Received: 20 August 2023 | Accepted: 4 December 2023

DOI: 10.1002/jmv.29317

SHORT COMMUNICATION

JOURNAL OF MEDICAL VIROLOGY WILEY

Pan-pox-specific T-cell responses in HIV-1-infected individuals after JYNNEOS vaccination

Marta Sisteré-Oró[1] | Juan Du[2] | Diana D. J. Wortmann[1] | Marina D. Filippi[1] | Esperanza Cañas-Ruano[2] | Itziar Arrieta-Aldea[2] | Agustín Marcos-Blanco[2] | Xavier Castells[3] | Santiago Grau[4] | Natalia García-Giralt[2,5] | Daniel Perez-Zsolt[6] | Rytis Boreika[6] | Nuria Izquierdo-Useros[6,7,8] | Robert Güerri-Fernandez[2,8,9] | Andreas Meyerhans[1,10]

[1]Infection Biology Laboratory, Department of Medicine and Life Sciences, Universitat Pompeu Fabra (UPF), Barcelona, Spain

[2]Infectious Diseases Unit, Hospital del Mar Research Institute, Barcelona, Spain

[3]Preventive Medicine Unit, Hospital del Mar Research Institute, Barcelona, Spain

[4]Department of Pharmacy, Hospital del Mar, Barcelona, Spain

[5]Centro de Investigación Biomédica en Red de Fragilidad y Envejecimiento Saludable (CIBERFES), Instituto de Salud Carlos III, Madrid, Spain

[6]IrsiCaixa, AIDS Research Institute, Can Ruti Campus, UAB, Badalona, Spain

[7]Germans Trias i Pujol Research Institute (IGTP), Badalona, Spain

[8]CIBER Enfermedades Infecciosas (CIBERINFEC), Instituto de Salud Carlos III, Madrid, Spain

[9]Department of Medicine and Life Sciences (MELIS), Universitat Pompeu Fabra, Barcelona (UPF), Barcelona, Spain

[10]ICREA, Catalan Institution for Research and Advanced Studies Pg. Lluís Companys 23, Barcelona, Spain

Correspondence
Robert Güerri-Fernandez, Infectious Diseases Unit, Hospital del Mar Research Institute, 08003 Barcelona, Spain.
Email: rguerri@psmar.cat

Andreas Meyerhans, Infection Biology Laboratory, Department of Medicine and Life Sciences, Universitat Pompeu Fabra (UPF), 08003 Barcelona, Spain.
Email: andreas.meyerhans@upf.edu

Funding information
Red de Fragilidad y Envejecimiento Saludable (CIBERFES), Grant/Award Number: CB16/10/00245; Centro de Investigación Biomédica en Red Enfermedades Infecciosas, Grant/Award Number: CB21/13/00002; FIS Project, Grant/Award Number: PI19/00019; Spanish Ministry of Science and Innovation, Grant/Award Number: PID2022-141395OB-I00; Spanish Ministry of Science and Innovation, Grant/Award Number: PID2020-117145RB-I00; EU HORIZON-HLTH-2021CORONA-01, Grant/Award Number: 101046118

Abstract

People living with human immunodeficiency virus (HIV) are the individuals most affected by the current Monkeypox virus outbreak that was first announced in May 2022. Here we report Pan-pox-specific T-cell responses in a cohort of HIV-1-infected individuals after receiving the nonreplicative, attenuated smallpox vaccine JYNNEOS from Bavarian Nordic. Intradermal (i.d.) and subcutaneous (s.c.) vaccination was safe without major side effects. Dose-sparing i.d. vaccination was superior to s.c. vaccination and promoted T-cell polyfunctionality, and the expression of the gut-homing marker α4β7 integrin on lymphocytes. HIV-1-infected individuals with CD4 T-cell counts ≤500/mm^3 blood required at least a booster vaccination to exhibit efficient virus-specific T-cell responses. The magnitude of the Th1 response after this booster directly correlated with the CD4 T-cell count of the vaccinees. Further studies with a larger number of participants are warranted to confirm and expand our observations.

KEYWORDS
HIV-1-infected individuals, JYNNEOS vaccination, monkeypox (mpox), pan-pox-specific T-cell responses

1 | INTRODUCTION

Monkeypox (mpox) is a zoonotic virus infectious disease that can spread autochthonously between humans via direct contacts and respiratory routes.[1] It is caused by the mpox virus (MPXV), a member of the genus *Orthopoxvirus* of the family of *Poxviridae*, and is associated with fever, headache, muscle pain, swollen lymph nodes, rash, respiratory and rectal symptoms, exhaustion, and others.[2] Depending on the MPXV strain, and the age and immunocompetence of the host, disease symptoms can be very severe and may lead to death in few cases.[3] MPXV is antigenically related to other *Orthopoxviruses* including variola virus, the cause of smallpox.[4] This antigenic relatedness is the reason why smallpox vaccines provide >85% protection against MPXV infection in humans.[5]

