***Preliminary Report - Early release, subject to modification***

# Quantification of the neutralization resistance of the Omicron Variant of Concern.

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**Summary:** The reduction in neutralization of the newly-emerged SARS-CoV-2 B.1.1.529 (Omicron) variant of concern[1] is highly variable. Fold-reduction, relative to the pandemic founder, ranged from 1 to 23, with quartiles of 2.5, 5.5, and 11, measured by lentiviral pseudotype neutralization assay. This was estimated from two cohorts: 17 random recent blood donors in Stockholm, and a set of 17 previously-infected hospital workers with higher mean neutralizing antibody titers against the WuHu-1 founder variant than those observed in the blood donors. Almost all serum samples evaluated retained some neutralization activity against the Omicron variant. The First WHO International Standard (20/136) showed a ±40-fold reduction in the neutralization of Omicron ($IC_{50}$ from 0.6 IU/ml to 23.4 IU/ml). These preliminary data on the neutralization sensitivity of the Omicron variant require both internal and independent confirmation, and the clinical impact of natural and vaccine-induced immunity with respect to protection from infection and severe disease needs urgent investigation.

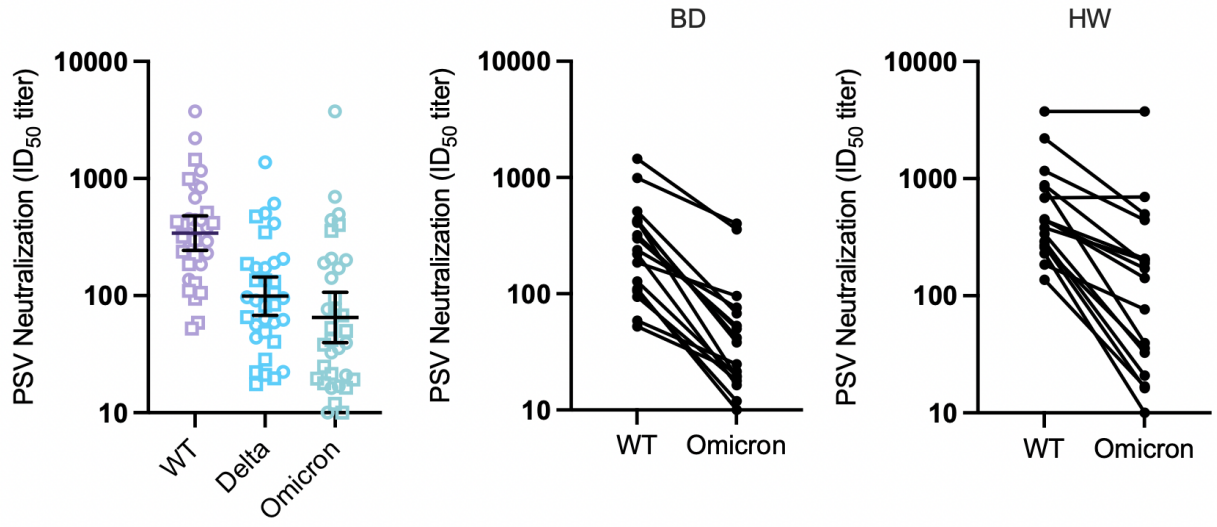


**Figure 1:** Pseudovirus neutralization titers for Blood Donors (BD; N=17) and Hospital Workers (HW; N=17) against the pandemic founder variant (WT), the B.1.617.2 variant (Delta) and the B.1.1.529 variant (Omicron).

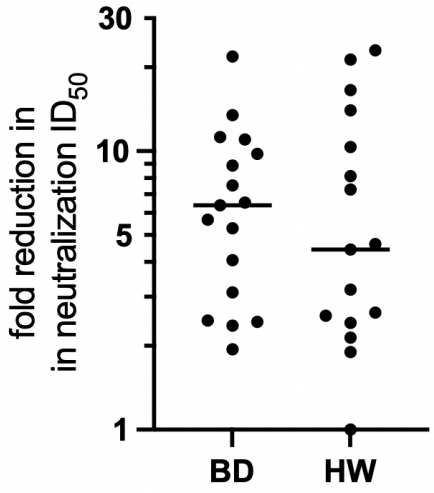


**Figure 2:** Fold reduction in neutralization of the Omicron variant compared to the pandemic founder variant for Blood Donors (BD, N=17) and Hospital Workers (HW, N=17) from Stockholm, Sweden.

**Donor sample description:** Two cohorts were studied. Cohort 1 comprised serum samples with detectable neutralization against the Wu-Hu-1 founder variant from 17 anonymized blood donors ("BD"), donated during week 48, 2021, in Stockholm, Sweden. Cohort 2 comprised 17 serum samples from Hospital Workers ("HW") at the Karolinska University Hospital in Stockholm, who were invited to participate in a study that aimed to characterize their antibody responses following SARS-CoV-2 infection and subsequent vaccinations. Participants were confirmed PCR positive in May 2020 and serum samples collected in November 2021 were analyzed.

**Assay details:** A region of spike (with codons corresponding to amino acid positions 43 to 1000) incorporating all of the Omicron variant reference mutations was amplified from cDNA derived from a later-confirmed B.1.1.529 clinical sample obtained from a set of anonymized early cases of suspected Omicron infections. The spike fragment was subcloned by Gibson Assembly into a codon-optimized SARS-CoV-2 Spike expression vector (in pcDNA3.1), harbouring a mutation that introduces a stop codon that truncates the last 19 amino acids of the cytoplasmic tail (facilitating efficient incorporation onto lentiviral particles). The spike-encoding expression vector was confirmed by sequencing to be identical, by amino acid, to the Omicron consensus. Omicron Spike-pseudotyped lentivirus particles were generated by the co-transfection of HEK293T cells with the Omicron variant spike plasmid, an HIV gag-pol packaging plasmid (Addgene #8455), and a lentiviral transfer plasmid encoding firefly luciferase (Addgene #170674). Neutralization was assessed in HEK293T-ACE2 cells, and all samples were run simultaneously.

**Statistical analysis:** Individual $ID_{50}$ values for each sample against each variant were calculated in GraphPad Prism v9 by fitting a four-parameter logistic curve to neutralization by serial 3-fold dilutions of serum, as described previously[2].

**Ethics:**
HW cohort: Informed consent was obtained from all participants as part of an ethics approval (Decision number 2020-01620) from the National Ethical Review Agency of Sweden. BD cohort and Omicron-positive samples were anonymized, and not subject to ethical approvals.

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1. Classification of Omicron (B.1.1.529): SARS-CoV-2 variant of Concern. https://www.who.int/news/item/26-11-2021-classification-of-omicron-(b.1.1.529)-sars-cov-2-variant-of-concern.
2. Sheward, D. J. *et al.* Beta RBD boost broadens antibody-mediated protection against SARS-CoV-2 variants in animal models. *Cell Rep Med* **2**, 100450 (2021).