

ORIGINAL ARTICLE

A Monovalent Chimpanzee Adenovirus Ebola Vaccine — Preliminary Report

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ABSTRACT

BACKGROUND

The West African outbreak of Ebola virus disease has caused more than 8500 deaths. A vaccine could contribute to outbreak control in the region. We assessed a monovalent formulation of a chimpanzee adenovirus 3 (ChAd3)–vectored vaccine encoding the surface glycoprotein of *Zaire ebolavirus* (EBOV), matched to the outbreak strain.

METHODS

After expedited regulatory and ethics approvals, 60 healthy adult volunteers in Oxford, United Kingdom, received a single dose of the ChAd3 vaccine at one of three dose levels: 1×10^{10} viral particles, 2.5×10^{10} viral particles, and 5×10^{10} viral particles (with 20 participants per group). Safety was assessed over the next 4 weeks. Antibodies were measured on enzyme-linked immunosorbent assay (ELISA) and T-cell responses on enzyme-linked immunospot (ELISpot) and flow-cytometry assays.

RESULTS

No safety concerns were identified at any of the dose levels studied. Fever developed in 2 of the 59 participants who were evaluated. Prolonged activated partial-thromboplastin times and transient hyperbilirubinemia were observed in 4 and 8 participants, respectively. Geometric mean antibody responses on ELISA were highest (469 units; range, 58 to 4051; 68% response rate) at 4 weeks in the high-dose group, which had a 100% response rate for T cells on ELISpot, peaking at day 14 (median, 693 spot-forming cells per million peripheral-blood mononuclear cells). Flow cytometry revealed more CD4+ than CD8+ T-cell responses. At the vaccine doses tested, both antibody and T-cell responses were detected but at levels lower than those induced in macaques protected by the same vaccine.

CONCLUSIONS

The ChAd3 monovalent vaccine against EBOV was immunogenic at the doses tested. (Funded by the Wellcome Trust and others; ClinicalTrials.gov number, NCT02240875.)

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THE CURRENT OUTBREAK OF EBOLA VIRUS disease (EVD) in West Africa has led to more than 8500 deaths.¹ An effective vaccine may be necessary to contain this international public health emergency. No new vaccine has been first tested in humans and rapidly developed within months in an attempt to control a major infectious disease outbreak. However, both the chimpanzee adenovirus (ChAd) and modified vaccinia virus Ankara (MVA) vectors,² which were developed by the Vaccine Research Center of the National Institute of Allergy and Infectious Diseases in collaboration with Okavios, had already been manufactured to clinical grade at the time of the acceleration of the EVD outbreak in early August 2014. These events provided the opportunity to design a rapid clinical-development program that could lead to deployment of the vaccine in 2015.

ChAd vectors make up a new-generation vaccine technology that first reached clinical assessment in 2007³⁻⁵ and are generally used clinically with an MVA booster dose.⁶⁻¹⁴ A single dose of 10¹⁰ or 10¹¹ virus particles of chimpanzee adenovirus type 3 (ChAd3, also called cAd3) encoding the *Zaire ebolavirus* (EBOV) wild-type surface glycoprotein had shown efficacy in cynomolgus macaques, which encouraged the assessment of a single-dose vaccine in this phase 1 trial, called EBL01.² The Guinea outbreak strain of ebolavirus is 97% identical in amino acid sequence to the well-characterized Zaire strain.¹⁵ Although the original clinical-development plan for this Ebola vaccine included the use of a bivalent vaccine formulation of Zaire and Sudan strains¹⁶ that would use both ChAd3 and MVA primarily for biodefense, the ChAd3 vaccine encoding just the Zaire strain appeared to be a potentially advantageous monovalent formulation for outbreak control on the basis of efficacy data in macaques and was thus selected for testing in this study.

Biomanufacturing of large amounts of the ChAd3 vaccine was a limiting factor in the development of an accelerated plan to undertake large-scale efficacy trials and deployment. The use of the monovalent formulation halves the manufacturing challenge, as compared with use of the bivalent vaccine, which includes a second vector encoding the Sudan strain glycoprotein¹⁶ with only 60% identity to the Guinea outbreak strain. Moreover, previous preclinical and clinical

assessments of viral vectors encoding multiple antigens or mixtures of vectors encoding different antigens have sometimes shown reduced immunogenicity when more than a single antigen vector was used.^{17,18} A further consideration in the trial design was the possibility that a lower dose of ChAd might be sufficiently immunogenic with this insert, as suggested by previous studies of clinical-dose response with ChAd vectors,³ a factor that could thus allow the deployment of more vaccine doses from each manufacturing run.