With the worldwide eradication of smallpox in 1978 and the cessation of the mass smallpox vaccination campaigns, the incidence of mpox has continuously increased.[6] Mpox usually appeared as individual outbreaks that were geographically limited to endemic areas in Central and West Africa.[7] This situation dramatically changed in spring 2022 when a worldwide outbreak beyond the endemic regions was reported,[8] and human-to-human MPXV transmission has been estimated to occur since at least 2016. The cases were mainly among men having sex with men, many of which being persons living with human immunodeficiency virus (HIV) infection.[8,9] Within this latter group, individuals with high HIV loads and/or low CD4 T-cell counts were especially susceptible to severe mpox disease.[9] To protect this highly vulnerable group of individuals against MPXV infection and reduce virus spreading, vaccination would be the preferred option. However, a specific MPXV vaccine does not exist and, thus, a vaccination campaign would rely on the protective crossreactivity of smallpox vaccines. From the available ones, the nonreplicative JYNNEOS vaccine, approved in 2019, seems the best option.[10] It induces negligible antibody responses but robust T-cell responses[11,12] that play a critical role against poxvirus infections.[13] Furthermore, it has less side effects than vaccines such as ACAM2000™, LC16 or Dryvax that are based on replicative vaccinia viruses and are contraindicated for immuno-compromised individuals.[14] Importantly, American and European health authorities proposed a dose sparing intradermal (i.d.) route of JYNNEOS administration instead of the usual subcutaneous route (s.c), which extends the available vaccine doses by a factor of 5 and thus better covers increasing vaccine demands without compromising vaccine efficacy.[14,15] However, clinical trials that evaluate JYNNEOS vaccination responses and compare different administration routes in HIV-1-infected individuals are lacking. Here we analyzed JYNNEOS vaccination-induced pan-pox-specific T-cell responses from HIV-1-infected individuals whose virus load was controlled by antiretroviral therapy and whose CD4 T-cell counts were either ≤500/mm^3 blood (loCD4-group) or ≥500/mm^3 blood (hiCD4-group). The vaccine responses were compared with those from vaccinated healthy control (HC) individuals. We demonstrate that the dose-sparing i.d. vaccination route is preferable over the s.c. route, and that loCD4 individuals may require at least one vaccine booster to generate an efficient pan-pox-specific T-cell response.

2 | MATERIALS AND METHODS

2.1 | Sample collection and participant characteristics

This is an observational study that prospectively collected data and blood samples from HIV-1-infected individuals and HCs who were vaccinated with JYNNEOS, a modified vaccinia Ankara attenuated vaccine developed by Bavarian Nordic. The study groups consisted of (i) HIV-1-infected individuals with CD4 T-cell counts ≤ 500/mm^3 blood (n = 10; loCD4), (ii) HIV-1-infected individuals with CD4 counts ≥ 500/mm^3 (hiCD4) vaccinated s.c (n = 7), (iii) HIV-1-infected individuals with CD4 counts ≥ 500/mm^3 (hiCD4) vaccinated i.d (n = 7), (iv) HC vaccinated i.d (n = 7) as positive controls, and (v) unvaccinated healthy individuals (n = 7) serving as negative controls for the T-cell assays. All groups received a single JYNNEOS dose (0.5 mL; s.c vaccinees or 0.1 mL; i.d. vaccinees), except the loCD4 individuals who received a booster after 28 days due to their immunocompromised condition. Blood samples were collected before (Day 0) (pre-vaccine) and after first vaccination (28 days after vaccination) (1st vacc), with an additional collection 28 days after the booster vaccination for the loCD4 group (2nd vacc). Additionally, from 24 immunized participants, serum samples were taken 4–12 months post vaccination.

Participant characteristics of the 24 HIV-1-infected individuals are outlined in Table 1. The median age of the HIV-1-infected individuals was 34 years (interquartile range [IQR]: 23–44), all were male, and only one had a coinfection. All were on highly active antiretroviral therapy, with 95.83% (23/24) having an undetectable HIV-1 viral load. The vaccinated HC individuals had a median age of 43 years (IQR: 27–59) and were all male. Unvaccinated HC individuals had a median age of 47 years (IQR: 27–67), with 28.56% (2/7) being male. All participants provided written informed consent and the study protocol was approved by the institutional review board of the Hospital del Mar, Barcelona.

2.2 | Quantification of pan-pox-specific immune responses

Specific T-cell responses were assessed from peripheral blood mononuclear cells (PBMCs) as previously described using enzyme-linked immunospot (ELISpot)[16] and intracellular cytokine staining (ICS)[17] with adaptations as specified in the Supporting Information S8: Extended Method). The gating strategy used for ICS by flow cytometry is shown in Supporting Information S1: Figure S1. To quantify specific Th1 responses from CD69+CD4+ and CD8+ T cells, tumor necrosis factor-α (TNF-α) and/or interleukin-2 (IL-2), and/or

TABLE 1 Characteristics of HIV-1-infected individuals.

