Short Communication

Safety and antibody response of recipients who unexpectedly received undiluted prime dose of BNT162b2 COVID-19 vaccine in Taiwan

Wang-Huei Sheng [a,b], Chung-Cheng Wang [c,d], Ching-Chi Chu [e], Yueh-Juh Lin [e], Sui-Yuan Chang [f,g], Si-Man Ieong [f], Shan-Chwen Chang [a,b,*]

[a] Department of Internal Medicine, National Taiwan University Hospital, Taipei City, Taiwan
[b] School of Medicine, National Taiwan University College of Medicine, Taipei City, Taiwan
[c] Department of Urology, En Chu Kong Hospital, New Taipei City, Taiwan
[d] Department of Biomedical Engineering, Chung Yuan Christian University, Chungli, Taiwan
[e] Division of Cardiology, Department of Internal Medicine, En Chu Kong Hospital, New Taipei City, Taiwan
[f] Department of Clinical Laboratory Sciences and Medical Biotechnology, National Taiwan University College of Medicine, Taipei, Taiwan
[g] Department of Laboratory Medicine, National Taiwan University Hospital, Taipei City, Taiwan

We reported 25 recipients (14 females and 11 males) aged from 18 to 65 years who unexpectedly received a primary dose of undiluted BNT162b2 vaccine (180 μg). The most common adverse reactions included injection site pain (n = 22), followed by fever (9), fatigue (8), chest tightness (6), and dizziness (6). The most common laboratory abnormalities were anemia (n = 4) and elevated liver transaminase level (4), followed by abnormal leukocyte counts (3) and elevated D-dimer level (3). The adverse reactions and laboratory abnormalities of these recipients were mild and spontaneously recovered within a few weeks. Significant elevations of anti-SARS-CoV-2 spike IgG titers after a booster dose of the BNT162b2 were found. Similar to reports of BNT162b2 clinical trials, the adverse reactions and laboratory abnormalities of these recipients were mild, and they spontaneously recovered within a few weeks. These results provide clinical and immunological effects of undiluted BNT162b2 vaccine inoculation.

* Corresponding author. Department of Internal Medicine, National Taiwan University Hospital, 7, Chung Shan South Road, Taipei City, 10002, Taiwan. Fax: +886 2 23224793.
E-mail address: changsc@ntu.edu.tw (S.-C. Chang).

Please cite this article as: W.-H. Sheng, C.-C. Wang, C.-C. Chu et al., Safety and antibody response of recipients who unexpectedly received undiluted prime dose of BNT162b2 COVID-19 vaccine in Taiwan, Journal of the Formosan Medical Association, https://doi.org/10.1016/j.jfma.2022.07.009

Introduction

The BNT162b2 (BioNTech/Pfizer) mRNA vaccine was the first coronavirus disease 2019 (COVID-19) vaccine approved under emergency use authorization by the US Food and Drug Administration in individuals ≥16 years old.[1] It is a multi-dose vial administered intramuscularly after dilution as a series of two doses (30 μg in 0.3 mL each) at least 21 days apart.[2] Each vial contains six doses of 30 μg of tozinameran, a BNT162b2 RNA embedded in lipid nanoparticles, and the vaccine (0.45 mL) should be diluted with 1.8 mL of 0.9% sodium chloride solution use.[2]

Vogel et al. reported the preclinical development of BNT162b1 and BNT162b2 vaccines with different titrated doses.[3] They found that both vaccines could elicit a dose-dependent antibody response and the BNT162b2 appears to be more efficient than the BNT162b1.[3] Walsh et al. reported a Phase I clinical trial for BNT162b1 and BNT162b2 and evaluated the safety and immunogenicity between different doses of BNT162b1 and BNT162b2 vaccines in adults.[4] They found adverse reactions of the first dose were usually mild-to-moderate and primarily occurred within 7 days after injection. The most common reactions were local injection site pain, fatigue, chills, and fever.[4]

The mRNA vaccine-associated myocarditis is considered to be the most potentially serious adverse event.[5] In a nationwide vaccination campaign surveillance from Israeli, the overall risk of myocarditis between the first and second doses was 1.76 per 100,000 persons.[5] Symptoms of myocarditis usually developed within a few days (<7 days) after the vaccination.