With input from the World Health Organization (WHO),¹⁹ we therefore developed a plan for rapid clinical assessment of the safety and immunogenicity of a monovalent formulation of the ChAd3 vaccine against EBOV in August 2014 at three clinical sites (Oxford, United Kingdom; Lausanne, Switzerland; and Bamako, Mali), with an aim to immunize and evaluate 240 participants by late November. We report here on the safety and immunogenicity of this monovalent vaccine — now prioritized for use in a phase 3 trial and, potentially, for outbreak control — in its first trial in Oxford, United Kingdom.

METHODS

STUDY PARTICIPANTS

The study was conducted at the Centre for Clinical Vaccinology and Tropical Medicine at the University of Oxford. Participants were healthy adults between the ages of 18 and 50 years who provided written informed consent (Table 1).

ETHICS AND REGULATORY APPROVAL

The study was reviewed and approved by the U.K. National Research Ethics Service, Committee South Central–Oxford A, the Medicines and Healthcare Products Regulatory Agency, and the Oxford University Clinical Trials and Research Governance team, who monitored compliance with Good Clinical Practice guidelines. Safety oversight was provided by an independent data and safety monitoring board. The ChAd3 vaccine was provided by the Vaccine Research Center of the National Institute of Allergy and Infectious Diseases and GlaxoSmithKline.

STUDY DESIGN

EBL01 was a phase 1, dose-escalation, open-label study assessing the safety and immunogenicity

of the experimental monovalent ChAd3 vaccine against EBOV. Eligible participants were assigned to receive the ChAd3 vaccine as a single intramuscular injection in one of three dose groups: group 1 (1×10^{10} viral particles), group 2 (2.5×10^{10} viral particles), and group 3 (5×10^{10} viral particles). The first nine vaccinations occurred in a stepwise dose-escalation manner, with three participants in the low-dose group being vaccinated and followed for a minimum of 48 hours before proceeding to immunize volunteers in the next dose group. Full details regarding the study conduct are provided in the protocol, which is available with the full text of this article at NEJM.org.

STUDY VACCINE

The ChAd3 drug substance was manufactured at Advent, a subsidiary of Okairos (now Glaxo-SmithKline), and the drug product was manufactured at the Vaccine Research Center Vaccine Pilot Plant, under contract with the Vaccine Clinical Materials Program, Leidos Biomedical Research. The vaccine is a sterile, aqueous, buffered solution that includes the ChAd3-vectored vaccine encoding the surface glycoprotein of EBOV in single-dose vials of 9.1×10^{10} particle units per milliliter, as determined on high-performance liquid chromatography. Different dose levels were achieved by adjusting the volume of vaccine injected from 110 μl (in group 1) to 275 μl (in group 2) and 550 μl in group 3.

ASSESSMENT OF SAFETY

Participants were observed for 60 minutes after vaccination. Follow-up visits were scheduled for days 1, 7, 14, 28, 90, and 180 after vaccination, with an additional visit at day 10 for participants in group 3. All participants were given access to an electronic diary card on which to record all solicited symptoms for 7 days after vaccination and unsolicited symptoms for 28 days after vaccination. A review of symptoms occurred at each follow-up visit, in addition to testing that included a full blood count and the measurement of urea and electrolytes, liver enzymes, activated partial-thromboplastin time, prothrombin time, and fibrinogen. Severity grading of adverse events and the assignment of a causal relationship for unsolicited adverse events were conducted according to predefined criteria.

Table 1. Characteristics of the Participants at Baseline.*

Characteristic	Group 1 (N=20)	Group 2 (N=20)	Group 3 (N=20)	All Participants (N=60)
Sex — no. (%)				
Male	6 (30)	13 (65)	13 (65)	32 (53)
Female	14 (70)	7 (35)	7 (35)	28 (47)
Age				
Group — no. (%)				
18–20 yr	0	0	1 (5)	1 (2)
21–30 yr	9 (45)	8 (40)	11 (55)	28 (47)
31–40 yr	6 (30)	8 (40)	3 (15)	17 (28)
41–50 yr	5 (25)	4 (20)	5 (25)	14 (23)
Mean — yr	31.7 \pm 9.0	33.5 \pm 8.0	31.3 \pm 9.3	32.2 \pm 8.7
Range — yr	21–48	22–49	18–48	18–49
Race — no. (%) [†]				
White	19 (95)	19 (95)	18 (90)	55 (92)
Black	0	0	0	0
Asian	0	1 (5)	1 (5)	2 (3)
Mixed	1 (5)	0	1 (5)	3 (5)
Body-mass index [‡]				
Value — no. (%)				
<18.5	0	1 (5)	1 (5)	2 (3)
18.5–24.9	11 (55)	11 (55)	16 (80)	37 (62)
25–29.9	8 (40)	6 (30)	3 (15)	18 (30)
≥ 30	1 (5)	2 (10)	0	3 (5)
Mean	24.5 \pm 3.3	24.6 \pm 3.4	22.7 \pm 2.3	24.0 \pm 3.1
Range	18.5–30.5	17.4–33.0	17.1–27.4	17.1–33.0

* Plus-minus values are means \pm SD. There were no significant differences between the study groups.