Group	Administration route	Patient	Gender	Age	Current coinfection	Age of HIV diagnosis	Start of HAART (year)	HAART	Current viral load (copies/mL)	Current CD4 + T-cell count	Nadir CD4 + T-cell count
HIV-1 loCD4	s.c.	Patient 1	Male	44	No	2018	2018	DTG-3TC	uLD	188	216
	s.c.	Patient 2	Male	44	No	2012	2012	DAR/c-TAF-FTC	uLD	116	77
	s.c.	Patient 3	Male	34	Yes (HHV8)	2016	2016	BIC-TAF-FTC	uLD	349	35
	s.c.	Patient 4	Male	44	No	2012	2012	DAR/c-TAF-FTC	uLD	264	45
	s.c.	Patient 5	Male	34	No	2019	2019	DAR/c-DTG	uLD	404	339
	s.c.	Patient 6	Male	35	No	2015	2015	ELV/c-TAF-FTC	uLD	500	13
	s.c.	Patient 7	Male	39	No	2020	2020	BIC-TAF-FTC	uLD	304	278
	s.c.	Patient 8	Male	37	No	2019	2019	DAR/c-TAF-FTC	37	402	175
	s.c.	Patient 9	Male	28	No	2019	2019	BIC-TAF-FTC	uLD	259	117
	i.d.	Patient 10	Male	45	No	2015	2015	EFV-TDF-FTC	uLD	499	
HIV-1 hiCD4	s.c.	Patient 11	Male	34	No	2015	2015	DTG-3TC	uLD	857	
	s.c.	Patient 12	Male	43	No	2009	2009	RPV-TAF-FTC	uLD	757	232
	s.c.	Patient 13	Male	35	No	2014	2014	DAR/c-TAF-FTC	uLD	541	175
	s.c.	Patient 14	Male	43	No	2007	2007	DTG-RPV	uLD	698	340
	s.c.	Patient 15	Male	28	No	2017	2018	RPV-TAF-FTC	uLD	701	
	s.c.	Patient 16	Male	30	No	2019	2019	BIC-TAF-FTC	uLD	739	741
	s.c.	Patient 17	Male	25	No	2018	2018	BIC-TAF-FTC	uLD	807	
	i.d.	Patient 18	Male	49	No	2007	2011	DTG-3TC	uLD	940	550
	i.d.	Patient 19	Male	41	No	2016	2016	BIC-TAF-FTC	uLD	959	359
	i.d.	Patient 20	Male	23	No	2022	2022	BIC-TAF-FTC	uLD	904	429
	i.d.	Patient 22	Male	39	No	2018	2018	BIC-TAF-FTC	uLD	1804	609
	i.d.	Patient 23	Male	39	No	2018	2018	DTG-3TC	uLD	1471	515
	i.d.	Patient 24	Male	30	No	2016	2016	DTG-3TC-ABC	uLD	603	
	i.d.	Patient 25	Male	43	No	2019	2019	DTG-3TC-ABC	uLD	788	490

Abbreviations: HHV8, human herpesvirus-8; HIV, human immunodeficiency virus; i.d., intradermally; uLD: undetectable HIV-1 viral load; HAART, highly active antiretroviral therapy; s.c., subcutaneously.

interferon-γ (IFN-γ)-positive CD4+ or CD8+ T cells were analyzed using FlowJo software's Boolean tool "make or gate." Values above 0.04 were considered positive. The α4β7 analyses followed a similar gating strategy (Supporting Information S1: Figure S1), with positivity set at 0.015 for frequencies of α4β7+ cells from TNF-α and/or IL-2, and/or IFN-γ-positive CD69+ CD4 and CD8 T cells. T-cell polyfunctionality was assessed by Boolean tool "make and gate."

Binding antibodies against the MPXV surface proteins A29 (Sino Biologicals, SIB-40891-V08E-100) and A35 (Sino Biologicals, SIB-40886-V08H-100) were determined by enzyme-linked immunosorbent assays,[16] whereas neutralizing antibodies were determined by reduction of MPXV infectivity of Vero E6 cells.[18] An extended Method description is given in the Supporting Information S8.

3 | RESULTS AND DISCUSSION

JYNNEOS vaccination has been shown to reduce the incidence of mpox and provide some protection against MPXV infection.[19] To potentially protect highly vulnerable HIV-1-infected individuals during the recent MPXV outbreak and to evaluate their poxvirus-specific T-cell responses after vaccination, 24 HIV-1-infected individuals, all outpatients of the Infectiology Unit of the Hospital del Mar, Barcelona, participated in this study (see Table 1 for details). They were grouped according to their CD4+ T-cell counts as loCD4 or hiCD4, and received either the standard s.c. JYNNEOS vaccination (before August 2022) or the emergency dose-sparing i.d. vaccination after its approval in August 2022.[14] Seven HC individuals that work in the hospital and are exposed to MPXV-infected individuals also received the JYNNEOS vaccine as a prophylaxis. They were all i.d. vaccinated and served as positive controls for the induced T-cell responses. PBMCs from seven unvaccinated HCs served as negative controls. None of the participating individuals had been infected with MPXV. There were no changes in CD4+ T-cell counts or viral loads nor major side effects due to vaccination. The binding and neutralizing antibody titers from the available sera of 24 study participants (see Supporting Information S2: Figure S2) were low as previously described.[11,12]

To first evaluate JYNNEOS vaccine-induced T-cell responses in HIV-1-infected individuals ($n = 24$) and HCs ($n = 7$), PBMCs were isolated from all participants before and 28 days after the first vaccination time point, stimulated with a peptide pool of 127 pan-pox-specific HLA-I and HLA-II epitopes, and subjected to IFN-γ ELISpot and ICS assays. Results for the latter are given as composite Th1 responses in Figures 1–3, whereas the individual cytokine responses (IFN-γ, TNF-α, and IL-2) are given in Supporting Information S3–S5: Figures S3–S5. Strong and specific Th1 responses were observed for both groups of immunized participants (Figure 1A,B), emphasizing the high immunogenicity of the vaccine even after a single immunization. However, although all HCs responded well to the vaccine, nearly 30% (six of 22) of the HIV-1-infected individuals did not adequately respond (Figure 1A). All six low responders were vaccinated s.c. Three were from the loCD4- and three from the hiCD4 group. As all HC individuals were vaccinated i.d., it remains uncertain whether the low response was due to a lower immunogenicity of a single s.c. vaccination or to a lower vaccine response due to HIV-1 infection. Nonetheless, our previous results in immunizing HIV-1-infected individuals with the potent T-cell response-inducing messenger RNA-based coronavirus 2019 vaccine BNT162b2 argue to strongly consider a low immune responsiveness of this patient group.[16]

