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Simulating rabies post-exposure prophylaxis among patients with human immunodeficiency virus infection using a six-dose Essen regimen administrated with human diploid cell vaccine: A single-arm pilot study in Chinese population

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ABSTRACT

To explore the immunogencity and safety of HDCV using a six-dose Essen regimen in human immunodeficiency virus (HIV) infected patients. We conducted a single-arm pilot study by simulating post-exposure prophylaxis (PEP) in HIV-infected patients. All patients were administrated with HDCV using a 6-dose Essen regimen (consisting of 2 doses on day 0, and 4 doses each on day 3, 7, 14, and 28). Rabies virus-neutralizing antibody (RVNA) titers were detected on day 0, 7, 14, and 45, separately. The adverse reactions were also observed. In addition, we divided the patients with the baseline CD4+ T-cell counts of 500 cells/μL to examine the correlation between primary CD4+ T-cell counts and RVNA titers among HIV patients. Thirty patients included in the study were mostly male (96.7%), with a median age of 30.5 years and stable antiretroviral therapy (ART) treatment. Patients had RVNA titers of 0.84 IU/mL on day 7, 9.94 IU/mL on day 14, and 4.02 IU/mL on day 45 after vaccination, with significant differences between day 7 and day 14. The seroconversion rate reached 100% on day 14. Only three patients developed transient adverse reactions (including fever and redness, swelling, pain, and induration at the injection site）. There was no significant difference in antibody titers and safety profile between patients with CD4+T-cell counts below and above 500 cells/μL. A favorable immune response was achieved in HIV patients using the six-dose Essen regimen with HDCV. The safety profile of HDCV is satisfactory, with no major adverse events.

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Introduction

Acquired Immune Deficiency Syndrome (AIDS) remains a serious problem that affects global public health. Human Immunodeficiency Virus (HIV) mainly targets CD4+T-cell counts, compromising cellular immunity and resulting in significant immune dysfunction, which brings them an increased risk for numerous infectious diseases.[1] Rabies is an acute zoonotic viral disease caused by RNA viruses of the family Rhabdoviridae, genus Lyssavirus, with a nearly 100% mortality rate after the onset of the disease.[2,3] Notably, rabies post-exposure prophylaxis (PEP) requires comprehensive interventions beyond vaccination alone, as intramuscular immunization fails to confer absolute protection following mammalian injuries. Essential wound management protocols should be strictly implemented, comprising (a) local wound care through sequential procedures: (1) copious irrigation with soap and detergent solution for ≥15 min; (2) application of virucidal agents (e.g., 70% ethanol or 10% povidone-iodine); (b) anatomical-site-appropriate infiltration of rabies immunoglobulin (RIG) or monoclonal antibody cocktails (RmAb) to achieve immediate viral neutralization, a critical determinant of PEP efficacy. Currently, the number of rabies deaths worldwide is approximately 59,000 per year, with the majority of patients belonging to Asia and Africa, and China is one of the high-risk countries for rabies.[4] Vaccination remains an important measure in rabies post-exposure prophylaxis (PEP). However, the impairment of the immune system can diminish the effectiveness of vaccines given to individuals with HIV, obstructing their ability to achieve the desired protective outcomes.[5] Research indicates that HIV patients with low CD4+T cell counts struggle to develop adequate antibody levels after rabies vaccination.[6–8] Moreover, even those with high CD4+T cell counts and stable antiretroviral therapy (ART) still exhibit significantly lower levels of protective antibody titers than healthy individuals. A clinical study found that 5 years after 30 HIV patients (median CD4 count: 537 cells/μL) received two doses of the rabies vaccine, the seroconversion rate among HIV patients was only 63%, compared to 86% in non-HIV-infected individuals.[9] Another study examined 27 HIV patients with a history of moderate or severe immunodeficiency (CD4+T-cell counts < 300 cells/μL) and observed that their immune function improved following stable ART treatment (CD4+T-cell counts > 500 cells/μL). However, tests of immune

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function indicators revealed that, despite their CD4+T-cell counts t returning to normal levels, their immune function remained impaired.[10] In that case, the immunodeficiency status still exists among HIV patients with CD4+T cell counts above 500 cells/μL, suggesting an improved vaccination schedule is required for all HIV patients.