[†] Race was self-reported.

[‡] The body-mass index is the weight in kilograms divided by the square of the height in meters.

ELISA ANALYSES

We assessed anti-glycoprotein IgG responses to EBOV using enzyme-linked immunosorbent assay (ELISA) by means of a commercial kit (AE 320620-1, Alpha Diagnostic International [ADI]), according to the manufacturer's instructions, with serum diluted at 1:100 and 1:500. Optical density was read at 450 nm on an EL800 microplate reader (BioTek). Values are reported with or without the subtraction of the prevaccination optical density and were converted to micrograms per milliliter with the use of the reference serum provided by the manufacturer. We also used end-

Table 2. Adverse Events.*

Symptom and Intensity	Group 1 (N=19)	Group 2 (N=20)	Group 3 (N=20)	All Participants (N=59)
Local				
Pain				
Mild	10 (53)	14 (70)	7 (35)	31 (53)
Moderate	2 (11)	2 (10)	3 (15)	7 (12)
Mild swelling	3 (16)	1 (5)	2 (10)	6 (10)
Mild redness	3 (16)	7 (35)	3 (15)	13 (22)
Mild warmth	10 (53)	7 (35)	4 (20)	21 (36)
Mild itch	2 (11)	4 (20)	0	6 (10)
Systemic				
Moderate fever				
	0	1 (5)	1 (5)	2 (3)
Feverishness				
Mild	3 (16)	5 (25)	6 (30)	14 (24)
Moderate	0	2 (10)	1 (5)	3 (5)
Severe	0	1 (5)	0	1 (2)
Myalgia				
Mild	7 (37)	6 (30)	11 (55)	24 (41)
Moderate	0	3 (15)	0	3 (5)
Arthralgia				
Mild	1 (5)	3 (15)	3 (15)	7 (12)
Moderate	0	1 (5)	1 (5)	2 (3)
Headache				
Mild	7 (37)	11 (55)	8 (40)	26 (44)
Moderate	2 (11)	3 (15)	1 (5)	6 (10)
Fatigue				
Mild	11 (58)	8 (40)	10 (50)	29 (49)
Moderate	2 (11)	2 (10)	3 (15)	7 (12)
Nausea				
Mild	3 (16)	1 (5)	4 (20)	8 (14)
Moderate	1 (5)	2 (10)	1 (5)	4 (7)
Severe	0	1 (5)	0	1 (2)
Malaise				
Mild	6 (32)	5 (25)	7 (35)	18 (31)
Moderate	2 (11)	1 (5)	2 (10)	5 (8)
Severe	0	1 (5)	0	1 (2)
Use of acetaminophen, NSAID, or aspirin for symptoms	8 (42)	8 (40)	7 (35)	23 (39)

* Shown are the maximum solicited local and systemic reactogenicity symptoms collected for 7 days after vaccination. Frequency is calculated as the number of participants counted once at the time of the worst severity of the event. Intensity categories in which all the values were zero are not shown. NSAID denotes nonsteroidal antiinflammatory drug. The case definitions for these adverse events can be found in the protocol.

point titration to perform assays to compare the responses directly with titers that were associated with protection in macaque efficacy trials² and with a method used in a trial of a bivalent ChAd3 vaccine in humans conducted by the National Institutes of Health (NIH).¹⁶ The end-point dilution ELISA for the NIH EC₉₀ (the concentration at which there is a 90% decrease in antigen binding) assay was performed as reported previously,² and values are also presented with or without the subtraction of the prevaccination optical density to maintain consistency with previous studies in which this assay was used.

T-CELL ASSAYS

Ex vivo enzyme-linked immunospot (ELISpot) and intracellular cytokine staining assays were performed largely as described previously^{7,11} with the use of overlapping peptide pools. (Additional details are provided in the Methods section in the Supplementary Appendix, available at NEJM.org.)

RESULTS

STUDY POPULATION

From September 17, 2014, to November 18, 2014, a total of 60 of the 88 participants who were screened for eligibility were vaccinated (Fig. S1 in the Supplementary Appendix). One participant in group 1 withdrew on day 1 after vaccination owing to an aversion to venipuncture. The participant had reported no symptoms at the time of withdrawal but declined to attend additional follow-up visits. E-mail correspondence on day 10 after vaccination confirmed that the participant remained well, with no symptoms to report. All the remaining 59 participants completed at least 28 days of follow-up.