The CD4+ T-cell count in HIV-1-infected individuals is inversely related to their immunodeficiency. To test whether and how the CD4+ T-cell count relates to JYNNEOS vaccine responsiveness, pan-pox-specific T-cell responses of the loCD4 group were analyzed 28 days after the first (1st vacc) and 28 days after the second s.c. vaccination (2nd vacc) (Figure 2). Albeit not all vaccinees responded, the second vaccination increased median ELISpot (1st vacc: 78 [0–156]; 2nd vacc: 130 [6–254]) and Th1 CD4 (1st vacc: 0.455 [0.07–0.84]; 2nd vacc: 0.815 [0–1.63]) and CD8 T-cell responses (1st vacc: 0.340 [0–0.68]; 2nd vacc: 0.480 [0–0.96]) (Figure 2A–D). There was a significant linear relation between CD4+ T-cell counts and the vaccine-induced IFN-γ ELISpot response after the second vaccination (Figure 2B). Thus, the standard two-dose s.c. JYNNEOS vaccination efficiently induces pan-pox-specific T-cell responses in HIV-1-infected individuals in a CD4+ T-cell-count-dependent manner.

To cover the increased vaccine demands for protecting against mpox, the dose sparing i.d. route of JYNNEOS administration was proposed by the health authorities in the US and Europe.[14,15] Accordingly, we adapted our vaccine administration route so that study participants after August 2022 were given 0.1 mL i.d. instead of 0.5 mL s.c. JYNNEOS shots. This enabled us to compare both administration routes in the hiCD4 group of HIV-1-infected individuals (Figure 3A–C). Importantly, all HIV-1-infected individuals generated significant pan-pox-specific T-cell responses after i.d. vaccination, which was not the case after s.c. vaccination. Furthermore, IFN-γ ELISpot levels were significantly higher after i.d. than after s.c. vaccine administration. This strongly supports the proposed dose-sparing vaccination route also for the protection of immunocompromised individuals. Figures combining all vaccination responses (Supporting Information S6: Figure S6) and T-cell polyfunctionalities (Supporting Information S7: Figure S7) are provided to enable simple head-to head comparisons.

The integrin α4β7 plays a crucial role in the trafficking of T cells to the gut-associated lymphoid tissue. There, the cells downregulate α4β7 and differentiate into long-lived resident memory cells that participate in tissue protection against intestinal pathogens.[20] As sexual MPXV transmission was considered a dominant infection route in the current MPXV outbreak,[8,9] it is conceivable that mucosal immune responses will have an important impact in protection from infection. To test whether JYNNEOS vaccine-induced T-cell responses might have mucosal-homing potential, we evaluated the expression of the α4β7 gut-homing integrin on pan-pox-specific T cells via ICS (Figure 3D,E). Interestingly, the i.d. immunized groups of HCs (7/7 individuals for CD4 T cells and 6/7 individuals for CD8 T