Overdose with the BNT162b2 vaccine injections has been reported in Australia, Germany, Italy, and the United States.[6] In this study, we reported 25 recipients who erroneously received undiluted BNT162b2 vaccine and adverse events and laboratory abnormalities were prospectively monitered.

Materials and methods

On September 27, 2021, 25 people (14 females and 11 males) accidently received an injection of an undiluted multi-dose vial of BNT162b2 vaccine (180 μg) each person. This was identified later in the same day, and all of the recipients were informed immediately. It was suggested that they be admitted to the hospital for monitoring and clinical care. All recipients were closely monitored daily for 1 week (Days 2–7) and then by outpatient clinic visits at the 2nd, 4th, and 6th week. Physical examination, venous blood for biochemical tests and anti-SARS-CoV-2 spike antibody, cardiopulmonary function tests including cardiac enzymes, electrocardiography (ECG), chest X-ray examination, and cardiac echography were provided at each visit. Voluntary second doses of the BNT162b2 vaccine inoculation were provided for all 25 people 2 months after the first dose. This study had been approved by the Institutional Review Boards (Ethics Committee) of En Chu Kong Hospital, New Taipei City, Taiwan (ECK-IRB 1101102).

All recipients were instructed to report any suspected adverse reactions, such as local (erythema, swelling, heat, and pain over the inoculated site) and systemic symptoms (fever, chill, myalgia, arthralgia, chest tightness, chest pain, palpitations, shortness of breath, exercise intolerance, and fainting) from the first day after the undiluted prime dosing of the BNT162b2 vaccine. A standard form questionnaire was designed to evaluate the adverse reactions of vaccination according to WHO guidelines.[7]

An anti-SARS-CoV-2 spike in IgG was determined using an Abbott SARS-CoV-2 IgG II Quant assay (06S60, Abbott, USA). Results were reported as arbitrary units (AU) per milliliter, and the cutoff value was 50.0 AU/mL. The mathematical relationship of the Abbott AU/mL unit to the WHO unit (binding antibody unit per mL [BAU/mL]) followed the equation: $BAU/mL = 0.142{*}AU/mL$.

Results

The underlying diseases of the 25 recipients included hypertension ($n = 7$), coronary artery disease (2), hyperlipidemia (2), thalassemia (1), ankylosing spondylitis (1), iron-deficiency anemia (1), uterine myoma (1), reflux esophagitis (1), hyperthyroidism (1), and chronic hepatitis B (1). The most common adverse reactions included injection site pain ($n = 22$), followed by fever (9), fatigue (8), chest tightness (6), dizziness (6), myalgia (5), and palpitations (5) (Table 1). The median duration from symptom onset to recovery was 3–6 days and the symptoms of 21 recipients (84%) were resolved within 7 days. Most symptoms were mild to moderate and none of the recipients reported severe adverse reactions.

The most common laboratory abnormalities were anemia ($n = 4$) and elevated liver transaminase level (4), followed by abnormal leukocyte counts (3) and elevated D-dimer level (3) (Table 2). Two recipients had a persistent level of anemia due to underlying iron-deficiency anemia in one and thalassemia in the other. One recipient had elevated liver transaminase levels due to underlying chronic hepatitis B. All the laboratory abnormalities were transient and most were clinically asymptomatic. For all recipients, there were no significant changes of left ventricular ejection fraction (LVEF) individually and cardiac echography of each recipient was followed up by the same cardiologist. The median LVEF was 69.0% (range, 67.5%–78.2%).

The anti-SARS-CoV-2 spike IgG increased at the 2nd week (median, 11.26 BAU/mL; interquartile range [IQR], 1.37–16.39 BAU/mL; at Days 9–15) from the baseline (0.36 BAU/mL; IQR, 0.15–0.76 BAU/mL; at Days 2–4). Compared

Table 1 Adverse reactions reported after the administration of undiluted prime dose at five scheduled time points. The number indicate the case number with symptoms.