SAFETY

A full listing of the frequency and maximum severity of solicited, unsolicited, and laboratory adverse events according to dose group are provided in Table 2, and in Tables S1 and S2 in the Supplementary Appendix. A majority of the adverse events that were reported in all dose groups were mild in severity, with no unexpected serious adverse reactions or serious adverse events (Table 2). Two participants (one in group 2 and one in group 3) had an episode of moderate fever (temperature, 38.1°C and 38.9°C, respectively). Both events oc-

curred within the first 24 hours after vaccination, and neither persisted for more than 24 hours.

A prolonged activated partial-thromboplastin time was observed in four participants during the first 2 weeks after vaccination: in three participants in group 2 (two with a grade 1 elevation and one with a grade 2 elevation) and in one participant in group 3 with a grade 1 elevation. None of the prolongations were associated with symptoms or clinical features of coagulopathy. The elevation had fully resolved in all participants by 10 weeks after vaccination. No further abnormality was found in any of these participants on extended hematologic and coagulation evaluation.

Transient induction of an antiphospholipid antibody causing an *in vitro* artifact on the laboratory assay for activated partial-thromboplastin time was reported previously after the administration of adenovirus vectors.^{20,21} Transient mild lymphocytopenia was noted on day 1 after vaccination in five participants in group 1, four in group 2, and eight in group 3, and moderate lymphocytopenia was noted in two participants each in group 2 and group 3 on day 1. Transient mild-to-moderate elevations in bilirubin were recorded in six participants in group 2 and group 3 combined. Transient hyperbilirubinemia in the severe range was recorded in two participants (one in group 2 and one in group 3) who had a prevaccination diagnosis of Gilbert's syndrome.

ANTIBODY RESPONSES

We measured optical density to assess IgG responses against the surface glycoprotein of EBOV with a single serum dilution before immunization and at days 14 and 28 (Fig. 1A and 1B). Antibody responses were highest at 28 days after vaccination, with no significant difference in response at any time point among the three dose groups. Vaccination induced a significant increase in the antibody titer in all groups ($P < 0.001$ by the Wilcoxon matched-pairs test) (Fig. 1, and Fig. S2A in the Supplementary Appendix).

We also used the EC₉₀ end-point titration method described in Methods to compare the responses directly with titers that were associated with protection in macaque efficacy trials² and in a trial of bivalent ChAd3 vaccine in humans conducted by the NIH¹⁶ (Fig. 1C and 1E, and Fig. S2B in the Supplementary Appendix). In the analyses of samples obtained from 58 par-

ticipants on day 28, the geometric mean titer after the subtraction of prevaccination responses was 235 units in group 1, 402 in group 2, and 469 in group 3 (Fig. 1E).

