BMJ Open

BMJ Open is committed to open peer review. As part of this commitment we make the peer review history of every article we publish publicly available.

When an article is published we post the peer reviewers' comments and the authors' responses online. We also post the versions of the paper that were used during peer review. These are the versions that the peer review comments apply to.

The versions of the paper that follow are the versions that were submitted during the peer review process. They are not the versions of record or the final published versions. They should not be cited or distributed as the published version of this manuscript.

BMJ Open is an open access journal and the full, final, typeset and author-corrected version of record of the manuscript is available on our site with no access controls, subscription charges or pay-per-view fees (http://bmjopen.bmj.com).

If you have any questions on BMJ Open's open peer review process please email info.bmjopen@bmj.com

BMJ Open: first published as 10.1136/bmjopen-2021-055611 on 11 October 2021. Downloaded from http://bmjopen.bmj.com/ on April 9, 2024 by guest. Protected by copyright.

BMJ Open

Booster dose of mRNA SARS-CoV-2 vaccine for kidney transplant recipients without adequate humoral response with or without Immunosuppression reduction – protocol for a randomised controlled trial (BECAME study)

Journal:	*BMJ Open*
Manuscript ID	bmjopen-2021-055611
Article Type:	Protocol
Date Submitted by the Author:	17-Jul-2021
Complete List of Authors:	Yahav, Dafna; Rabin Medical Center, Rozen-Zvi, Benaya; Rabin Medical Center, Mashraki, Tiki; Rabin Medical Center, Department of Nephrology and Hypertension Atamna, Alaa; Tel Aviv University, Sackler school of medicine; Rabin Medical Center, Infectious diseases unit Ben-Zvi, Haim ; Rabin Medical Center, Department of Medicine E, Beilinson Hospital Bar-Haim, Erez; Israel Institute for Biological Research, Department of Biochemistry and Molecular Genetics; IIBR Rahamimov, R.; Rabin Medical Center
Keywords:	Renal transplantation < NEPHROLOGY, COVID-19, IMMUNOLOGY

SCHOLARONE™
Manuscripts

BMJ Open: first published as 10.1136/bmjopen-2021-055611 on 11 October 2021. Downloaded from http://bmjopen.bmj.com/ on April 9, 2024 by guest. Protected by copyright.

I, the Submitting Author has the right to grant and does grant on behalf of all authors of the Work (as defined in the below author licence), an exclusive licence and/or a non-exclusive licence for contributions from authors who are: i) UK Crown employees; ii) where BMJ has agreed a CC-BY licence shall apply, and/or iii) in accordance with the terms applicable for US Federal Government officers or employees acting as part of their official duties; on a worldwide, perpetual, irrevocable, royalty-free basis to BMJ Publishing Group Ltd ("BMJ") its licensees and where the relevant Journal is co-owned by BMJ to the co-owners of the Journal, to publish the Work in this journal and any other BMJ products and to exploit all rights, as set out in our licence.

The Submitting Author accepts and understands that any supply made under these terms is made by BMJ to the Submitting Author unless you are acting as an employee on behalf of your employer or a postgraduate student of an affiliated institution which is paying any applicable article publishing charge ("APC") for Open Access articles. Where the Submitting Author wishes to make the Work available on an Open Access basis (and intends to pay the relevant APC), the terms of reuse of such Open Access shall be governed by a Creative Commons licence – details of these licences and which Creative Commons licence will apply to this Work are set out in our licence referred to above.

Other than as permitted in any relevant BMJ Author's Self Archiving Policies, I confirm this Work has not been accepted for publication elsewhere, is not being considered for publication elsewhere and does not duplicate material already published. I confirm all authors consent to publication of this Work and authorise the granting of this licence.

