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CORRESPONDENCE

Third-Dose BNT162b2 Vaccination Elicits Markedly High-Level SARS-CoV-2–Neutralizing Antibodies in Vaccinees Who Responded Poorly to a Second Dose in Japan

To the Editor—We read with great interest the article by Saciuk et al demonstrating that, in a retrospective cohort study in Israel, an additional dose of BNT162b2 vaccine 6 months after initial 2-dose vaccination bolsters protection against infection, with a vaccine effectiveness of 89%, as assessed during August–October 2021, when the majority of infections were due to the Delta variant [1]. Recent severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) studies using pseudovirus have shown that a third dose of BNT162b2 well elicits neutralizing antibodies against VOC (variants of concern) including Omicron (B.1.1.529, BA.1) [2–4]. However, Cheng et al very recently reported a single-source outbreak of Omicron BA.2 sublineage in Hong Kong, which indicates high transmissibility of Omicron/BA.2 [5] and poses further concerns on the efficacy of anti–SARS-CoV-2 vaccines.

In the present prospective study, enrolling 225 health care workers (see demographic characteristics in Supplementary Table 1), who received 3 doses of BNT162b2 in Japan, we consecutively determined SARS-CoV-2 neutralizing activity (50% neutralizing titer, NT_{50}) of their sera using VeroE6^{TMPRSS2} cells and its kinetics/profiles over 300 days following the first dose; this is a continuation of our previous study [6]. We also determined NT_{50} of selected sera using VeroE6^{TMPRSS2} and HeLa$^{hACE2\text{-}TMPRSS2}$ cells against infectious VOC, including Delta and Omicrons (BA.1 and BA.2), whose emergence has been associated with a steep increase in coronavirus disease 2019 (COVID-19) cases and hospitalizations (experimental details are provided in the "Methods" section of Supplementary Materials).

There was significant neutralizing activity on day 28 after the first dose (NT_{50} = 501, 1 week after the second dose), while there was a continual decrease until day 280. NT_{50} values further decreased to 51 by day 280 when approximately 85% of the participants had NT_{50} values of <100 and approximately 36% had less than 20 NT_{50} or undetectable (Supplementary Figure 1). However, 2 weeks after administration of the third dose (205 participants [91.1%] remained in the cohort on day 300), there was a substantial rise in neutralizing activity, achieving an average NT_{50} of 3531. There was a concern that individuals who poorly responded to the second dose might again fail to produce sufficient neutralizing antibodies. Therefore, we specifically determined neutralization activity in vaccinees, who had achieved the lowest 10% level of neutralization following the second dose (n = 22, average-NT_{50} = 110 on day 28; inset in Supplementary Figure 1). Notably, by day 300, all these low responders achieved markedly greater levels of neutralizing activity with an average NT_{50} of 2341 (range 482–9113 in VeroE6^{TMPRSS2}; Table 1). In HeLa$^{hACE2+TMPRSS2}$ cells, sera from these low responders substantially neutralized SARS-CoV-2$^{05\text{-}2N}$, Alpha, Beta, Gamma, and Delta (geometric mean [gMean] NT_{50} = 1777, 1350, 480, 1015, and 959, respectively), but had only marginal activity against Omicron/BA.1 with a gMean-NT_{50} of 52 (range ≤ 20–197; Table 1). The same sera had similar neutralization profiles in VeroE6^{TMPRSS2} cells. On the other hand, sera from participants who achieved the highest 10% level of neutralization on day 300 (n = 22, average NT_{50} = 10 885 in VeroE6^{TMPRSS2}) had high neutralizing activity against SARS-CoV-2$^{05\text{-}2N}$, Alpha, Beta, Gamma, and Delta (gMean NT_{50} = 9774, 4906, 2279, 3271, and 3377, respectively) in HeLa$^{hACE2+TMPRSS2}$ cells and good neutralizing activity against Omicron/BA.1 (gMean NT_{50} = 500, range 171–979). Notably, all day 280 sera of the highest 10% of participants failed to neutralize both Omicron/BA.1 and BA.2 (NT_{50} values ≤20; Supplementary Figure 2); however, these participants' sera on day 300 also neutralized Omicron/BA.2 well (gMean-NT_{50} = 702, range 262–1653; Table 1).

The present data clearly show that a third dose of BNT162b2 elicits high-level SARS-CoV-2-neutralizing antibodies even in those who poorly responded to the second dose, although low responders to the vaccines may be vulnerable to infection with Omicron sublineages BA.1 and BA.2. Of note, however, in terms of the effectiveness of a third dose of BNT162b2 against Omicron sublineages, the morbidity and mortality have yet to be determined between individuals who received the third dose but contracted symptomatic Omicron-related COVID-19 and those not receiving the third dose who contracted symptomatic Omicron-related COVID-19.

Supplementary Data

Supplementary materials are available at *The Journal of Infectious Diseases* online (http://jid.oxfordjournals.org/). Supplementary materials consist of data provided by the author that are published to benefit the reader. The posted materials are not copyedited. The contents of all supplementary data are the sole responsibility of the authors. Questions or messages regarding errors should be addressed to the author.

Notes

Author contributions. M. A. and H. M. had access to all data in this study and hold all responsibility for the integrity of the data and the accuracy of the data analysis. M. A. and H. M. contributed the

Table 1. Neutralization Activity of Highest and Lowest 10% of Responders' Sera against SARS-CoV-2s, Including the Wuhan Strain and 6 VOC

	SARS-CoV-$2^{05\text{-}2N}$, Wuhan	SARS-CoV-2^{QK002}, Alpha	SARS-CoV-$2^{TY8\text{-}612}$, Beta	SARS-CoV-$2^{TY7\text{-}501}$, Gamma	SARS-CoV-2^{1734}, Delta	SARS-CoV-$2^{NCGM\text{-}929\text{-}1N}$, Omicron/BA.1	SARS-CoV-2^{2037}, Omicron/BA.2, in VeroE6^{TMPRSS2}[a]
gMean NT_{50} of highest 10% of sera in HeLa$^{hACE2+TMPRSS2}$ cells	9774 (3745–27 921)	4906 (1926–11 018)	2279 (844–8996)	3271 (1723–8849)	3377 (1157–8053)	500 (171–979)	702 (262–1653)
gMean NT_{50} of lowest 10% of sera in							
VeroE6^{TMPRSS2} cells	1654 (482–9113)	1544 (458–4335)	483 (114–3486)	928 (278–2877)	1014 (371–4246)	130 (≤20–487)	115 (≤20–649)
HeLa$^{hACE2+TMPRSS2}$ cells	1777 (410–7608)	1350 (429–4337)	480 (96–2861)	1015 (528–2410)	959 (386–5905)	52 (≤20–197)	ND

gMean NT_{50} titers of day 300 sera of lowest 10% of responders (n = 22) and highest 10% of responders (n = 22) against SARS-CoV-$2^{05\text{-}2N}$ and 6 VOC were determined in cell-based assays using each SARS-CoV-2 strain and VeroE6^{TMPRSS2} cells or HeLa$^{hACE2+TMPRSS2}$ cells. The numbers in parentheses denote ranges of NT_{50} values determined for each strain.

Abbreviations: gMean, geometric mean; ND, not determined; NT_{50}, 50% neutralizing titer; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; VOC, variant of concern.

[a]Only VeroE6^{TMPRSS2} cells were used for SARS-CoV-2^{2037} (Omicron/BA.2) because SARS-CoV-2^{2037} did not propagate well in HeLa$^{hACE2+TMPRSS2}$ cells.

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