# Evaluating the Safety and Immunogenicity of the rZIKV/D4D30-713 Live Attenuated Chimeric Zika Candidate Vaccine in Healthy Flavivirus-Naïve Adults


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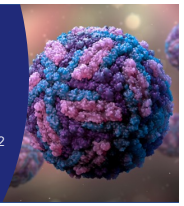


## BACKGROUND

- Zika virus (ZIKV) is a flavivirus associated with a serious congenital Zika syndrome, with a recent epidemic originating in Brazil in 2015.
- Novel ZIKV vaccine candidates are in development.
- We report results of a phase I study of the first live attenuated ZIKV vaccine for use in humans.
- rZIKV/D4D30-713 is a chimeric Zika vaccine, expressing the premembrane (prM) and envelope (E) genes of a contemporary ZIKV strain within a dengue DEN4Δ30 background.


- Chimerization has been used previously in flavivirus vaccine development, including:
  - Tetravalent dengue: yellow fever 17D with prM and E coding of DENV-1, DENV-2, DENV-3, DENV-4
  - Monovalent dengue: rDEN2/4D30, rDEN3/4D30
  - West Nile: rWN/DEN4D30

## METHODS

- We conducted a phase I, randomized, placebo-controlled double-blind trial, evaluating two different doses, $10^3$ PFU and $10^4$ PFU, of rZIKV/D4Δ30 in healthy adult subjects 18 – 50.
- Between October 2018 and August 2021, 28 subjects were enrolled in each dose cohort (20 to vaccine; 8 to placebo) at the Johns Hopkins Center for Immunization Research and the University of Vermont Vaccine Testing Center.
- Subjects were followed out to 26 weeks post-vaccination.
- Primary outcomes included safety and reactogenicity (adverse events graded by severity) and immunogenicity assessed by seroconversion by day 90 post-vaccination (ZIKV peak titer ≥ 1:10).
- The study was IRB approved (WCG 20181303, UVM CHRMS 18-0566).

**Table 1.** Characteristics of study population.

| | $10^3$ PFU (N=20) | Placebo for $10^3$ PFU group (N=8) | $10^4$ PFU (N=20) | Placebo for $10^4$ PFU group (N=8) |
|---|---|---|---|---|
| Age at vaccination, mean (years) N (stdev) | 29.75 (8.57) | 30.5 (9.99) | 36.1(6.97) | 35.88 (8.39) |
| Age range (years) N-N | (18-50) | (21-47) | (24-47) | (23-46) |
| Sex, male N (%) | 11 (55%) | 3 (37.5%) | 3 (15%) | 2 (25%) |
| Race, White N (%) | 14 (70%) | 5 (62.5%) | 14 (70%) | 6 (75%) |

## OBJECTIVES

- Safety and reactogenicity (adverse events graded by severity).
- Immunogenicity assessed by neutralizing antibody titers at 28, 56, and 90 days post-vaccination
- Seroconversion by day 90 post-vaccination (ZIKV peak titer ≥ 1:10).
- Recovery of infectious vaccine virus from blood/serum/urine/vaginal secretions/semen.
- Detection of vaccine virus by PCR from blood/serum/urine/vaginal secretions/semen

## RESULTS

**Table 2.** Adverse events following one dose of $10^3$ PFU ZIKV/DEN4Δ30 or placebo

| | | ZIKV/DEN4Δ30 (n=20) | Placebo (n=8) | P value (1-sided)[1] |
|---|---|---|---|---|
| Local | | | | |
| | Erythema | 5% | 0.0% | 0.7143 |
| | Tenderness | 0% | 0.0% | n/a |
| | Pain | 0% | 0.0% | n/a |
| | Pruritus | 0% | 0.0% | n/a |
| | Induration | 0% | 0.0% | n/a |
| Systemic | | | | |
| | Fever | 0% | 0% | n/a |
| | Headache | 50.0% | 62.5% | 0.8454 |
| | Rash | 0.0% | 0.0% | n/a |
| | Neutropenia[2] | 0.0% | 0.0% | n/a |
| | Elevated ALT | 0.0% | 0.0% | n/a |
| | Myalgia | 20.0% | 25.0% | 0.7922 |
| | Arthralgia | 10.0% | 12.5% | 0.8120 |
| | Retro-orbital Pain | 5.0% | 12.5% | 0.9259 |
| | Fatigue | 25.0% | 25.0% | 0.6940 |
| | Muscle weakness | 5.0% | 0.0% | 0.7143 |
| | Prolonged PT | 0.0% | 0.0% | n/a |
| | Prolonged PTT | 0.0% | 0.0% | n/a |
| | Thrombocytopenia | 0.0% | 0.0% | n/a |