For peer review only

Booster dose of mRNA SARS-CoV-2 vaccine for kidney transplant recipients without adequate humoral response with or without Immunosuppression reduction – protocol for a randomised controlled trial (BECAME study)

Dafna Yahav[1,2*], Benaya Rozen-Zvi[2,3*], Tiki Mashraki[3,4], Alaa Atamna[1,2], Haim Ben-Zvi[2,5], Erez Bar-Haim[6], Ruth Rahamimov[2,3,4]

[1] Infectious Diseases Unit, Rabin Medical Center, Beilinson Campus, Petah-Tikva, Israel

[2] Sackler Faculty of Medicine, Tel Aviv University, Tel Aviv, Israel

[3] Department of Nephrology and Hypertension, Rabin Medical Center, Beilinson Campus, Petah-Tikva, Israel

[4] Department of Transplantation, Rabin Medical Center, Beilinson Campus, Petah-Tikva, Israel

[5] Clinical Microbiology Laboratory, Rabin Medical Center, Beilinson Hospital, Petah-Tikva, Israel

[6] Department of Biochemistry and Molecular Genetics, Israel Institute for Biological Research, 74100 Ness-Ziona, Israel.

[*] BRZ and DY contributed equally for this work

Corresponding author: Prof. Dafna Yahav, Infectious Diseases Unit, Rabin Medical Center, Beilinson Hospital, 39 Jabotinsky Road, Petah-Tikva, 49100, Israel. Phone: 972-3-9377512, Fax: 972-3-9376512. E-mail: dafna.yahav@gmail.com

Keywords: kidney transplant recipients; COVID-19; vaccine; immunosuppression reduction; randomized controlled trial

Word count: 1936

For peer review only

Abstract

Introduction: Inadequate antibody response to mRNA SARS-CoV-2 vaccination has been described among kidney transplant recipients. Immunosuppression level and specifically, use of antimetabolite in the maintenance immunosuppressive regimen, are associated with inadequate response. In light of the severe consequences of COVID-19 in solid organ transplant recipients, we believe it is justified to examine new vaccination strategies in these patients.

Methods and analysis: BECAME is a single center, open label, investigator-initiated randomised controlled, superiority trial, aiming to compare immunosuppression reduction combined with a third BNT162b2 vaccine dose versus third dose alone. The primary outcome will be seropositivity rate against SARS-CoV-2. A sample size of 154 patients was calculated for the seropositivity endpoint assuming 25% seropositivity in the control group and 50% in the intervention group. A sample of participant per arm will be also teste for T-cell response. We also plan to perform a prospective observational study, evaluating seropositivity among ~500 kidney transplant recipients consenting to receive a third vaccine dose, who are not eligible for the randomised controlled trial.

Ethics and dissemination: The trial is approved by local ethics committee of Rabin medical center (RMC-0192- 21). Results of this trial will be published; trial data will be available. Protocol amendments will be submitted to the local ethics committee.

Registration: The study is registered in clinicaltrials.gov (NCT04961229).

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Article summary

Strengths and limitations of this study

- This randomised controlled trial addresses a question of crucial importance for organ transplant recipients during the COVID-19 pandemic

- Allocation concealment will reduce the risk of bias, though blinding will not be possible

- Antibody level measures for all participants at several timepoints, and partial sampling for T-cell response will provide an overview on protectivity of vaccine

- Currently, no neutralizing antibody testing is planned, limiting the evaluation of protective effect of the vaccine

For peer review only

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Introduction

COVID-19 outbreak has great impact on solid organ transplant recipients. Mortality rates among kidney transplant recipients have been reported between 13 to 50%, with high rates of complications including acute kidney injury (AKI) in 30-89% of hospitalized patients. (1) Severe consequences of COVID-19 were also demonstrated among vaccinated kidney transplant recipient who were infected with SARS-CoV-2, with considerable mortality. (2)

The Pfizer mRNA-based BNT162b2 vaccine, the first vaccine approved by the FDA against SARS-CoV-2 infection has been delivered to over 5 million people in Israel since December 2020. Immunocompromised patients were excluded from the phase III trial evaluating this vaccine, and thus, the efficacy and safety of the vaccine in this patient population are currently not well studied. (3) A large study from Israel has validated the effectiveness of this vaccine in the general population, (4) however the number of vaccinated solid organ transplant recipients in this cohort (n=435) was too small for performing evaluation of vaccine effectiveness for this sub-group. (5)

The American Society of Transplantation and other transplantation societies in the world have recommended vaccinating transplant candidates and recipients against SARS-CoV-2 despite lack of data regarding efficacy in these populations, based on encouraging clinical results in other populations. (6)

Early phase 1/2 studies showed that BNT162b2 elicited strong antibody response in healthy adults. The titer of the neutralizing antibodies increased with dose and