Currently, intramuscular injection is the primary method of rabies vaccination used worldwide. The WHO recommends that intramuscular rabies vaccine be administered for 2 weeks (1-1-1-1-0) or 3 weeks (2-0-1-0-1).[4] But the vaccination program commonly used in China is Essen (1-1-1-1-1), a five-dose regimen on days 0, 3, 7, 14, and 28, and Zagreb (2-1-1), a four-dose regimen that involves two sites IM on day 0 and one site IM on days 7 and 21.[11] People with HIV adhere to the same vaccination procedures as the general population, with no specific recommendations for their special needs. At present, several organizations worldwide have released guidelines on managing rabies post-exposure for individuals who are immunocompromised.[12–15] Organizations like the WHO and the UKHSA recommend that individuals with severe immunodeficiency or unvaccinated persons with altered immunocompetence should receive an additional vaccine dose and/or antibody testing within a specified period after completing the vaccine series to monitor the immunogenicity among those patients. However, thorough debridement procedures should be prioritized as the primary intervention following animal bites. Furthermore, the 2021 WHO consolidated guidelines on HIV stress that HIV patients may require supplemental doses or revaccination after ART-induced immune reconstitution.[16] While many guidelines have given their recommendations, there is limited research on its efficacy and application in HIV patients. This highlights the critical need to explore tailored post-exposure prophylaxis protocols for HIV patients and to evaluate the effectiveness of intramuscular injection in this population. According to the WHO guidelines and the current practical situation in China, we chose to double the first dose based on the immunization program of five doses of Essen.

This study aims to evaluate the use of a six-dose Essen regimen (with a double dose at the initial injection) for post-exposure prophylaxis in HIV patients who have not previously received the rabies vaccine. The objectives include assessing the effectiveness of the vaccine response, exploring the relation between CD4+T-cell counts and antibody titers, and developing a safe and effective post-exposure vaccination protocol tailored explicitly to the HIV-infected population. Additionally, the study aims to provide evidence-based support to enhance post-exposure prophylaxis standards in China.

Materials and methods

Study design and population

A single-center, single-arm pilot study was conducted at The Second Hospital of Nanjing. Thirty patients in total were recruited and visited between May 2023 and July 2024. The inclusion criteria were as follows: 1. Age between 18 and 65 years; 2. HIV infection; 3. No history of injury from mammals and no history of rabies vaccination (RVNA titer < 0.5 IU/mL before the first dose of vaccine). The exclusion criteria were as follows: 1. Females who were breastfeeding or planning to become pregnant during the trial; 2. History of severe vaccine side effects such as allergy, urticaria, dyspnea, angioneurotic edema,or abdominal pain; 3. Asthma, unstable for the past 2 years, requiring emergency treatment, hospitalization, intubation, oral or intravenous corticosteroids; 4. Epilepsy, Guillain-Barré syndrome; 5. Those with a fever with an axillary temperature of >37.0°C before the vaccine; 6. Patients with infections or co-infections such as active tuberculosis, fungal and cytomegalovirus infections.

The study protocol was approved by the Ethics Committee of The Second Hospital of Nanjing (No. 2022-LS-ky038). It was in accordance with the revised 2008 Declaration of Helsinki, and all patients signed an informed consent form.

Intervention description

We simulated PEP in HIV patients without rabies exposure by a six-dose Essen regimen, which means two sites IM at baseline (day 0) and one site IM on days 3, 7, 14, and 28, respectively. All rabies vaccines were manufactured by Chengdu Kanghua Biological Products Co., Ltd, reconstituted to 1 mL each, containing rabies vaccine potency of not less than 2.5 IU.