Symptoms, (n)	Visit 1	Visit 2	Visit 3	Visit 4	Visit 5
	Day 2–4 (n = 25)	Day 5–7 (n = 25)	Day 9–15 (n = 25)	Day 24–37 (n = 25)	Day 40–52 (n = 25)
Local reactions					
Injection site pain	22	10	2	0	0
Erythema and swelling	2	1	0	0	0
Systemic reactions					
Fever	9	5	0	0	0
Fatigue	8	5	0	0	0
Chest tightness	6	3	2	0	0
Dizziness	6	2	2	0	0
Myalgia	5	2	0	0	0
Palpitation	5	2	1	0	0
Dyspnea	3	1	1	0	0
Headache	3	1	1	0	0
Sweats	2	1	1	0	0
Skin rashes	2	1	0	0	0
Arthralgia and arthritis	1	1	0	0	0
Chills	1	0	0	0	0
Cough	1	1	0	0	0
Diarrhea	1	0	0	0	0
Numbness	1	1	1	0	0
Nausea and vomits	1	0	0	0	0

with the anti-SARS-CoV-2 spike IgG at the 2nd week, there was significant elevation in the anti-SARS-CoV-2 spike IgG level of 20-fold at the 4th week (232.04 BAU/mL; IQR, 97.94–357.29 BAU/mL; at Days 24–37). Six recipients received a second dose of the BNT162b2 vaccine with a median interval of 66 days (range 60–76 days) between the first and second doses of the BNT162b2 vaccine inoculation. The anti-SARS-CoV-2 spike IgG significantly increase after a booster dose of the BNT162b2 (450.98 BAU/mL; IQR, 340.67–662.00 BAU/mL; at Day 86–104).

Table 2 Results of blood biochemistry, chest plain films and cardiac echography after the administration of undiluted prime dose at five scheduled time points. The number indicate the abnormality case number.

	Visit 1	Visit 2	Visit 3	Visit 4	Visit 5
	Day 2–4 (n = 25)	Day 5–7 (n = 17)	Day 9–15 (n = 21)	Day 24–37 (n = 16)	Day 40–52 (n = 10)
Anemia	4	3	3	3	2
Elevated liver transaminase	4	2	2	1	1
Elevated D-dimer	3	3	3	2	0
Abnormal blood leukocyte count	3	2	1	0	0
Elevated CPK	1	1	0	0	0
Elevated serum creatinine	1	1	0	0	0
Decrease blood platelet count	0	0	0	0	0
Elevated CPK-MB	0	0	0	0	0
Elevated troponin-I	0	0	0	0	0
Abnormal findings of electrocardiography	3	3	2	2	2
Abnormal findings of chest plain film	1	1	1	1	1
Abnormal findings of cardiac echo	1	1	1	1	1

CPK, creatine phosphokinase.

Discussion

Our observational cohort provides information about the experience and management of recipients with undiluted BNT162b2 dosing. The undiluted BNT162b2 vaccine was associated with adverse reactions that were mild to moderate and spontaneously subsided within 2 weeks. The recipients inoculated with the undiluted BNT162b2 vaccine dose had a trend of more slowly antibody decay and prompt immunological response to the second dose, even administration 2 months after the first dose. Compatible with other studies,[4,8–10] we confirmed a favorable safety profile for the BNT162b2 COVID-19 vaccine.

High dose inoculation of the BNT162b2 vaccine was reported in animal experiments and clinical trials.[3,8] In an experimental rhesus macaques model receiving intramuscular 30 or 100 μg doses of the BNT162b2 vaccine, anti-SARS-CoV-2 RBD-binding IgG was detectable by Day 14 after the initial dosing, and antibody titers markedly increased 7 days after second dose.[3] Similar immunological responses were found between the two different dose groups. Antibody titers rapidly declined in the 30-μg dose BNT162b2 group compared to the 100-μg dose group.[3] In a Phase I clinical trial evaluating the immunogenicity of different BNT162b2 vaccine doses (10 μg, 20 μg, 30 μg) among adults, the geometric mean RBD-binding IgG were similar in the 30-μg and 100-μg groups and higher than in the 10-μg group.[8] Our results are compatible with these reports,[3,8] and a sustained antibody response was found even before the inoculation with the second dose 2 months later.