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

increased after the second injection in comparison to the first. (7,8) High rates of antibody response to two doses of the vaccine were also documented in healthy population, accompanied by a distinct Th1 type T cell response. (9) While the role of neutralizing antibodies in protection from SARS-CoV-2 was demonstrated, it is expected that a steady T cell response has a central role against SARS-CoV-2 infection. (10) Solid organ recipient are expected to gain lower immune response to vaccinations with effectiveness varied between different vaccines and different transplanted organ populations. (11) Several studies from Israel demonstrated low rates of antibody response to the BNT162b2 vaccine among solid organ transplant recipients, including 36% seropositivity among 308 kidney transplant recipients 2-4 weeks after the second vaccine dose; (12) 47% seropositivity among 80 liver transplant recipients; (13) 49% among 37 heart transplant recipients; (14) and 18% among 168 lung transplant recipients. (15) Most demonstrated an association between mycophenolic acid dose and calcineurin inhibitors (CNI) blood levels and antibody response. A large study evaluating response to either BNT162b2 vaccine or the mRNA-1273 (Moderna) vaccine among 658 organ transplant recipients also demonstrated low seropositivity rates of 54% a median of 29 days after two vaccine doses. (16)

We plan a randomised controlled trial aiming to evaluate whether a third booster dose of mRNA SARS-CoV-2 vaccine BNT162b2 with or without immunosuppression reduction improves the humoral response in kidney transplant recipients.

Methods and analysis

Study hypothesis and aims

We aim to evaluate the effect of a third mRNA vaccine dose with and without immunosuppression reduction on rates of seropositivity among kidney transplant

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

recipients. We hypothesize that immunosuppression reduction combined with a third dose will demonstrate superiority over a third dose alone in terms of seropositivity rates.

Study design:

The study is a single center, randomised controlled, superiority open-label trial, with an observational cohort as below:

- Randomised controlled trial (2 arms):

Third dose of BNT162b2 vaccine with or without reduction of mycophenolic acid dose (see below).

Observational arm: a third vaccine dose with no change in immunosuppression for patients that are excluded from the randomised trial (see exclusion criteria)

Setting

The study will be conducted at Rabin Medical Center in Israel, in the transplantation follow-up clinic.

Study population:

We will include in both the RCT and observational study adult (age≥18 years) kidney transplant recipients that received two doses of BNT162b2 vaccine at least 3 weeks prior to enrollment, and were seronegative (IgG against the spike protein of SARS-CoV-2 below 50 AU/ml) at least two weeks after the second vaccine dose.

Additional inclusion criteria for the RCT:

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

- Recipients treated by three anti-rejection medications including: prednisone, tacrolimus, mycophenolate mofetil or mycophenolic acid.

- Tacrolimus trough blood levels 5-10 nGr/ml (lower or higher doses will have to be adjusted before re-considering for inclusion)

Exclusion criteria for both RCT and observational part:

- Past infection with SARS-CoV-2
- Pregnancy
- Age below 18 years
- Active infection

Additional exclusion criteria for RCT only:

- Recipients at a high risk for acute or chronic humoral rejection including:

  - Recipients with positive panel-reactive antibody (PRA) (any positive value) at any time before or after transplantation
  - Recipients that had an acute rejection in the last year
  - Recipients less than 6 months after transplantation
  - Recipients that are considered at high risk for rejection according to the primary care nephrologist
  - Recipients taking less than 3 anti-rejection medications
  - Recipients currently treated with mTOR inhibitors (everolimus, sirolimus) and/or azathioprine
  - Recipients treated with plasmapheresis in the previous 3 months

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

- Recipients treated with eculizumab in the last year
- Recipient treated with IVIG in the previous 3 months
- Recipient treated with rituximab in the previous 6 months

The study will be discontinued at any time according to patient request.

Patient randomisation

Patients will be randomised to immunosuppression reduction versus no immunosuppression reduction in a 1:1 ratio. Randomisation will be performed using a computer-generated list of random numbers that will be allocated centrally through a web site.

Interventions:

All recipients more than 6 months post transplantation and at least 3 weeks following second vaccine dose will be approached and invited to a first study visit.

At first visit:

- Signed informed consent will be obtained from participants willing to participate by study investigators who usually work in the transplantation clinic.