Procedure of rapid fluorescent focus inhibition test (RFFIT)

The RVNA titers were measured by RFFIT (Tested by Nanjing Zhongzhiheng Information Technology Co.). Briefly, the CVS-11 strain as challenged rabies virus was added into diluted serum sample in the 96 well tissue culture plates and then incubated at 37℃ for 1 h. Fifty microliters of cell suspension was added, and the plates were then incubated in a CO_2 incubator at 37℃ for 24 h. The plates were fixed with acetone solution, then stained by Fluorescein Isothiocyanate (FITC) conjugated anti-rabies antibody, and the plates were incubated in a CO_2 incubator at 37℃ for 1 h. The plates were observed using an inverted fluorescence microscope. The highest serum dilution demonstrated 50% inhibition of fluorescence foci, which was the endpoint of dilution. The titers were converted to IU/mL by comparing them with a reference serum. RVNA titers >0.5 IU/mL were considered seroconversion.[17]

Baseline data collection

Patient characteristics such as sex, height, weight, age, and duration of ART treatment were obtained with the patient's consent. Phenotypic analysis of T-cell subsets was done by flow cytometry, and baseline CD4+ T cell and CD8+T cell counts were collected. The baseline RVNA tests were conducted at day 0 before primary vaccination.

Immunogenicity evaluation

The primary endpoint to evaluate the immunogenicity of the six-dose Essen regimen using HDCV was the RVNA titer at day 14. In addition, secondary endpoints including the RVNA titers at

days 7 and 45 and the seroconversion rate at day 7, 14 and 45 were analyzed. In addition, a subgroup analysis was performed to assess the correlation between RVNA titers and baseline CD4+ T-cell count threshold.

Safety assessments

All subjects were kept under observation for 30 min after each vaccination for immediate reaction and at 24, 48, and 72 h for local and systemic reactions. Weekly follow-ups were conducted from day 4 to week 4 after each vaccination, combined with unsolicited reports from the subjects. The occurrences of injection site adverse reactions (e.g., tenderness, itching, swelling, hardness, cellulitis) and systemic adverse reactions (e.g., fever, headache, malaise, nausea, vomiting, allergy, arthralgia, respiratory distress) were collected. Adverse reactions were determined by the State Drug Administration's 'Guiding Principles for Adverse Reaction Classification Criteria in Clinical Trials of Preventive Vaccines.'[18] Adverse reactions or adverse events should be recorded in the case report form and data recording form normatively.

Statistical analysis

Data analysis was performed using R4.2.3. Patient demographic and clinical data were analyzed using descriptive statistics. The one-sample Kolmogorov–Smirnov test for normality was used for measurement data. Quantitative data, conforming to the normal distribution, was expressed as Mean ± SD, whereas skewed distribution data was expressed as median (interquartile range [IQR]), and categorical variables were expressed using frequency distributions. Differences between groups were analyzed using the independent samples t-test (for normal distribution data) or Mann–Whitney U-test (for skewed distribution data). Friedman test (for skewed distribution data) or paired t-test (for normal distribution data) was used to examine between independent groups. $p < .05$ was considered statistically significant.

Result

Characteristics of the study population

We enrolled 30 HIV patients in our study (Figure 1), and the pertinent baseline characteristics are presented in Table 1. The study population was predominantly male (96.7%), with a median