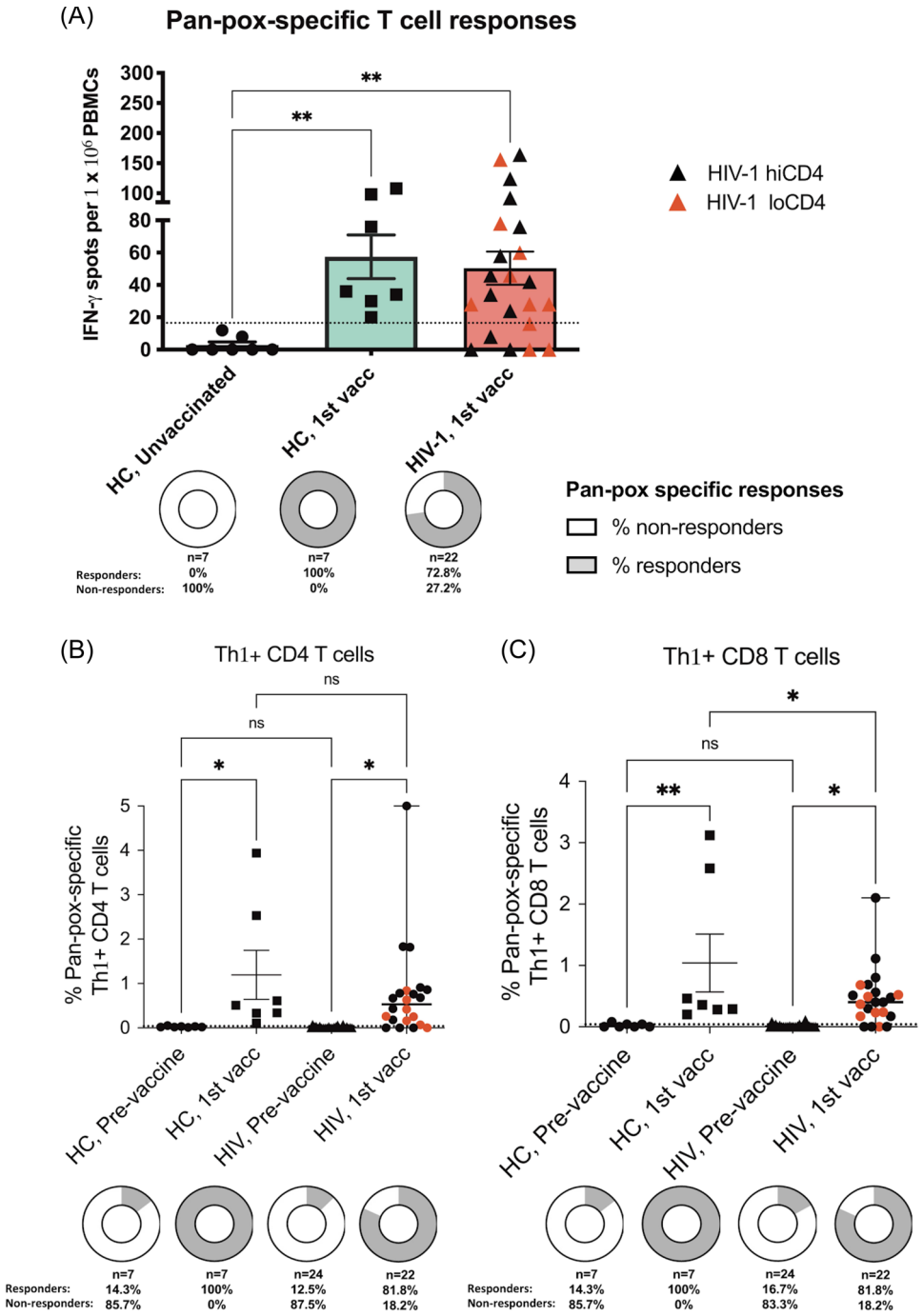

FIGURE 1 Pan-pox-specific T-cell responses in healthy controls (HCs) and human immunodeficiency virus-1 (HIV-1)-infected individuals after JYNNEOS vaccination. (A) Interferon-γ (IFN-γ) enzyme-linked immunospot (ELISpot) values (SFU per 10^6 peripheral blood mononuclear cells [PBMCs]) for HCs and HIV-1-infected individuals before vaccination and 28 days after the first vaccination. Percentages (%) of pan-pox-specific Th1 CD4+ (B) or CD8+ (C) responses determined by intracellular cytokine staining (ICS) for HC and HIV-1-infected individuals at prevaccination and after the first vaccination. Red triangles in the HIV-1-infected individuals group refer to the HIV-1 loCD4 group, whereas black triangles refer to the HIV-1 hiCD4 group. Statistically significant differences between the groups were calculated using a one-way analysis of variance test for parametric data or a Kruskal–Wallis test for nonparametric data. Nonsignificant differences were indicated as "ns" and differences with p values < 0.05 were considered significant and were indicated with asterisks: *$p < 0.05$; **$p < 0.01$; ***$p < 0.001$. Donut charts under figures A, B, and C show the number "n" of individuals analyzed and percentages of responders (gray-colored in chart) and non-responders (white-colored in chart).

