

1 **Supplemental Material**

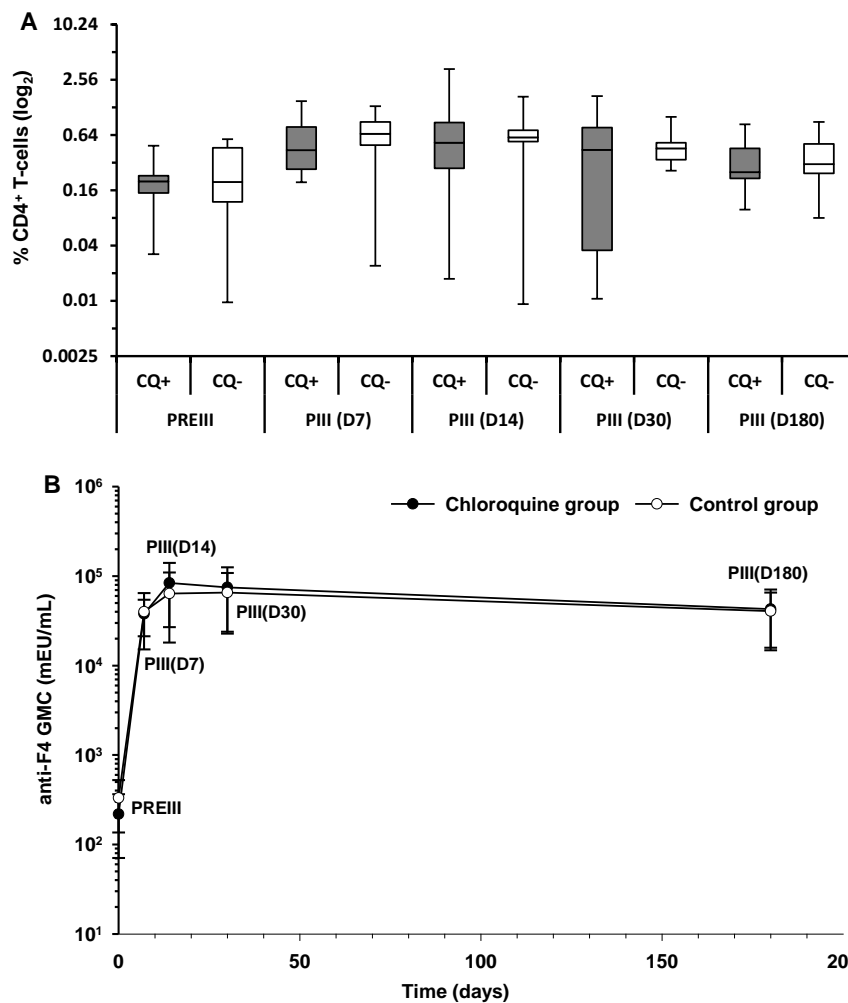
2 **Immunogenicity and safety of a booster dose of an investigational adjuvanted**

3 **polyprotein HIV-1 vaccine in healthy adults and effect of administration of chloroquine.**

4 Geert Leroux-Roels, Patricia Bourguignon, Julie Willekens, Michel Janssens, Frédéric

5 Clement, Arnaud Didierlaurent, Laurence Fissette, François Roman, Dominique Boutriau

6 **Supplement 1.** (A) Frequency of CD4⁺ T-cells expressing IL-2 and at least one other
 7 cytokine in response to the F4 fusion protein and (B) geometric mean concentrations for
 8 anti-F4 antibodies after administration of the booster doses of the F4/AS01_B-adjuvanted
 9 HIV Type 1 investigational vaccine in the Chloroquine and the Control groups (ATP
 10 immunogenicity cohort)