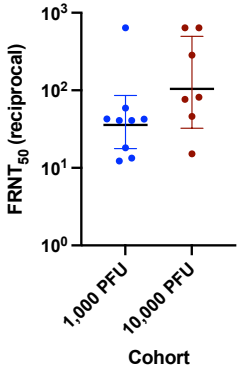
1. *P value is the probability is greater for rZIKV/DEN4Δ30 than placebo*
2. *Neutropenia was defined as an ANC ≤ 1,000/mm³.*

**Table 3.** Adverse events following one dose of $10^4$ PFU ZIKV/DEN4Δ30 or placebo

| | | ZIKV/DEN4Δ30 (n=20) | Placebo (n=8) | P value (1-sided)[1] |
|---|---|---|---|---|
| Local | | | | |
| | Erythema | 5% | 0.0% | 0.7143 |
| | Tenderness | 0% | 0.0% | n/a |
| | Pain | 0% | 0.0% | n/a |
| | Pruritus | 0% | 0.0% | n/a |
| | Induration | 0% | 0.0% | n/a |
| Systemic | | | | |
| | Fever | 5.0% | 0% | 0.714 |
| | Headache | 40.0% | 62.5% | 0.933 |
| | Rash | 0.0% | 0.0% | n/a |
| | Neutropenia[2] | 0.0% | 0.0% | n/a |
| | Elevated ALT | 0.0% | 0.0% | n/a |
| | Myalgia | 15.0% | 25.0% | 0.877 |
| | Arthralgia | 10.0% | 0.0% | 0.503 |
| | Retro-orbital Pain | 0.0% | 25.0% | 1.000 |
| | Fatigue | 20.0% | 25.0% | 0.792 |
| | Muscle weakness | 0.0% | 0.0% | n/a |
| | Non-purulent conjunctivitis | 0.0% | 0.0% | n/a |
| | Numbness | 5.0% | 0.0% | 0.714 |
| | Hyporeflexia | 5.0% | 0.0% | 0.714 |
| | Prolonged PT | 0.0% | 0.0% | n/a |
| | Prolonged PTT | 0.0% | 0.0% | n/a |
| | Thrombocytopenia | 0.0% | 0.0% | n/a |

1. *P value is the probability is greater for rZIKV/DEN4Δ30 than placebo*
2. *Neutropenia was defined as an ANC ≤ 1,000/mm³.*

**Figure 1.** Peak ZIKV FRNT titers in flavivirus-naive subjects who received $10^3$ PFU or $10^4$ PFU of ZIKV/DEN4Δ30.




*Line represents geometric mean of the peak titers, with error bars representing 95% confidence interval of the geometric mean titer. Only those who seroconverted are presented.*

- Fifty-five of 56 enrolled subjects completed the study.
  - One volunteer ($10^4$ PFU group) withdrew before study day 28 due to work conflicts.
- The most common adverse events were headache, fatigue, and myalgias.
  - These events did not occur significantly more frequently in vaccine recipients compared with placebo recipients.
  - One severe adverse event was deemed unrelated to study product (new breast cancer diagnosis).
- In the $10^3$ PFU group, 9 of 20 (45%) seroconverted.
- In the $10^4$ PFU group, 7 of 19 (37%) seroconverted.
- ZIKV could not be recovered by culture or PCR in the blood, saliva, or urine from any subject.

## CONCLUSIONS

- The vaccine appears to be safe and well-tolerated in healthy flavivirus-naïve adults.
- However, the vaccine was insufficiently immunogenic, with only around a 40% seroconversion and no recoverable virus in bodily fluids.
- The seroconversion proportion was similar between the two dosing groups, with higher peak titers in the $10^4$ PFU group.
- Given the lack of dose effect on seroconversion, further dose increases are unlikely to improve seroconversion.
- Chimerization can be highly attenuating to viruses.
- Our results suggest rZIKV/DEN4Δ30 is over-attenuated and thus will not be further developed as a candidate ZIKV vaccine.
- Additional Zika vaccine candidates will be studied.

## REFERENCES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