

**Neutralization of variant under investigation B.1.617 with sera of BBV152 vaccinees**

\*#<sup>1</sup>Pragya D. Yadav, Ph.D, <sup>1</sup>#Gajanan N. Sapkal, Ph.D, <sup>1</sup>Priya Abraham, M.D, Ph.D, <sup>2</sup>Raches Ella, M.S., <sup>1</sup>Gururaj Deshpande, Ph.D,<sup>1</sup> Deepak Y. Patil, Ph.D, <sup>1</sup>Dimpal A Nyayanit, Ph.D, <sup>3</sup>Nivedita Gupta, Ph.D., <sup>1</sup>Rima R. Sahay, M.D., <sup>1</sup>Anita M Shete, Ph.D, <sup>3</sup>Samiran Panda, M.D., Ph.D, <sup>3</sup>Balram Bhargava, D.M.,<sup>2</sup>V. Krishna Mohan,Ph.D.

#Equal first author

<sup>1</sup>Indian Council of Medical Research-National Institute of Virology, Pune, Maharashtra, India, Pin-411021

<sup>2</sup>Bharat Biotech International Limited, Genome Valley, Hyderabad, Telangana, India Pin-500 078

<sup>3</sup>Indian Council of Medical Research, V. Ramalingaswami Bhawan, P.O. Box No. 4911, Ansari Nagar, New Delhi, India Pin-110029

**\*Corresponding author**

Dr. Pragya D. Yadav,

Scientist 'E' and Group Leader,

Maximum Containment Facility,

Indian Council of Medical Research-National Institute of Virology,

Sus Road, Pashan, Pune, Maharashtra, India Pin-411021.

Phone: +9120-26006111, Fax No. 91-20-26122669

Email: [hellopragya22@gmail.com](mailto:hellopragya22@gmail.com)

Dear Editor,

The emergence of SARS-CoV-2 variants in places where the virus is uncontained poses a global threat from the perspective of public health and vaccine efficacy. Peng et al. recently reported the increased transmissibility with the newly emerged VOC (20C/S:452R and 20C/S:452R) with the L452R mutation in San Francisco [1]. We report the immunological characteristics of a VUI B.1.617, playing a critical role in the current surge of COVID-19 in the western state of Maharashtra, India.

Several SARS-CoV-2 variants B.1.1.7, B.1.351 and B.1.1.28.1 have been reported in India during year 2021[2,3]. We had sequenced 146 nasopharyngeal/oropharyngeal swabs of COVID-19 cases [4]. Among these, 15 sequences had a combination of L452R and E484Q mutations which raised concern as both are found in the receptor-binding domain (RBD) of the spike protein. However, the combined effect of these mutations is still unknown.

A total of 23 non-synonymous changes were observed in the sequences. Out of which, seven conserved non-synonymous changes were found at spike protein (G142D, E154K, L452R, E484Q, D614G, P681R, Q1071H) along with other conserved mutations (Figure 1A). These sequences were classified as VUI B.1.617. Phylogenetic analysis revealed three distinct sub-clusters of B.1.617 lineages with mutations in spike first T95I; second H1101D and third V382L, V1175Y (Figure 1 B). So far 21 countries have reported the presence of B.1.617 variants [5]. Virus isolation was attempted from fifteen specimens using Vero-CCL-81 cells [6]. Twelve specimens displayed cytopathic effects on the 4th post-infection day which further passaged and tittered for performing plaque reduction neutralization test (PRNT) [7]. These isolates were obtained, from clinical specimens of asymptomatic individuals (age range: 14-55 years) and cases with a low-grade fever, cough, and sore throat (age range: 26-77 years).

The neutralization efficacy of the VUI B.1.617 variant was compared with B1 (D614G) and B.1.1.7 variant using sera of 28 BBV152 vaccinated individuals, collected during the phase II clinical trial [8]. D614G vs. B.1.617 GMT ratio was 1.95, (95% CI:1.60-2.38 and p-value <0.0001). Similarly, the GMT ratio comparison of B.1.1.7 was significantly higher than the GMT for B.1.617 (GMT ratio 1.84, 95% CI: 1.50-2.27, p value< 0.0001) (Figure 1C and 1D). The comparison of D614G and B.1.1.7 showed equivalent responses with a GMT ratio of 1.06 and 95% CI (1.02-1.10).

Sera samples collected from COVID-19 recovered individuals (n=17) infected with lineage B.1.1.7, B.1.351, B.1.1.28.2, and B1 were used to perform PRNT50 against B.1.617 variants and the results were compared with the vaccine recipients' sera samples. The GMT values for vaccine recipients were 88.48 (95% CI: 62.02-126.2) and for recovered cases 86.85 (95% CI 52.04-144.9). The sera of BNT162b2 vaccinees which effectively neutralized B.1.1.7 and P.1 variants; was reduced with B.1.351 variant [9]. The B.1.617 variant performance with vaccine sera was better than recovered cases. The result of B.1.1.7 variant neutralization with BBV152 vaccine sera and findings of B.1.617 emphasize that this vaccine is robust against emerging mutation and maintains the efficacy of the vaccine (Figure 1 E) [10]. Assessing the clinical efficacy of BBV152 against such variants is underway.

## **Author Contributions**

PDY, GNS and PA contributed to study design, data collection, data analysis, interpretation and writing and critical review. RE, RRS, AMS, DYP, DAN and GRD contributed to data collection, interpretation, writing and critical review. NG, SP, VKM, and BB contributed to the critical review and finalization of the paper.