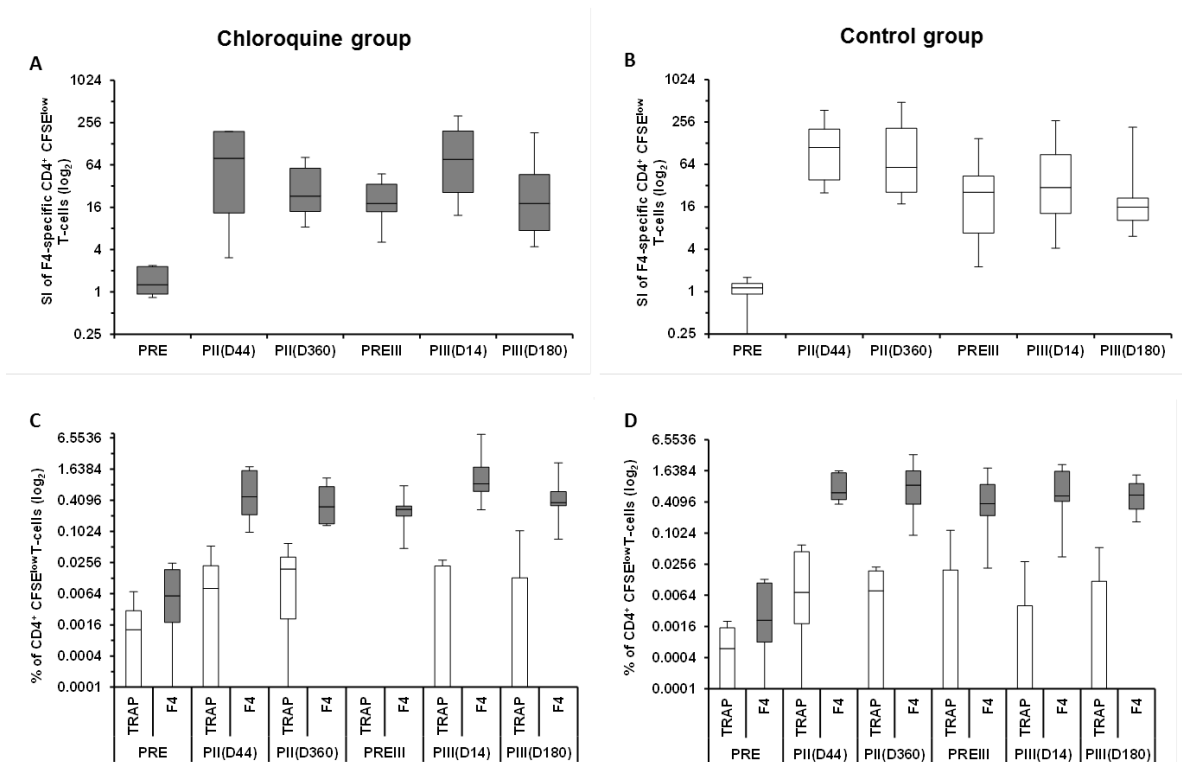


11

12 Footnote: CQ=chloroquine. The percentage of CD4⁺/CD8⁺ T-cells expressing cytokines in response
 13 to the fusion protein F4 was determined by adding the individual frequencies of the CD4⁺/CD8⁺ T-
 14 cell response to each of the four individual antigens. PREIII = pre-booster dose= approximately
 15 three years post-primary vaccination; PIII(D7), PIII(D14), PIII(D30), PIII(D180)= 7, 14, 30, and 180
 16 days after the booster dose. The boxplot: the central box shows the interquartile range (Q1–Q3),

17 with the thick horizontal line representing the median (Q2), and the whiskers (above and below the
18 box), the maximum and the minimum. GMC= geometric mean concentration; Error bars represent
19 95% confidence intervals.

20 **Supplement 2.** (A and B) Stimulation index of CD4⁺ CFSE^{low} T-cells stimulated with F4
 21 peptide pools and (C and D) frequency of F4- and TRAP-specific CD4⁺ CFSE^{low} T-cells
 22 expressing at least one cytokine following primary and booster doses of the F4/AS01_B
 23 investigational vaccine in the Chloroquine (A and C) and the Control (B and D) groups (ATP
 24 immunogenicity cohort)



25
 26 Footnote: CFSE= 5,6-CarboxyFluorescein diacetate Succinimidyl Ester; F4= pool of peptides covering
 27 the sequence of F4 antigens; PRE= pre-vaccination in the initial study; PII(D44)= 44 days after
 28 primary vaccination; PII(D360)= 360 days after primary vaccination; PREIII = pre-booster dose=
 29 approximately three years post-primary vaccination; PIII(D14)= 14 days after the booster dose;
 30 PIII(D180)= 180 days after the booster dose; SI= stimulation index; TRAP= tartrate-resistant acid
 31 phosphatase (irrelevant peptides). The boxplot: the central box shows the interquartile range (Q1–
 32 Q3), with the thick horizontal line representing the median (Q2), and the whiskers (above and
 33 below the box), the maximum and the minimum.

34 **Supplement 3.** CD4⁺ T-cell response to the F4/AS01-adjuvanted HIV type 1 vaccine after stimulation with p17, p24, RT, and Nef peptide
 35 pools: responder rates (pooled ATP immunogenicity cohort for 10µg primed participants)

Timepoint	N	Responders by number of antigens, % (95% CI)				Responders by antigen, % (95% CI)			
		≥1 antigen	≥2 antigens	≥3 antigens	All 4 antigens	Nef	P17	P24	RT
Day 0	10	60.0 (26.2–87.8)	60.0 (26.2–87.8)	30.0 (6.7–65.2)	20.0 (2.5–55.6)	30.0 (6.7–65.2)	20.0 (2.5–55.6)	60.0 (26.2–87.8)	60.0 (26.2–87.8)
Day 7	9	100 (66.4–100)	88.9 (51.8–99.7)	88.9 (51.8–99.7)	77.8 (40.0–97.2)	88.9 (51.8–99.7)	77.8 (40.0–97.2)	88.9 (51.8–99.7)	100 (66.4–100)
Day 14	9	100 (66.4–100)	88.9 (51.8–99.7)	88.9 (51.8–99.7)	66.7 (29.9–92.5)	66.7 (29.9–92.5)	88.9 (51.8–99.7)	88.9 (51.8–99.7)	100 (66.4–100)
Day 30	9	88.9 (51.8–99.7)	88.9 (51.8–99.7)	77.8 (40.0–97.2)	66.7 (29.9–92.5)	77.8 (40.0–97.2)	66.7 (29.9–92.5)	88.9 (51.8–99.7)	88.9 (51.8–99.7)
Day 180	10	100 (69.2–100)	80.0 (44.4–97.5)	70 (34.8–93.3)	40.0 (12.2–73.8)	50.0 (18.7–81.3)	60.0 (26.2–87.8)	80.0 (44.4–97.5)	100 (69.2–100)

36

37 Footnote: CI= confidence interval; N=number of participants with results available for the 4 antigens

38

39