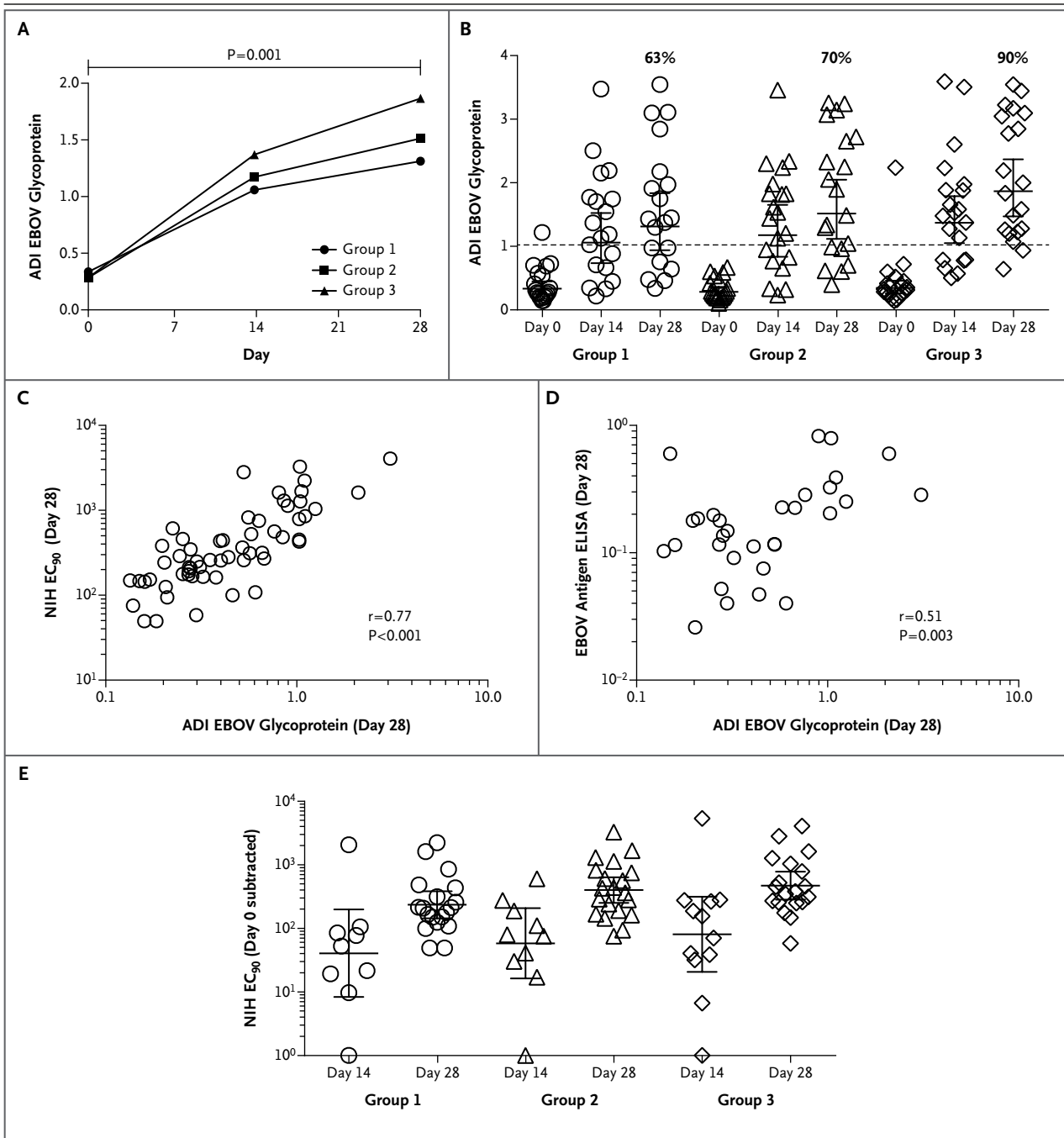
The results of the two above-mentioned assays, the ELISA with the ADI kit and the end-point dilution by means of the NIH method,^{2,16} were highly correlated with each other. We used linear regression to determine a conversion factor of 1 optical-density unit on the ADI scale to a dilution factor of 1202 on the NIH EC₉₀ assay (Fig. 1C), which yielded geometric mean values of 481 units in group 1, 514 in group 2, and 681 in group 3. We also used a heat-killed virus antigen ELISA to assess the induction of antibody titers against the Guinea outbreak strain of Ebola virus, using the Guinea strain whole virus rather than the surface glycoprotein from the Zaire strain as the antigen. In these analyses, 6 of 35 participants (17%) who were tested had a positive response (2 of 10 in group 1, 1 of 12 in group 2, and 3 of 13 in group 3) (Fig. 1D).

ELISPOT RESPONSES

T-cell responses to the 10 peptide pools were assessed by means of interferon- γ ELISpot assays on days 0, 7, 14, and 28 (Fig. 2A). Before vaccination, responses to the EBOV glycoprotein were below the level of detection of the assay in 72% of the participants (median, 50 spot-forming cells [SFCs] per million peripheral-blood mononuclear cells; 95% confidence interval, 53 to 63) (Fig. 2B). ELISpot responses peaked at day 14 (Fig. 2C), with median responses of 431 SFCs (interquartile range, 203 to 783) in group 1, 387 SFCs (interquartile range, 281 to 834) in group 2, and 693 SFCs (interquartile range, 348 to 866) in group 3. There was no significant difference in the magnitude of the immune response at any time point among the different dose groups on the basis of the Kruskal–Wallis test.

FLOW CYTOMETRY WITH INTRACELLULAR CYTOKINE STAINING

At 28 days after vaccination, we assessed the CD4+ and CD8+ T-cell responses to vaccination according to the secretion of interferon- γ , interleukin-2, or tumor necrosis factor α . Cytokines were predominantly detected from CD4+ T cells, and cytokine expression tended to be higher in group 3 than in group 1 ($P = 0.06$ by the Kruskal–Wallis test with Dunn's correction for multiple



comparisons) (Fig. 3A). Expression of the degranulation marker CD107a was also detected from CD8+ T cells (Fig. 3B). Both CD4+ and CD8+ T cells showed polyfunctional and monofunctional phenotypes, with dual positive cells predominating (Fig. 3C and 3D).