- Anti-spike antibody response will be assessed using SARS-CoV-2 IgG II Quant (Abbott©) assay. (17) Participants who have a documented seronegative test in the last 6 weeks will not be tested again.

Participants will be invited for an additional visit once negative serology will be reported, within 7 days of serology collection. At this second visit all participants who

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

gave informed consent to participate in either the prospective non-randomised study or RCT will receive a single vaccine dose.

In addition, participants in the RCT will be randomised into two groups:

1) Third booster dose of BNT162b2 (one standard dose) with no change in immunosuppression protocol

2) Third booster dose of BNT162b2 (one standard dose) with immunosuppression reduction according to protocol (mycophenolic temporary cessation 4 days before (5 half-lives) and one week (expected antibody response) after vaccination (to allow for antibody response).

Patients who will test seronegative will be informed by the study coordinator by phone in which study arm they will be participating and receive instructions for immunosuppression reduction both during the phone call and by written instructions provided to each patient during the first visit (see Appendix). Participants in the observational study will receive a third vaccine standard dose, without any change in immunosuppression (beyond routine care)

For all groups:

- Antibodies titer against spike protein will be evaluated again 2 weeks and 3, 6, 12 months after the third vaccine dose

- T-cell response will be evaluated for a subset of patients in each group (estimated 20 patients per arm) before booster dose, at 2 weeks after booster dose, and at 3 months. For T cell response quantification peripheral blood mononuclear cell (PBMC) will be stimulated for 24 hours with spike protein and secreted interferon-gamma (IFNg) will be measured by ELISA.

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

- Follow-up for adverse events, rejection and SARS-CoV-2 infection will be performed at 2 weeks and at three, 6 and 12 months post third vaccination dose

Outcomes:

The primary outcome of both RCT and observational study is positive humoral response against SARS-CoV-2, defined as anti-spike protein titer above 50 AU/ml at 2 weeks post vaccination

Secondary outcomes

- Positive humoral response against SARS-CoV-2 at 3-, 6-, and 12-months post vaccination
- Log transformed titer of anti S protein at 2 weeks and 3, 6, and 12 months
- Log transformed change in anti-spike protein titer at 2 weeks and 3, 6, and 12 months
- Adverse events to booster dose at 2 weeks post vaccine. Severity of adverse events will be assessed using CTCAE v4.0 criteria
- Acute rejection of the allograft at 2 weeks, 3,6, and 12 months (either documented by biopsy or clinically suspected, defined as increase in creatinine by 20% from baseline, without any other plausible explanation)
- SARS-CoV-2 infection during the follow up period (until 12 months following vaccine)
- Other viral reactivation during the follow up period (VZV, CMV), tested according to clinical suspicion)
- Number of hospitalizations until 12 months

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Monitoring outcomes:

Participants in both RCT and observational study will be invited for a clinic visit at 2 weeks, 3, 6, and 12 months at the post-transplant follow up clinic at Rabin Medical Center. During their visit they will be questioned for adverse events (see Appendix for questionnaire); blood and urine tests will be checked as in the usual post-transplant follow ups for creatinine levels, urine protein or microalbumin, tacrolimus levels and anti-spike protein antibodies. Data will be entered to the REDCap software, and the primary investigator will be responsible to check for timely data entry, missing data and suspected faulty data.

Predefined subgroup analysis:

- Recipients 65 years and older

Sample size:

The trial is designed to demonstrate superiority of immunosuppression reduction in terms of seropositivity rates. For the seropositivity endpoint we calculated a sample size of 77 participants per arm, assuming a 25% seropositivity rate in the control group and 50% in the intervention group, with a 1-sided hypothesis with 2.5% α-risk and 90% power.

For the observational study we assume 500 participants.

Stopping rules:

We plan to assess for acute rejection episodes following first 20 participants allocated to the immunosuppression reduction arm. If an acute rejection episode will be demonstrated within 2 weeks follow up from the booster dose – the study will be terminated. Acute rejection will be defined as 20% increase from baseline creatinine with no other plausible explanation. Need for histological proof of rejection will be

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

discussed case by case by an independent safety monitoring committee. In addition, interim analyses will be performed after the recruitment of each 50 participants. At each analysis, rejection episodes will be evaluated and rates of seropositivity will be assessed. An independent safety monitoring committee of three nephrologists or ID physicians, specialist in managing transplant patients, will meet after each interim analysis to decide whether the study should be terminated.