## **Acknowledgement**

Authors gratefully acknowledge the staff of ICMR-NIV, Pune including Mr. Prasad Sarkale, Mr. Shrikant Baradkar, Dr. Abhinendra Kumar, Mrs. Triparna Majumdar, Mrs. Savita Patil, Mr. Rajen Lakra, Mr. Vishwajeet Dhanure, Ms. Pranita Gawande, Mrs. Kaumudi Kalele, Mrs. Ashwini Waghmare, Ms. Jyoti Yemul, Ms. Manisha Dudhmal, Ms. Aasha Salunkhe and Mr. Chetan Patil for extending excellent technical support. The study was approved by the Institutional Biosafety Committee and Institutional Human Ethics Committee of ICMR-NIV, Pune, India. under project 'Molecular epidemiological analysis of SARS-COV-2 circulating in different regions of India' (20-3-18N).

## **Financial support & sponsorship**

Financial support was provided by the Indian Council of Medical Research (ICMR), New Delhi at ICMR-National Institute of Virology, Pune under intramural funding 'COVID-19'.

## **Conflicts of Interest**

Authors do not have a conflict of interest.

## References

1. Peng J, Sabrina A, Mitchell AM, et al. Estimation of secondary household attack rates for emergent SARS-CoV-2 variants detected by genomic surveillance at a community-based testing site in San Francisco. medRxiv. **2021**.
2. Yadav PD, Nyayanit DA, Sahay RR, et al. Isolation and characterization of the new SARS-CoV-2 variant in travellers from the United Kingdom to India: VUI-202012/01 of the B.1.1.7 lineage. J Travel Med **2021**; 28.
3. Menon GI. The Novel Coronavirus Variants and India's Uncertain Future. The Wire Science. 2021; Available at: <https://science.thewire.in/health/sars-cov-2-variants-b117-b1617-india-second-wave-uncertain-future/>. Accessed 22 April 2021.
4. Yadav PD, Nyayanit DA, Shete AM, et al. Complete genome sequencing of Kaisodi virus isolated from ticks in India belonging to *Phlebovirus* genus, family *Phenuiviridae*. Ticks Tick Borne Dis **2019**; 10:23–33.
5. PANGO lineages. Available at: [https://cov-lineages.org/lineages/lineage\\_B.1.617.html](https://cov-lineages.org/lineages/lineage_B.1.617.html). Accessed 22 April 2021.
6. Sarkale P, Patil S, Yadav PD, et al. First isolation of SARS-CoV-2 from clinical samples in India. Indian J Med Res **2020**; 151:244–250.
7. Deshpande GR, Sapkal GN, Tilekar BN, et al. Neutralizing antibody responses to SARS-CoV-2 in COVID-19 patients. Indian J Med Res. **2020**; 152:82-7. Ella R, Reddy S, Jogdand H, et al. Safety and immunogenicity of an inactivated SARS-CoV-2 vaccine, BBV152: interim results from a double-blind, randomised,

multicentre, phase 2 trial, and 3-month follow-up of a double-blind, randomised phase 1 trial. *Lancet Infect Dis.* **2021**; 21(5): P637-646.

9. Liu Y, Liu J, Xia H, Zhang X, et al. Neutralizing activity of BNT162b2-elicited serum. *N Engl J Med.* **2021**.
10. Sapkal GN, Yadav PD, Ella R, et al. Inactivated COVID-19 vaccine BBV152/COVAXIN effectively neutralizes recently emerged B.1.1.7 variant of SARS-CoV-2. *J Travel Med.* **2021**. taab051, <https://doi.org/10.1093/jtm/taab051>

Accepted Manuscript

**Figure 1: Characteristics and neutralization of VUI B.1.617 variant:** **A)** The common nucleotide changes observed in the majority of the isolates and clinical sequences. **B)** A neighbor-joining tree was generated using a Tamura 3-parameter model with gamma distribution and a bootstrap replication of 1000 cycles. Isolates are marked in red color and sequences from foreign travelers marked in pink color. The representative sequences from other clades are represented as B.1.1.7 (red), B.1.617 (blue), B.1.351 (black), B.1.525 (purple) Brazil P2 (light green), and B1 (light blue). Individual spike mutations specific to the clusters are marked using the arrows. One of sequences (MCL-21-H-741) had E484K mutation, that lead to its distinct clustering, in B.1.617 lineage (blue) belonged to a traveler who had returned from UAE to India.

**C)** Scatter plot depicting the neutralizing response of the individual sera (n=28) vaccinated with BBV152 (Covaxin) collected during phase II clinical trial for the prototype B1 (D614G) (pink), B.1.1.7 (red), B.1.617 (blue). The red solid line indicates the geometric mean titer and the error bar depicts a 95% confidence interval. **D)** Neutralization of the matched-pair samples compared to prototype D614G (pink), B.1.1.7 (red) and B.1.617 (blue). Neutralization reduction by a factor of 1.95 and 1.8 was observed against the B.1.617 variant for B1 (D614G) and B.1.1.7 variant respectively. A reduction factor of 1.06 was observed between the B1 (D614G) and B.1.1.7 variant. A two-tailed pair-wise comparison was performed using the Wilcoxon matched-pairs signed-rank test with a p-value of 0.05. \*\*\*\*

represent p-value <0.001 and \*\*p value=0.0038,ns= non-significant p-value. **E)** Neutralization of the COVID-19 recovered cases sera (n=17) of B.1.1.7 (n=2), B.1.351 (n=2), B.1.1.28.2 (n=2) and B1 lineage (n=11) infected individuals PRNT50 values against B.1.167 variant were compared with vaccine recipient serum samples. A two-tailed pair-wise comparison was performed using the Mann-Whitney test with a p-value of 0.05.

Accepted Manuscript



Figure 1