DISCUSSION

The safety and immunogenicity profile of this monovalent chimpanzee adenovirus–vectored vaccine supported its assessment either alone or as part of a heterologous prime-boost vaccination

Figure 1 (facing page). Antibody Responses to the *Zaire ebolavirus* Glycoprotein.

Panel A shows anti-glycoprotein IgG levels, as measured with the use of an enzyme-linked immunosorbent assay (ELISA) (Alpha Diagnostic International [ADI]), at baseline and on days 14 and 28 after vaccination with increasing doses of a chimpanzee adenovirus 3 (ChAd3)-vectored vaccine against *Zaire ebolavirus* (EBOV). The antibody levels in the three dose groups are shown in optical-density (OD) units without subtraction of background levels on the y axis. The analyses are for 19 patients in group 1, 20 in group 2, and 19 in group 3. No significant differences in responses among doses at any time point were detected. Lines represent geometric mean IgG responses for each group. Panel B shows individual responses at days 0, 14, and 28 for each group in geometric means and 95% confidence intervals. The percentage of participants with positive results at the peak time points on day 28 are indicated. The dotted line represents the positive threshold (optical density, 1.024), as calculated from the mean +3 SD of the day 0 responses for all participants. Panel C shows Spearman's correlation for the results on the ADI ELISA and the NIH EC₉₀ end-point dilution ELISA for 58 samples tested with the two assays for IgG antibodies against EBOV. Linear regression of these data indicates that 1 OD unit with the ADI assay is equivalent to 1202 units on the NIH EC₉₀ scale for serum diluted at 1:500. On the basis of this conversion, the geometric mean responses were 481 units for group 1, 514 for group 2, and 681 for group 3 on the NIH EC₉₀ scale, representing reciprocal serum dilutions. Panel D shows Spearman's correlation for the ADI ELISA and the EBOV antigen ELISA using heat-killed Guinea strain virus in samples obtained from 35 participants. Panel E shows titers for 58 participants with the use of the NIH EC₉₀ ELISA reported previously⁶ (with 19 participants in group 1, 20 in group 2, and 19 in group 3). The geometric mean titers in these groups were 235, 402, and 469, respectively, after the subtraction of prevaccination responses at baseline.

regimen for the prevention of infection and disease by the Guinea outbreak strain of EBOV. A considerable effort by many groups and agencies allowed for a successful application for funding, the filling of the monovalent vaccine, submission and approval of regulatory and ethical applications, completion of contractual arrangements, and initiation of this clinical trial in approximately 1 month. The prioritization of this trial by several funders, regulators, and reviewers in response to the declaration of an international public health emergency was key to rapid progress.

No safety concerns were identified for this

ChAd vector, with the majority of the recorded local and systemic adverse events being mild and short-lived. These findings are similar to the safety profile of other simian adenoviral vectors that have been assessed clinically.^{3-7,11-14,18} As of mid-December 2014, approximately 250 participants had received this monovalent vaccine, with no reports of serious vaccine-related adverse events.^{22,23}

Accrual of safety data from this trial allowed for initiation of immunization with the same monovalent vaccine in Bamako, in early October 2014,²³ and to date 91 participants have been immunized at that center.²² In previous trials of some viral-vectored vaccines, reduced immunogenicity has been observed among African populations, as compared with the responses among northern Europeans,^{11,24} which highlights the importance of immunogenicity assessments in West Africa for a vaccine that is intended for use in the control of the current Ebola outbreak.

In our study, immunogenicity assessments included measurements of glycoprotein-specific antibodies against EBOV by means of an ELISA analysis for which the manufacturer provided a conversion factor and control serum, which allowed for an estimated readout in micrograms per milliliter, as well as both interferon- γ ELISpot and flow-cytometry assays performed on fresh cells. These *ex vivo* T-cell assays provide a sensitive measure of T-cell immunogenicity that is not subject to diminution by cryopreservation of peripheral-blood mononuclear cells. Antibody responses, as measured on ELISA, showed a weak dose-response relationship, with a maximal geometric mean response at day 28 after immunization for the dose groups tested in this study. The range of ELISA values that were measured by means of end-point titration in the high-dose group (group 3) was 58 to 4051, with a geometric mean titer of 469, which was generally lower than the titers of 967 to 6600 that were reported in macaques protected by 2×10^{10} viral particles of the bivalent ChAd3 vector,² although some overlap is evident.