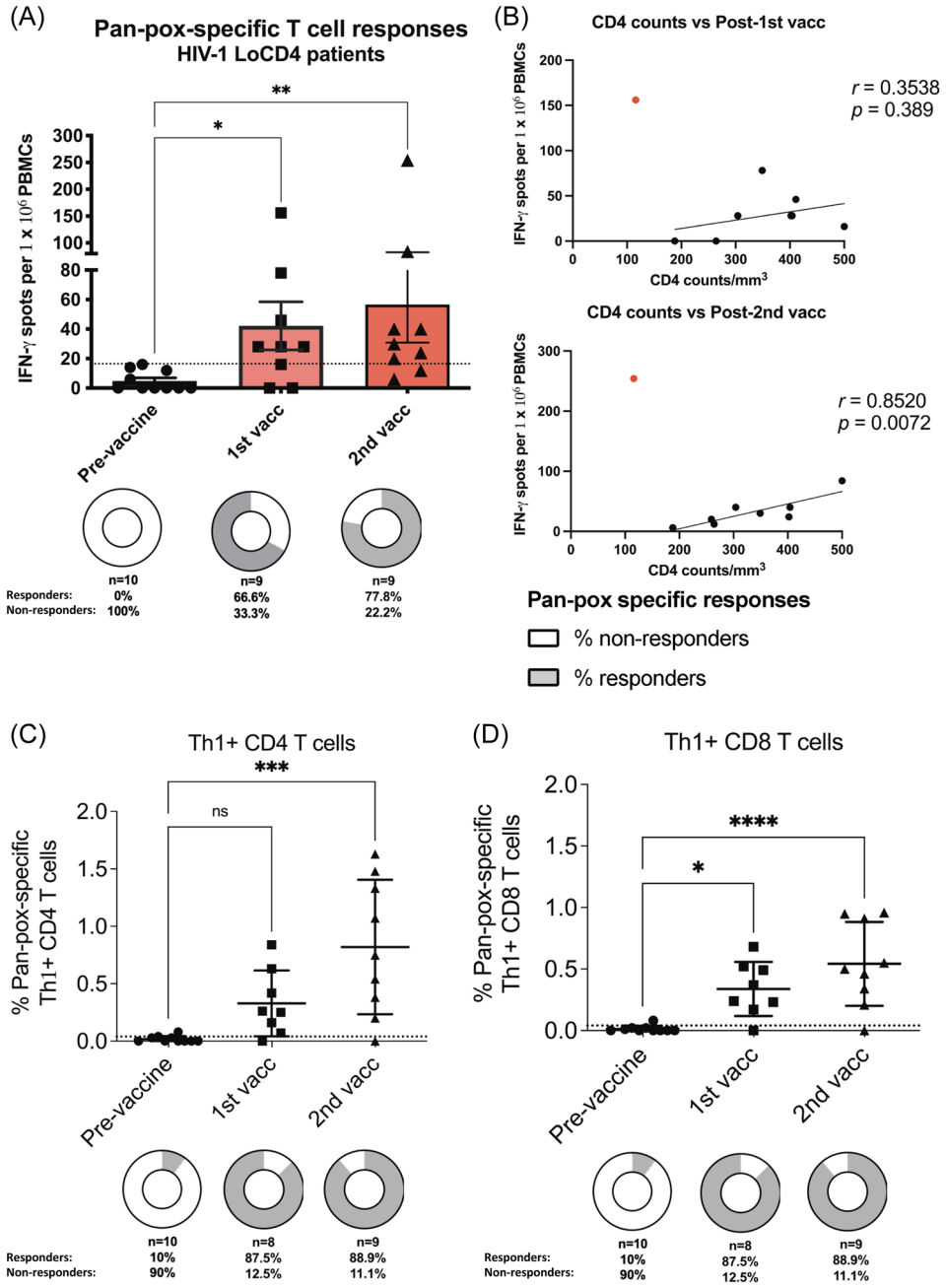

FIGURE 2 Pan-pox-specific T-cell responses in the human immunodeficiency virus-1 (HIV-1) loCD4 group after one or two JYNNEOS vaccinations. (A) Interferon-γ (IFN-γ) enzyme-linked immunospot (ELISpot) values (SFU per 10^6 peripheral blood mononuclear cells [PBMCs]) for the HIV-1 loCD4 at three defined time points: prevaccination (pre), after first vaccination (1st vacc; 28 days post vaccination), and after second vaccination (2nd vacc; 28 days post 2nd vaccination). (B) Correlation plots of CD4 counts/mm^3 blood (*x* axis) and IFN-γ values (*y* axis) for each individual as shown under (A) after the first and the second vaccination. Red point indicates a HIV-1-infected individual that was considered an outlier by Grubb's test and excluded from analyses. Percentages (%) of pan-pox-specific Th1 CD4+ (C) or CD8+ (D) T-cell responses determined by intracellular cytokine staining (ICS) for the HIV-1 loCD4 group at prevaccination and after the first and second vaccination. Statistically significant differences between the groups were calculated using a one-way analysis of variance test for parametric data or a Kruskal–Wallis test for nonparametric data. Nonsignificant differences were indicated as "ns" and differences with *p* values < 0.05 were considered significant and were indicated with asterisks: $^*p < 0.05$; $^{**}p < 0.01$; $^{***}p < 0.001$; p < 0.0001. Donut charts under figures A, C and D show the number "*n*" of individuals analyzed, and percentages of responders (gray-colored in chart) and nonresponders (white-colored in chart).

