0.5
0
CD40L
IL-2
TNF-α
IFN-γ
C
Week 0
Week 6
Week 28
Week 30
Week 48
F4/AS01B_3
F4/AS01B_2
Control
1 cytokine
2 cytokines
3 cytokines

Supplement 13. Antibody geometric mean concentrations (GMCs) against p17 (A), p24 (B), RT (C) and Nef (D) at each timepoint (ATP cohort for immunogenicity)
Footnote: F4/AS01$_B$_3 = participants randomized to receive three doses of F4/AS01$_B$ at Weeks 0, 4 and 28; F4/AS01$_B$_2 = participants randomized to receive two doses of F4/AS01$_B$ at Weeks 0 and 4, and one dose of placebo at Week 28;
Control = participants randomized to receive three doses of placebo at Weeks 0, 4 and 28. Errors bars represent 95% confidence intervals. ATP = According-to-Protocol. EU = ELISA Units



Supplement 14. Relationship between cell mediated immunogenicity (% $CD40L^+$ $CD4^+$ T-cells expressing at least IL-2) and change from baseline in (A) VL or (B) CD4 count at Week 48 (ATP cohort for immunogenicity)

Footnote: $F4/AS01_B$_3 = participants randomized to receive three doses of $F4/AS01_B$ at Weeks 0, 4 and 28; $F4/AS01_B$_2 = participants randomized to receive two doses of $F4/AS01_B$ at Weeks 0 and 4, and one dose of placebo at Week 28;
Control = participants randomized to receive three doses of placebo at Weeks 0, 4 and 28. ATP = According-to-Protocol.