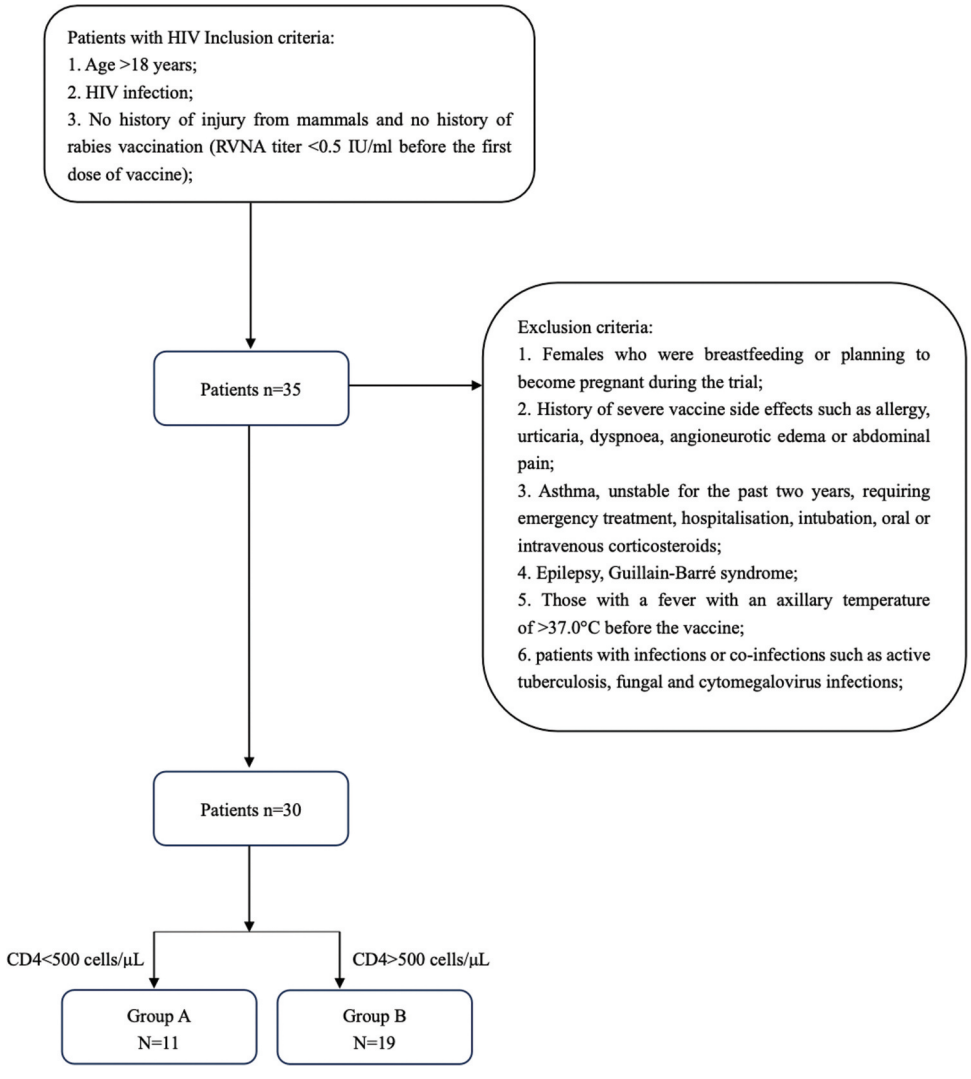

Figure 1. Study population inclusion flowchart.

Table 1. Basic characteristics of the study subjects and changes in antibody titers.

Characteristic	Patients ($n = 30$)
Age (median, IQR, years)	30.5 (27, 39.25)
Sex (n, %)	
Male	29 (96.7)
Female	1 (3.3)
BMI (mean±SD, kg/m^2)	23.51 ± 2.62
ART treatment time (median, IQR, years)	5 (1.00, 7.75)
CD3+T cell (median, IQR, cells/μL)	1322.50 (1141.25, 1864.50)
CD4+T cell (median, IQR, cells/μL)	616 (471.00, 751.25)
CD8+T cell (median, IQR, cells/μL)	683 (563.50, 801.75)
CD4+T CD8+T cell (median, IQR, cells/μL)	7.50 (4.25, 11.00)
Th/Ts (median, IQR)	0.865 (0.53, 1.13)

age of 30.5 years. Participants have been received ART treatment for a median duration of 5 years. Nineteen participants had CD4+ T-cell counts above 500 cells/μL, while three out of 30 patients had CD4+ T-cell counts <200 cells/μL, with a median CD4+T-cell counts of 616 cells/μL among all participants.

Immunogenicity after vaccination

As shown in Table 2, we observed that the median antibody titer of the participants was 0.84 (IQR, 0.37–3.15) IU/mL on day 7, 9.94 (IQR, 7.21–27.96) IU/mL on day 14, and 4.02 (IQR, 3.35–10.29) IU/mL on day 45. The results indicate a significantly increasing trend of RVNA titers from day 0 to day 14 (Figure 2).