Statistical analysis

Fisher's exact test will be performed for the proportion of participants with seropositivity, and comparisons of anti-S antibody titers will be tested with the Mann-Whitney U test, and 95% confidence intervals (95% CI) will be calculated. IgG concentrations below threshold of detection will be given the lowest detectable value multiplied by 0.5. A p-value < 0.05 will be considered statistically significant. All data will be analyzed using SPSS (Version 27).

Patient and Public Involvement

No patient involved

Ethics and dissemination

COVID-19 carry significant morbidity and mortality. RCTs and real-life studies show effectiveness of the vaccine. Nevertheless, a correlation between antibody response and protection has been described. In solid organ transplanted recipients, receiving T-cell immunosuppressive therapy, it is reasonable to assume that seronegative people will remain unprotected from SARS-CoV-2. In recent studies, no serious adverse events among transplant recipients receiving two mRNA vaccine doses were found. (12–15) Temporary reduction of immunosuppression during sepsis in kidney

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

transplant recipients was not associated with an increased risk of rejection or long-term graft failure. (18) The primary investigator will have access to the final trial dataset.

This trial's results will be made available through publication. Patient data (de-identified) will be available for other researchers by request. This will be considered for researchers presenting a methodologically adequate protocol, ethical approval, and signing data transfer agreement.

Planned timeline for the study – patients will be recruited during July to September 2021, follow up will be for 12 months. Analysis and writing of the manuscript are planned until June 2022. Current status of the study – still not recruiting.

Author contributions: All authors contributed to conception, design, trial management and planned data analysis. BRZ, RR, DY and AA contributed to trial database and randomisation site design. BRZ, DY and RR wrote the first draft of the manuscript. All authors revised the protocol and approved the final manuscript.

Funding: no external funding. The vaccine doses were provided by the Israeli ministry of health.

For peer review only

References:

1. Azzi Y, Bartash R, Scalea J, Loarte-Campos P, Akalin E. COVID-19 and Solid Organ Transplantation: A Review Article. Transplantation. 2021 Jan 1;105(1):37–55.

2. Tau N, Yahav D, Schneider S, Rozen-Zvi B, Sneineh MA, Rahamimov R. Severe consequences of COVID-19 infection among vaccinated kidney transplant recipients. American Journal of Transplantation [Internet]. [cited 2021 May 28];n/a(n/a). Available from: https://onlinelibrary.wiley.com/doi/abs/10.1111/ajt.16700

3. 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 Dec 31;383(27):2603–15.

4. Dagan N, Barda N, Kepten E, Miron O, Perchik S, Katz MA, et al. BNT162b2 mRNA Covid-19 Vaccine in a Nationwide Mass Vaccination Setting. N Engl J Med. 2021 Feb 24;

5. Roest S, Hoek RAS, Manintveld OC. BNT162b2 mRNA Covid-19 Vaccine in a Nationwide Mass Vaccination Setting. N Engl J Med. 2021 Apr 21;384(20).

6. COVID-19 Vaccine Information for Transplant Recipients [Internet]. American Society of Transplantation. 2021 [cited 2021 May 15]. Available from: https://www.myast.org/covid-19-vaccine-information-transplant-recipients

7. 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 Dec 17;383(25):2439–50.

8. Sahin U, Muik A, Derhovanessian E, Vogler I, Kranz LM, Vormehr M, et al. COVID-19 vaccine BNT162b1 elicits human antibody and TH1 T cell responses. Nature. 2020 Oct;586(7830):594–9.

9. Favresse J, Bayart J-L, Mullier F, Dogné J-M, Closset M, Douxfils J. Early antibody response in healthcare professionals after two doses of SARS-CoV-2 mRNA vaccine (BNT162b2). Clin Microbiol Infect. 2021 May 8;

10. Jin P, Li J, Pan H, Wu Y, Zhu F. Immunological surrogate endpoints of COVID-2019 vaccines: the evidence we have versus the evidence we need. Signal Transduction and Targeted Therapy. 2021 Feb 2;6(1):1–6.

11. Eckerle I, Rosenberger KD, Zwahlen M, Junghanss T. Serologic vaccination response after solid organ transplantation: a systematic review. PLoS One. 2013;8(2):e56974.