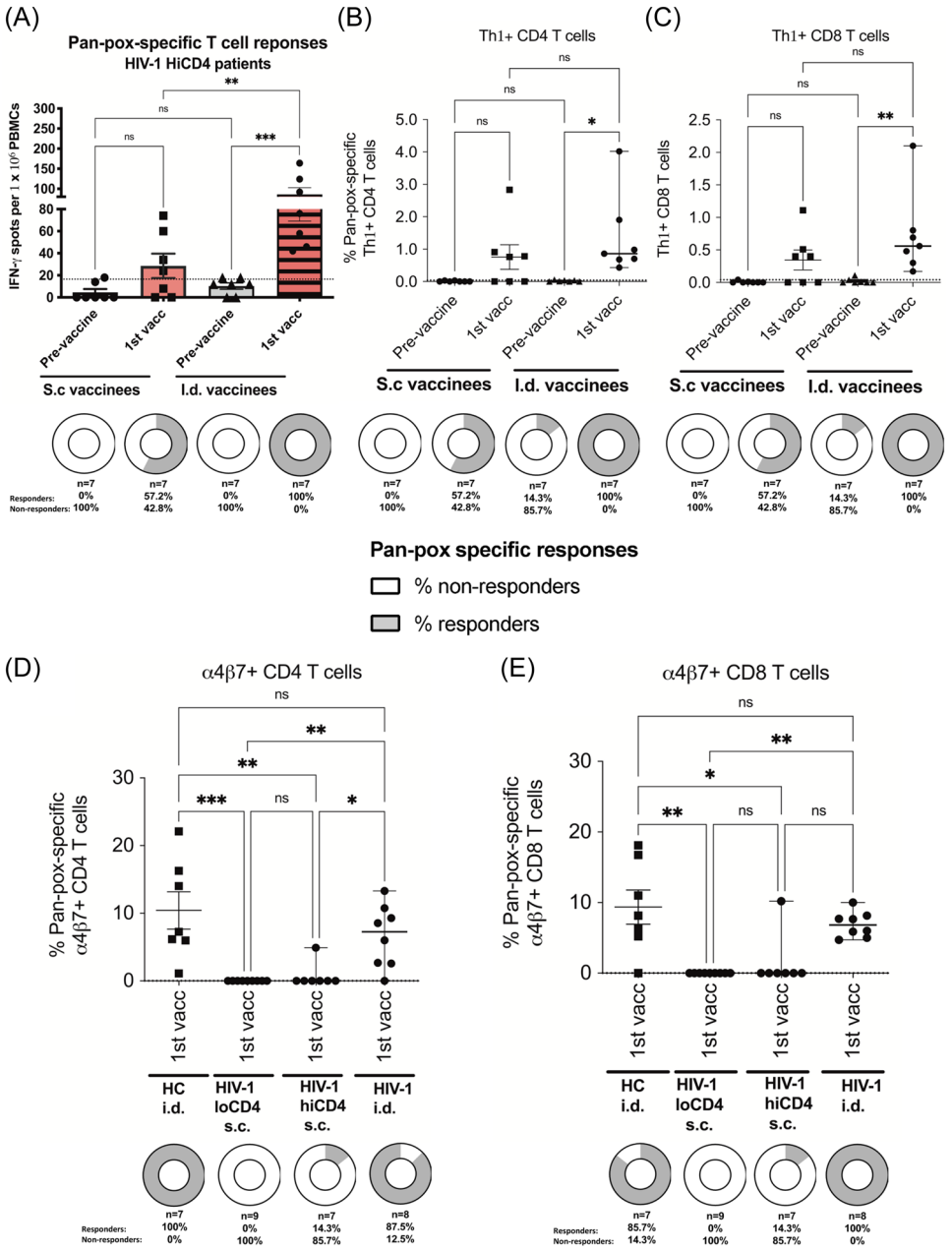

FIGURE 3 Comparison of subcutaneous (s.c.) and intradermal (i.d.) JYNNEOS vaccination routes. Pan-pox-specific interferon-γ (IFN-γ) enzyme-linked immunospot (ELISpot) (A), Th1 CD4+ (B), and Th1 CD8+ (C) T-cell responses after i.d. or s.c. JYNNEOS vaccination of human immunodeficiency virus-1 (HIV-1) hiCD4 group individuals. Pan-pox-specific α4β7+ CD4+ (D) or CD8+ (E) T-cell responses for HC and HIV-1-infected individuals after s.c. or i.d. JYNNEOS vaccination. Given are the differences in percentages to the prevaccination time point. Individual values for IFN-γ ELISpots and intracellular cytokine staining (ICS) assays are shown. Statistically significant differences between the groups were calculated using a one-way analysis of variance test for parametric data or a Kruskal–Wallis test for nonparametric data. Nonsignificant differences were indicated as "ns" and differences with p values < 0.05 were considered significant and were indicated with asterisks: *$p < 0.05$; **$p < 0.01$; ***$p < 0.001$. Donut charts under figures A–E show the number "n" of individuals analyzed and percentages of responders (gray-colored in chart) and nonresponders (white-colored in chart).

cells) and HIV-1-infected individuals (7/8 individuals for CD4 T cells and 8/8 individuals for CD8 T cells) showed higher integrin expression than the s.c immunized groups suggesting an additional potential benefit of i.d. vaccination over the standard s.c. JYNNEOS vaccination. This hypothesis awaits direct demonstration of pan-pox-specific T cells in mucosal tissue.

In summary, this study provides direct evidence for a superior immunogenicity of the dose-sparing i.d. JYNNEOS vaccination in HIV-1-infected individuals that are particularly susceptible to MPXV infection and pathogenicity. It emphasizes the need to specifically control vaccine responses in HIV-1-infected individuals with low CD4 T-cell counts as their vaccination responses may be severely

diminished. We hypothesize that i.d. vaccination may also improve protection by affecting polyfunctionality and homing characteristics of induced virus-specific T cells. Limitations of this study are the small number of vaccinees in each study group, and that the HC group solely received the dose-sparing i.d. vaccination due to vaccine supply shortages. Nonetheless, we provide an early indication of how to best proceed with preventive vaccination against mpox in a group of individuals with high-infection-risks and suggest clear directions for further studies to confirm and expand our observations.

AUTHOR CONTRIBUTION

Concept and funding acquisition: Robert Güerri-Fernandez, Nuria Izquierdo-Useros, and Andreas Meyerhans. *Experimental design*: Marta Sisteré-Oró, Robert Güerri-Fernandez, and Andreas Meyerhans. *Experiment performance*: Marta Sisteré-Oró, Juan Du, Diana D. J. Wortmann, Daniel Perez-Zsolt, and Rytis Boreika. *Patient recruitment and handling*: Juan Du, Esperanza Cañas-Ruano, Itziar Arrieta-Aldea, Agustín Marcos-Blanco, Xavier Castells, Santiago Grau, NG-G, and Robert Güerri-Fernandez. *Sample collection*: Juan Du, Esperanza Cañas-Ruano, Itziar Arrieta-Aldea, Agustín Marcos-Blanco, Xavier Castells, Santiago Grau, Natalia García-Giralt, and Robert Güerri-Fernandez. *Figures and tables*: Marta Sisteré-Oró and Marina D. Filippi. *Manuscript drafting*: Marta Sisteré-Oró, Marina D. Filippi, and Daniel Perez-Zsolt with corrections from Andreas Meyerhans, Nuria Izquierdo-Useros, and Robert Güerri-Fernandez. All authors contributed to the article and approved the submitted version.

ACKNOWLEDGMENTS

This work received support and funding from Centro de Investigación Biomédica en Red de Fragilidad y Envejecimiento Saludable (CIBERFES) (grant number CB16/10/00245), Centro de Investigación Biomédica en Red Enfermedades Infecciosas, CIBERINFEC (CB21/13/00002), FEDER funds, FIS Project "PI19/00019" funded by Instituto de Salud Carlos III (ISCIII) and co-funded by the European Union, the Spanish Ministry of Science and Innovation grant number PID2022-141395OB-I00, and the "Unidad de Excelencia María de Maeztu," funded by the MCIN and the AEI (DOI: 10.13039/501100011033). Nuria Izquierdo-Useros is supported by the Spanish Ministry of Science and Innovation (grant PID2020-117145RB-I00, Spain), EU HORIZON-HLTH-2021CORONA-01 (grant 101046118, European Union).

CONFLICT OF INTEREST STATEMENT

The authors declare no conflict of interest.

DATA AVAILABILITY STATEMENT

The data that supports the findings of this study are available in the Supporting Information of this article and from the corresponding author upon reasonable request.