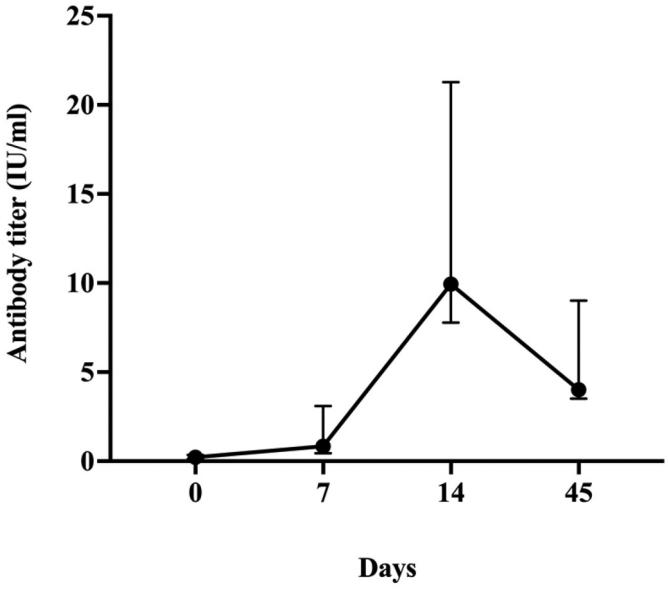

Figure 2. Changes in antibody titers in patients after vaccination.

The seroconversion rates on day 7, day 14, and day 45 were 66.7%, 100% and 100% (Table 2), separately. The difference between day 7 and day 14 is significant ($\chi 2 = 18.05$, $p < .01$), while no difference was observed between day 14 and day 45 ($p = .075$). In addition, among those three patients with CD4+ T-cell counts <200 cells/μL, one acquired protective antibodies on day 7, whereas the remaining two acquired protective antibodies on day 14.

Comparison of antibody titers in patients with different CD4+ T-cell counts threshold

In this analysis, we examined two patient groups: one comprising 11 individuals with CD4+ T -cell counts below 500 cells/μL (Group A) and the other consisting of 19 patients with CD4+ T-cell counts above 500 cells/μL (Group B). There were no significant differences in demographic information such as age, gender, and BMI between the two groups, except for CD4+ T, CD3+ T-cell counts, and Th/Ts. Importantly, for both groups, the antibody titers measured at days 0, 7, 14, and 45 post-vaccination revealed no significant differences. Similarly, the seroconversion rate on day 7, 14, and 45 were comparable between the two groups (Table 3).

Safety of HDCV injections for HIV patients

Among the 30 hIV patients administered the HDCV vaccine, three patients (10% of the cohort) experienced adverse reactions, with one in Group A and the other two in Group B (Table 3). Specifically, two of them encountered redness, swelling, and pain accompanied by hard nodules at the vaccination site. These symptoms persisted for 2–3 days and subsequently recovered without intervention. Notably, one of these two patients reported an adverse reaction following the initial dose, while the other experienced it after the third dose.

Table 2. Comparison of antibody titers and antibody positive conversion rate in patients on different days after vaccination.

	0-day	7-day	14-day	45-day	P_1	P_2	P_3	P_4
RVNA, median, IQR, IU/mL	0.22 (0.12,0.39)	0.84 (0.37,3.15)	9.94 (7.21,27.96)	4.02 (3.35,10.29)	0.008	<0.001	0.075	0.006
Seroconversion rate, n (%)		20 (66.7)	30 (100)	30 (100)		<0.001		0.075

RVNA: rabies virus neutralizing antibody; IQR: interquartile range.
P_1: 0-day vs. 7-day.
P_2: 7-day vs. 14-day.
P_3: 14-day vs. 45-day.
P_4: 7-day vs. 45-day.

Table 3. Comparison of characteristics and antibody titers of patients with CD4+ T-cell counts <500 cells/μL (Group A) and >500 cells/μL (Group B).