The adverse reactions and abnormalities of laboratory tests of this cohort were mild to moderate in severity and all resolved within 2 weeks. In the clinical trial evaluating the safety of BNT162b1 and BNT162b2 vaccine doses among adults, they found a similar incidence of adverse reactions after vaccination among 10-μg, 30-μg, and 100-μg groups.[8] However, the severity of adverse events was dose dependent. For example, pain at the injection site (58.3% of 10-μg vaccine group versus 100% in each of the 30-μg and 100-μg vaccine groups), fever $\geq$38.0 °C (8.3%, 8.3%, and 50.0% of 10-μg, 30-μg, and 100-μg groups, respectively).[8] Our observational cohort seems to have a relative lower incidence of adverse events. The participants in the present study had similar incidences of laboratory abnormalities with these clinical trials.[4,8,10,11]

This study had limitations. Although the study based on a prospective observational design, treatment was decided by recipients at their convenience. The time of visits may not have been performed on the same timeline. In addition, only 6 recipients received second dose of BNT162b2 vaccine inoculation. Therefore, the appropriate interval for second dose after undiluted prime dose are still not unknown. Of the six recipients who received the second dose, none had myocarditis. These results might support the recommendation of delayed inoculation by the US CDC.[12]

In conclusion, despite the limited number of cases, this report provides the clinical and immunological effects of undiluted BNT162b2 vaccine inoculation. The findings of this observational cohort supported the safety and efficacy of the BNT162b2 vaccine even with a larger dose. This report also provides valuable information for management of vaccine error.

Funding support

Declaration of competing interest

The authors have no conflicts of interest relevant to this article.

References

1. The U.S. Food and Drug Administration. *FDA approves first COVID-19 vaccine*. https://www.fda.gov/emergency-preparedness-and-response/coronavirus-disease-2019-covid-19/comirnaty-and-pfizer-biontech-covid-19-vaccine. [Accessed 10 March 2022].
2. The U. S. Food and Drug Administration. *Pfizer-BioNTech fact sheets (English) and FAQs*. https://www.fda.gov/emergency-preparedness-and-response/coronavirus-disease-2019-covid-19/comirnaty-and-pfizer-biontech-covid-19-vaccine#additional. [Accessed 10 March 2022].
3. Vogel AB, Kanevsky I, Che Y, Swanson KA, Muik A, Vormehr M, et al. BNT162b vaccines protect rhesus macaques from SARS-CoV-2. *Nature* 2021;**592**:283–9.
4. Walsh EE, Frenck RW, Falsey AR, Kitchin N, Absalon J, Gurtman A, et al. Safety and immunogenicity of two RNA-Based Covid-19 vaccine candidates. *N Engl J Med* 2020;**383**:2439–50.
5. Mevorach D, Anis E, Cedar N, Bromberg M, Haas EJ, Nadir E, et al. Myocarditis after BNT162b2 mRNA vaccine against Covid-19 in Israel. *N Engl J Med* 2021;**385**:2140–9.
6. Raposoa F, Lippib G. Antibody response induced by the boost overdose during COVID-19 heterologous prime-boost vaccination strategy. *Clin Chim Acta* 2021;**523**:201–4.
7. *WHO Guidelines on clinical evaluation of vaccines: regulatory expectations*. https://www.who.int/biologicals/BS2287_Clinical_guidelines_final_LINE_NOs_20_July_2016.pdf. [Accessed 10 May 2021].
8. Mulligan MJ, Lyke KE, Kitchin N, Absalon J, Gurtman A, Lockhart S, et al. Phase 1/2 study of COVID-19 RNA vaccine BNT162b1 in adults. *Nature* 2020;**586**:589–93.
9. Dagan N, Barda N, Balicer RD. Adverse effects after BNT162b2 vaccine and SARS-CoV-2 infection, according to age and sex. *N Engl J Med* 2021;**385**:2299.
10. Polack FP, Thomas SJ, Kitchin N, Absalon J, Gurtman A, Lockhart S, et al. Safety and efficacy of the BNT162b2 mRNA Covid-19 vaccine. *N Engl J Med* 2020;**383**:2603–15.
11. Frenck Jr RW, Klein NP, Kitchin N, Gurtman A, Absalon J, Lockhart S, et al. Safety, immunogenicity, and efficacy of the BNT162b2 Covid-19 vaccine in adolescents. *N Engl J Med* 2021;**385**:239–50.
12. Centers for Disease Control and Prevention. *COVID-19 vaccine administration errors and deviations*. https://www.cdc.gov/vaccines/covid-19/downloads/covid19-vaccine-errors-deviations.pdf. [Accessed 30 March 2022].