ORCID

Rytis Boreika http://orcid.org/0000-0001-6720-1577
Robert Güerri-Fernandez http://orcid.org/0000-0002-9794-9161
Andreas Meyerhans http://orcid.org/0000-0003-0214-6348

REFERENCES

1. Hernaez B, Muñoz-Gómez A, Sanchiz A, et al. Monitoring monkeypox virus in saliva and air samples in Spain: a cross-sectional study. *Lancet Microbe*. 2023;4(1):e21-e28. doi:10.1016/s2666-5247(22)00291-9
2. Kumar N, Acharya A, Gendelman HE, Byrareddy SN. The 2022 outbreak and the pathobiology of the monkeypox virus. *J Autoimmun*. 2022;131:102855. doi:10.1016/j.jaut.2022.102855
3. Kozlov M. Monkeypox goes global: why scientists are on alert. *Nature*. 2022;606(7912):15-16. doi:10.1038/d41586-022-01421-8
4. Grifoni A, Zhang Y, Tarke A, et al. Defining antigen targets to dissect vaccinia virus and monkeypox virus-specific T cell responses in humans. *Cell Host Microbe*. 2022;30(12):1662-1670. doi:10.1016/j.chom.2022.11.003
5. World Health Organization. Vaccines and immunization for Monkeypox. Published June 14, 2022. Accessed August 11 2023; https://apps.who.int/iris/bitstream/handle/10665/356120/WHO-MPX-Immunization-2022.1-eng.pdf
6. Rimoin AW, Mulembakani PM, Johnston SC, et al. Major increase in human monkeypox incidence 30 years after smallpox vaccination campaigns cease in the Democratic Republic of Congo. *Proc Natl Acad Sci USA*. 2010;107(37):16262-16267. doi:10.1073/pnas.1005769107
7. Lum FM, Torres-Ruesta A, Tay MZ, et al. Monkeypox: disease epidemiology, host immunity and clinical interventions. *Nat Rev Immunol*. 2022;22(10):597-613. doi:10.1038/s41577-022-00775-4
8. Thornhill JP, Barkati S, Walmsley S, et al. Monkeypox virus infection in humans across 16 countries — April–June 2022. *N Engl J Med*. 2022;387(8):679-691. doi:10.1056/nejmoa2207323
9. Mitjà O, Alemany A, Marks M, et al. Mpox in people with advanced HIV infection: a global case series. *Lancet*. 2023;401(10380):939-949. doi:10.1016/s0140-6736(23)00273-8
10. Rao AK, Petersen BW, Whitehill F, et al. Use of JYNNEOS (Smallpox and Monkeypox vaccine, live, nonreplicating) for preexposure vaccination of persons at risk for occupational exposure to orthopoxviruses: recommendations of the advisory committee on immunization practices — United States, 2022. *MMWR Morb Mortal Wkly Rep*. 2022;71(22):734-742.
11. Cohn H, Bloom N, Cai GY, et al. Mpox vaccine and infection-driven human immune signatures: an immunological analysis of an observational study. *Lancet Infect Dis*. 2023;S1473-3099(23):00352-00353. doi:10.1016/s1473-3099(23)00352-3
12. Zaeck LM, Lamers MM, Verstrepen BE, et al. Low levels of monkeypox virus-neutralizing antibodies after MVA-BN vaccination in healthy individuals. *Nature Med*. 2023;29(1):270-278. doi:10.1038/s41591-022-02090-w
13. Kennedy RB, Ovsyannikova IG, Jacobson RM, Poland GA. The immunology of smallpox vaccines. *Curr Opin Immunol*. 2009;21(3):314-320. doi:10.1016/j.coi.2009.04.004
14. National Institute of Allergy and infectious Diseases (NIH). Mpox (formerly monkeypox) vaccines. Accessed August 11 2023. https://www.niaid.nih.gov/diseases-conditions/mpox-vaccines
15. European Medicines Agency. Mpox. Accessed August 11 2023. https://www.ema.europa.eu/en/human-regulatory/overview/public-health-threats/mpox#recommendations-on-use-of-mpox-medicines-during-2022-outbreak-section
16. Sisteré-Oró M, Andrade N, Wortmann DDJ, et al. Anti-SARS-COV-2 specific immunity in HIV immunological non-responders after mRNA-based COVID-19 vaccination. *Front Immunol*. 2022;13:994173. doi:10.3389/fimmu.2022.994173
17. Prenafeta A, Bech-Sàbat G, Moros A, et al. Preclinical evaluation of PHH-1V vaccine candidate against SARS-CoV-2 in non-human primates. *iScience*. 2023;26(7):107224. doi:10.1016/j.isci.2023.107224

18. Moraes-Cardoso I, Benet S, Carabelli J, et al. *Immune Responses Associated with Mpox Viral Clearance in a Prospective Observational Cohort of People With and Without HIV in Spain*. Elsevier; 2023. https://ssrn.com/abstract=4578561; doi:10.2139/ssrn.4578561
19. Payne AB, Ray LC, Kugeler KJ, et al. Incidence of monkeypox among unvaccinated persons compared with persons receiving ≥1 JYNNEOS vaccine dose — 32 U.S. jurisdictions, July 31–September 3, 2022. *MMWR Morb Mortal Wkly Rep*. 2022;71(40):1278-1282. doi:10.15585/mmwr.mm7140e3
20. Masopust D, Choo D, Vezys V, et al. Dynamic T cell migration program provides resident memory within intestinal epithelium. *J Exp Med*. 2010;207(3):553-564. doi:10.1084/jem.20090858

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Sisteré-Oró M, Du J, Wortmann DDJ, et al. Pan-pox-specific T-cell responses in HIV-1-infected individuals after JYNNEOS vaccination. *J Med Virol*. 2023;96:e29317. doi:10.1002/jmv.29317