Characteristic	Group A ($n = 11$)	Group B ($n = 19$)	P value
Age, (median, IQR, years)	31 (27, 43.5)	30 (27.5, 36.5)	.914
Sex (n, %)			1.000
Male	11 (100)	18 (94.7)	
Female	0 (0)	1 (5.3)	
BMI	23.8 ± 3.8	23.4 ± 1.8	.729
ART treatment time (median, IQR, years)	2 (0.7, 5)	7 (3, 8.5)	.057
CD3+T cell, (median, IQR, cells/μL)	1104 (868.5, 1439.5)	1392 (1281.5, 1932)	.018
CD4+T cell, (median, IQR, cells/μL)	336 (205, 475)	673 (644.5, 870.5)	<.001
CD8+T cell, (median, IQR, cells/μL)	656 (537.5, 887.5)	710 (574.5, 799.5)	.621
CD4+T CD8+T cell, (median, IQR, cells/μL)	5 (3, 10.5)	9 (5, 11)	.226
Th/Ts (median, IQR,)	0.5 ± 0.3	1.1 ± 0.5	<.001
0-day antibody titer, (median, IQR, IU/mL)	0.2 (0.2, 0.3)	0.2 (0.1, 0.5)	.897
7-day antibody titer, (median, IQR, IU/mL)	1.3 (0.2, 4)	0.8 (0.4, 3.1)	.763
14-day antibody titer, (median, IQR, IU/mL)	9.2 (5.6, 22.1)	10.2 (7.7, 30.7)	.471
45-day antibody titer, (median, IQR, IU/mL)	3.5 (2.6, 7.5)	8 (3.4, 12.8)	.189
Day 7 antibody positive conversion rate (n, %)	7 (63.6)	13 (68.4)	1.000
Adverse Event, (n, %)	2 (18.2)	1 (5.3)	.54

Additionally, one patient developed a fever after the first vaccine dose. Furthermore, the incidence of adverse reactions did not significantly differ between Group A (2,18.2%) and Group B (1,5.3%) ($p = .54$).

Discussion

In our study, we implemented the six-dose Essen regimen for Rabies PEP in HIV patients, which can be regarded as a combination of the Essen and Zagreb regimens. On day 7, the seroconversion rate among HIV patients was only 66.7%. This suggests that administering two doses on day 0 followed by one dose on day 3 did not achieve a 100% seroconversion rate within 7 days. On the one hand, this result may be attributed to the limitation of inactivated vaccines in inducing immune responses, as previously confirmed by multiple clinical studies on inactivated vaccines.[19–21] Our study showed that even an additional dose on day 3 of the original Zagreb regimen does not significantly increase antibody titers. On the other hand, it may also result from the immunodeficiency in HIV patients, which obstructs their ability to generate the expected immune response. Studies indicate poor immunization in HIV patients vaccinated using standard procedures.[7,8] For instance, a 6-year-old HIV-infected girl from Thailand failed to respond to intramuscular pre-exposure rabies vaccination using HDCV administered on days 0, 7, and 28, as her antibody titers remained below detectable levels (<0.04 IU/mL). Subsequently, she also failed to respond to an intradermal postexposure rabies regimen, with antibody titers remaining low (<0.07 IU/mL).[7] Similar issues also exist with non-rabies vaccines,[21–25] such as the hepatitis B vaccine, IPV (inactivated polio vaccine), Influenza vaccine, or JEV (Japanese encephalitis virus) vaccine. Studies and guidelines indicate that the immune response in HIV patients may be lower than in fully immunocompetent persons when standard procedures are followed. For instance, in HIV-infected children, the immune response to the hepatitis B vaccine declined to 28–78% with the standard regimen, showing inadequate immunogenicity. However, following a booster dose, the seroconversion rate reached 51% at 30 ± 7 days, correlating with T cell immune memory development cells.[22] According to our study results, the overall antibody titers of the patients after vaccination started to increase on day 7, peaked on day 14, and decreased on day 45. This result is basically consistent with those obtained from the general population using standard PEP procedures, including Zagreb and Essen regimens.[26–29] However, the difference in RVNA titers between day 14 and day 45 is not significant. This might suggest that the Essen regimen with a doubling of first dose can lead better maintenance of short-term immunogenecity, which needs further studies with a larger sample size. All patients maintained adequate protective antibody titers on days 14 and 45, indicating that the 6-dose Essen regimen using HDCV is an appropriate vaccination protocol for PEP among HIV-infected patients. However, it should be noted that vaccination alone does not completely prevent the onset of rabies, life saving RIG/RmAbs and wound treatment in PEP is necessary.