T-cell responses that were measured on ELISpot assays were as expected for this viral vector, with responses peaking at day 14 at about 700 SFCs in the high-dose group.⁶ Flow cytometry on day 28 showed a predominantly

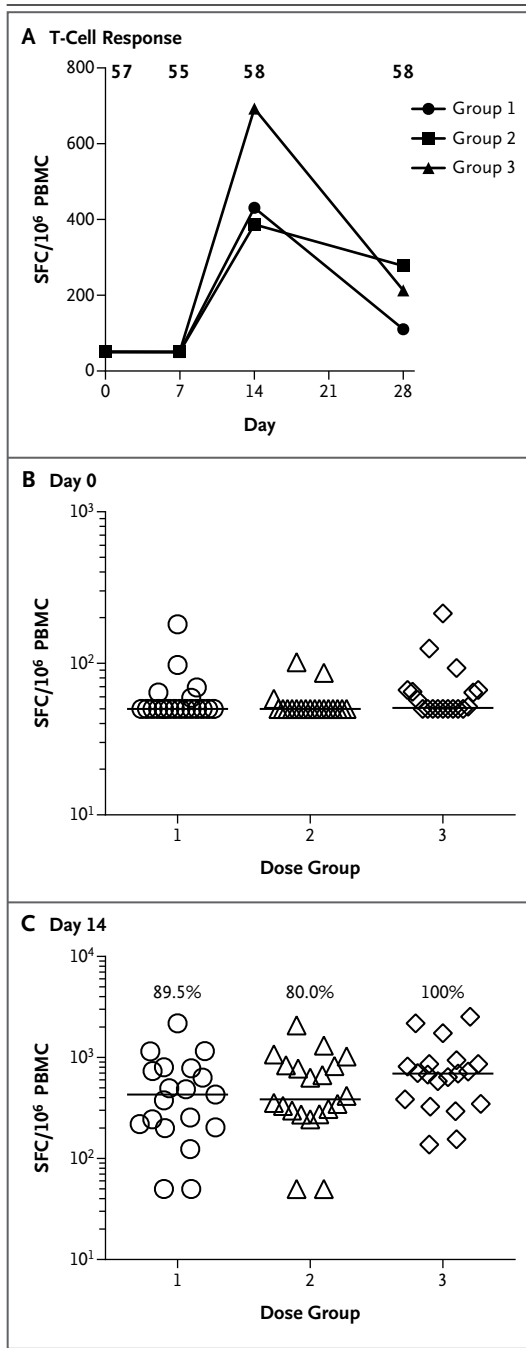


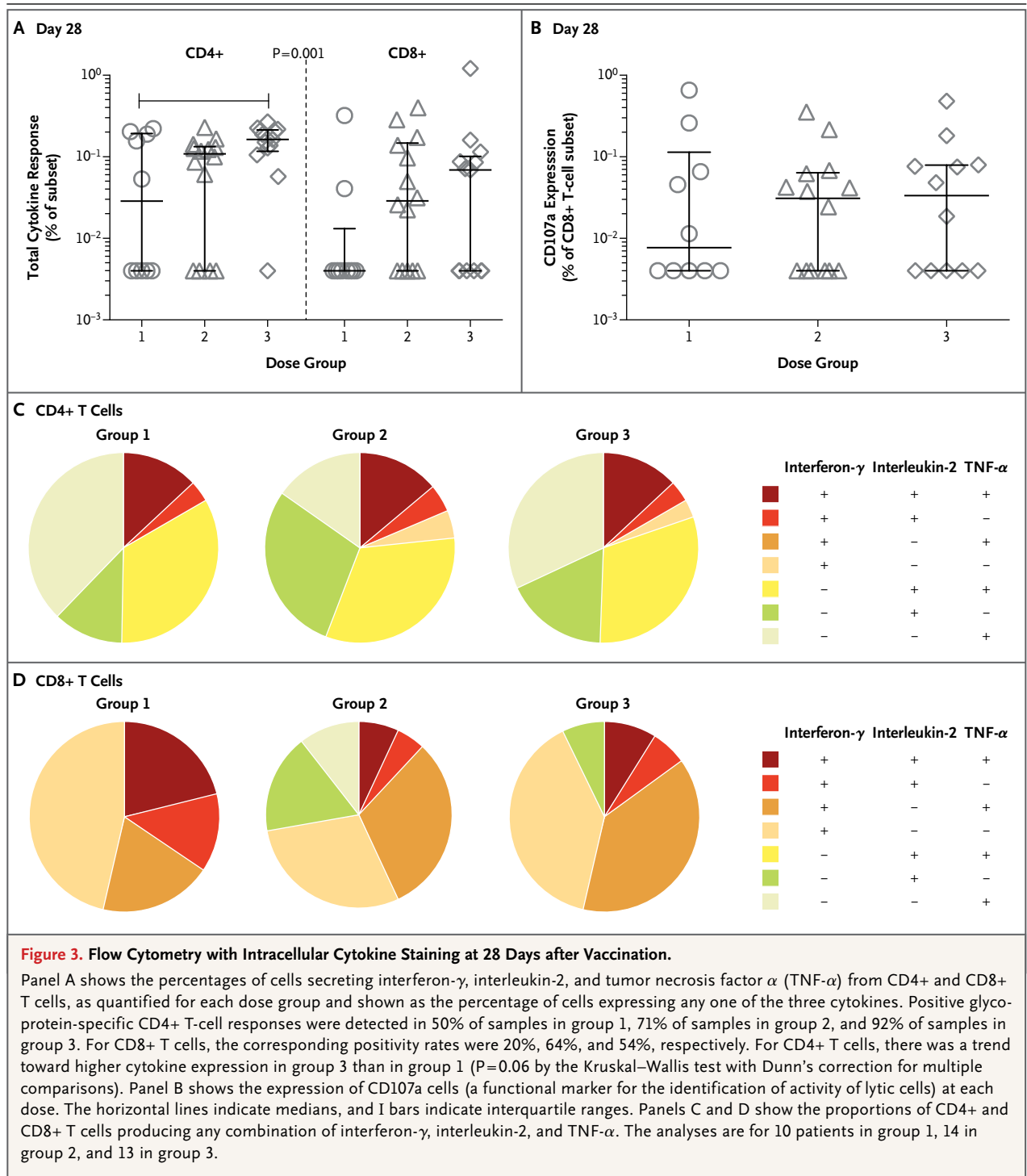
Figure 2. Responses to Peptides Spanning the Glycoprotein at Increasing Doses of Vaccine.