All patients in our study achieved adequate protective antibody titers. Baseline data revealed that the study population primarily consisted of young males with high CD4+ T-cell counts (above 500 cells/μL, with a median CD4+T cell count of 616 cells/μL). Previous studies have not provided sufficient evidence to establish a correlation between gender and vaccine immunogenicity.[30–32] Therefore, we speculate that the results may be partially attributed to the characteristics of our study population, including a median age (30 years), higher CD4+T-cell counts, and stable antiretroviral therapy (ART). Age has already been shown to correlate with the immunogenicity of rabies and other vaccines.[33–35] A systematic review indicates that adolescents under 18 years old (median age 11) and younger adults (median age 28) reach their GMT peak 12 days earlier than older adults (median age 62), with older adults showing lower GMT peak levels.[34] Studies on other types of vaccines have also found similar results. Young women (age 19–54) with HIV, higher CD4+T-cell counts, and stable ART had higher vaccine seroprotection rates and better immunogenicity after receiving the influenza vaccine compared to the older women group (age >55).[33]

Subgroup analysis between patients with CD4+ T-cell counts below and above 500 cells/μL was also executed to compare their immunogenicity, which shows no significant difference. Among 11 patients with CD4+ T-cell counts below 500 cells/μL, three were <200 cells/μL, four were 200–350 cells/μL and five were 350–500 cells/μL. WHO recommended that HIV patients with CD4 cells>350 cells/μL could be considered immune recovery, but some countries adopted a threshold of CD4+T-cell counts >500 cells/μL. Meanwhile, most guidelines recommended ART initiation at CD4+T cell counts <500 cells/μL, or even >500 cells/μL.[36] An extensive post-2015 life expectancy cohort analysis of HIV-infected patients found that infected individuals who started ART after 2015 and had a baseline CD4+ T-cell count of ≥500 cells/μL had a similar life expectancy to healthy individuals at age 40.[37] Studies have demonstrated that CD4+T cell counts >500 cells/μL have a lower risk of infectious diseases such as Hepatitis C Virus (HCV),[38] as well as opportunistic infectious.[39] Those results have emphasized a better immune profile in HIV patients with CD4+T cell counts >500 cells/μL. However, our study did not find a correlation between short-time immunogenicity and CD4+T cell count threshold. This might be the result of the additional dose of primary vaccination. Nevertheless, limited studies compared vaccination status between HIV patients with different CD4 thresholds, indicating that further studies with large sample sizes were needed. Additionally, this study included populations with short ART time and lower CD4+T cell counts. However, the results showed that they still reached effective protective antibody titers, indicating that the six-dose Essen regimen using HDCV had good immunogenicity. Recent studies further showed that 28 days after the initial rabies vaccine was administered with the Essen regimen, HDCV produced significantly higher antibody titers than other vaccine types, such as PHKCV and PVRV.[40] These results may provide new clinical evidence supporting the use of HDCV in PEP for HIV patients.

Of the 35 patients, three had less than 200 CD4+ T cells/μL, and only 1 patient received protective antibodies on day 7, whereas the remaining 2 received protective antibodies on day 14. In a prospective study, it was noted that after HIV patients received a 4-site id post-vaccination regimen (4-4-4-4-0-2-2) using purified Vero cell rabies vaccine (doubling the id dose of cell culture rabies vaccine), three out of seven patients with CD4+ T-cells <200 cells/mL had poor or even no vaccine response.[41] Therefore, this study focused on a five-dose Essen regimen and used the doubled dose of six-dose Essen regimen to vaccinate HIV patients with HDCV, who expressed satisfactory immune response results, and this vaccination procedure may be more beneficial for HIV patients. Interestingly, we found that at day 7, 10 patients had antibody titers <0.5 IU/mL and only 2 of them had CD4+ T cells <200 cells/μL. This partly suggests that the rapid response of patients to the vaccine does not seem to be related to the number of CD4+ T cells but is more likely related to individual differences in nonspecific immune responses.