Panel A shows median responses on interferon- γ enzyme-linked immunospot (ELISpot) assays to 10 summed glycoprotein peptide pools at increasing doses of the ChAd3 vaccine. Numbers indicate the number of data points included at each time point. Responses are measured in the number of spot-forming cells (SFCs) per million peripheral-blood mononuclear cells (PBMCs). Panel B shows responses before immunization at baseline. Panel C shows responses at the peak of the cell-mediated immune response at 14 days after vaccination, with the percentages of participants who had a positive response (as indicated by a significant increase over the background prevaccination level) shown for each group. In Panels B and C, the horizontal lines represent medians. The analyses are for 19 patients in group 1, 20 in group 2, and 19 in group 3.

of responses that we observed, about 0.07% of CD8+ T cells, appears to be lower than that reported for macaques protected by this vaccine.² As expected, there was substantial polyfunctionality in the observed T-cell responses, but the role of T-cell quality in protection against ebolavirus remains unclear.

If greater immunogenicity is required, one option is to use a higher dose of vaccine, as suggested by the improved immunogenicity of a bivalent formulation of this vaccine¹⁶ encoding the glycoproteins of both the Sudan and Zaire strains at a dose of 2×10^{11} viral particles, and two trials of higher-dose monovalent vaccines are in progress. Another possible option for inducing a higher response is to boost the immunogenicity of ChAd-primed antibody and T-cell responses by means of an MVA vector encoding the same antigen.³⁻⁵ Reported antibody and T-cell responses in humans with the use of a booster dose of an MVA vector are increased by a factor of approximately 30 for antibodies and by a factor of 5 to 10 for T-cell responses.^{3-5,7} In macaques, the activation of antibodies that was observed with an MVA vector encoding the EBOV glycoprotein was increased by a factor of 30, as compared with ChAd3 priming alone, and there was an increase in the frequency of antigen-specific T cells by a factor of approximately 5, with improved durability of vaccine efficacy.² Phase 1 assessment of a booster study of some of these vaccinees with an MVA vector began in late November 2014. Future phase 3 efficacy trials could consider the potential need for such a booster dose.

CD4+ T-cell response, in contrast to the predominant CD8+ T-cell responses induced by chimpanzee adenoviral vectors in many animal models. Although direct comparison is difficult because responses in the macaque study were reported as a percentage of the subpopulation of memory cells rather than as a percentage of all CD4+ and CD8+ lymphocytes,² the magnitude



In conclusion, the safety and immunogenicity profile of the ChAd3 vaccine in our study encourage further assessment of this vector alone and in heterologous prime-boost regimens in

future phase 1 and phase 2 trials, as well as consideration of the inclusion of this vaccine vector in proposed phase 3 trials in countries in which Ebola is endemic in early 2015.

The views expressed in this article are those of the authors and do not necessarily represent the position or policies of the World Health Organization.

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Disclosure forms provided by the authors are available with the full text of this article at NEJM.org.

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APPENDIX

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