Only three patients in our study experienced adverse reactions. However, one of the patients developed fever after the first dose of HDCV. It is worth noting that this patient tested positive for COVID-19 over the following 3 days, casting doubt on whether the fever was solely attributable to the vaccine. Encouragingly, all adverse reactions reported resolved spontaneously in the affected patients, indicating a favorable safety profile for the HDCV vaccine in this patient population. HDCV even compares favorably to rabies vaccines such as Purified Vero Cell Vaccine (PVRV) and Purified Chicken Embryo Cell Vaccine (PCECV).[42,43]

However, our study has some limitations. Firstly, our study was a single-arm clinical trial and did not have a healthy control group to obtain information about the changes in antibody titers in HIV patients after rabies vaccination and the comparison of antibody titers in healthy individuals. Secondly, most of the enrolled HIV patients were male, which may be related to the current main transmitting population and transmission route of HIV. Finally, due to the small number of patients with CD4+ T cells <200 cells/μL, the immunogenicity among patients with severe immunodeficiencies administering this modified vaccination procedure still needs further substantiation. In the future, prospective studies with large samples are needed for further research and confirmation.

A limitation we would also like to point out is that only 1 female participant participated in our study, and the rest of the 34 patients were male, which may have had some impact on the results. The reason for this may be due to the epidemiological characteristics of the area (the main mode of HIV transmission in the treatment centers in the area is MSM), which has a low percentage of female HIV patients. In addition, female HIV patients show higher sensitivity during the recruitment phase and lower willingness to participate in clinical trials, which poses a considerable recruitment challenge. A study published in The Lancet Infectious Diseases revealed that when women and men receive the same vaccine, females exhibit stronger innate immune responses to vaccination than males. This disparity may be attributed to sex-based differences in innate immunity, adaptive immunity, sex steroids modulate immunity, and host genetic factors.[44] A prospective randomized single-blinded investigation demonstrated that healthy female participants aged 18–64 developed enhanced protective antibody production following influenza immunization. Notably, adult females exhibited comparable antibody titers with half-dose vaccinations to those achieved by male counterparts receiving full-dose regimens, revealing significant sex-based variations in immunogenic responses to antigen exposure.[45] Therefore, we believe that despite the lack of female patients within the study cohort, some previous studies have confirmed that women may respond more strongly to the vaccine after vaccination than men, with a lower probability of false positives in the study. We fully recognize the importance of gender balance and will improve this issue in future studies.

Finally, we only analyzed rabies antibody titers at day 45 in patients, and long-term such as 180 and 360 days were not analyzed, and we will continue to follow patients to collect rabies antibody titer information before further analysis in subsequent trials.

Conclusion

HIV patients injected with HDCV using a five-dose Essen regime with a doubling of the first dose achieved a well immune response

with a 100% seroconversion rate at day 14 and a favorable safety profile. There was no significant difference in antibody titers between HIV patients with CD4+ T-cell counts below and above 500 cells/μL after HDCV injection.

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Disclosure statement

No potential conflict of interest was reported by the author(s).

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Author contributions

CRediT: **Huanyu Wu:** Data curation, Project administration, Writing – original draft; **Wei Ye:** Data curation, Project administration, Writing – original draft; **Xiaoming Deng:** Formal analysis, Software; **Lili Guo:** Supervision; **Chao Chen:** Validation; **Hao Jiang:** Funding acquisition, Methodology, Writing – review & editing.

Notes on contributor

Hao Jiang, male, born in 1984–01, M.D., currently deputy director of the Department of Emergency Medicine of Nanjing Hospital affiliated with Nanjing University of Traditional Chinese Medicine, graduated from Nanjing University with a Ph.D. in Clinical Medicine in 2018, is engaged in research related to acute and critical illnesses and animal-induced injuries, with the main research direction of pulmonary fungal and viral infections, fever with thrombocytopenia syndrome, rabies, and tetanus prevention research. He has published more than 10 SCI papers in domestic and international journals as the first author, edited a monograph of Clinical Internal Medicine Research, presided over a project of the Jiangsu Provincial Health Commission, and organized several municipal continuing education classes on animal injuries and special infections.

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