12. Rozen-Zvi B, Yahav D, Agur T, Zingerman B, Ben-Zvi H, Atamna A, et al. Antibody response to mRNA SARS-CoV-2 vaccine among kidney transplant recipients - Prospective cohort study. Clin Microbiol Infect. 2021 May 3;

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

13. Rabinowich L, Grupper A, Baruch R, Ben-Yehoyada M, Halperin T, Turner D, et al. Low immunogenicity to SARS-CoV-2 vaccination among liver transplant recipients. J Hepatol. 2021 Apr 20;

14. Itzhaki Ben Zadok O, Shaul AA, Ben-Avraham B, Yaari V, Ben Zvi H, Shostak Y, et al. Immunogenicity of the BNT162b2 mRNA Vaccine in Heart Transplanted Patients-A Prospective Cohort Study. Eur J Heart Fail. 2021 Apr 29;

15. Shostak Y, Shafran N, Heching M, Rosengarten D, Shtraichman O, Shitenberg D, et al. Early humoral response among lung transplant recipients vaccinated with BNT162b2 vaccine. Lancet Respir Med. 2021 May 5;

16. Boyarsky BJ, Werbel WA, Avery RK, Tobian AAR, Massie AB, Segev DL, et al. Antibody Response to 2-Dose SARS-CoV-2 mRNA Vaccine Series in Solid Organ Transplant Recipients. JAMA. 2021 May 5;

17. Abbott laboratories. SARS-CoV-2 IgG II Quant [Internet]. 2020 [cited 2021 Feb 24]. Available from: https://www.corelaboratory.abbott/int/en/offerings/segments/infectious-disease/sars-cov-2

18. Yahav D, Sulimani O, Green H, Margalit I, Ben-Zvi H, Mor E, et al. Immunosuppression reduction in kidney transplant recipients during bacterial infection-A retrospective study. Clin Transplant. 2019 Oct;33(10):e13707.

For peer review only

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Appendix

Questionnaire for adverse events

Did you have any of these symptoms after the first vaccine injection?

1st vaccine date:	yes	No	Severity
Local pain on injection site			
Muscle pain			
headache			
Fatigue			
Allergic reaction			
Fever			
Lymph node enlargement			
Need of medical treatment			
Need of hospitalization			

Did you have any of these symptoms after the second vaccine injection?

2nd vaccine date:	yes	No	Severity
Local pain on injection site			
Muscle pain			
headache			
Fatigue			
Allergic reaction			
Fever			
Lymph node enlargement			
Need of medical treatment			
Need of hospitalization			

For peer review only

Instructions for study participant - immunosuppression management

Dear participant,

You participate in this randomised controlled trial to evaluate the safety and effectiveness of a booster (third) dose of mRNA SARS-CoV-2 BNT162b2 vaccine with or without immunosuppression reduction.

In this study you will be randomised by a computer to be in one of two arms of the study. Following randomization, you will be informed by the study coordinator, who is a nurse in the transplantation clinic, to which arm you belong:

Arm 1 – immunosuppression continuation: continue with your medications as usual before and after the third vaccine injection date.

Arm 2 - immunosuppression reduction: stop taking Myfortic or Cellcept from 4 days before your scheduled vaccine date and 7 days after. Please continue with all your other medications as usual. 4 days before the vaccine – if the vaccine is on Thursday, you stop Myfortic or Cellcept on Sunday and resume on next Thursday; if the vaccine is on Wednesday, you stop Myfortic or Cellcept on Saturday and resume on next Wednesday.

If you have any questions, don't hesitate to contact the study team and/or the transplantation clinic.

For peer review only

SPIRIT
STANDARD PROTOCOL ITEMS: RECOMMENDATIONS FOR INTERVENTIONAL TRIALS

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description
Administrative information		
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym – page 1
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry – page 1
	2b	All items from the World Health Organization Trial Registration Data Set – the trial we be registered in the NIH registration data set and the Israeli ministry of health data set
Protocol version	3	Date and version identifier – 3-Jun-2021, Version 1
Funding	4	Sources and types of financial, material, and other support – page 13
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors – page 1, page 13
	5b	Name and contact information for the trial sponsor – no sponsor
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities – not applicable
	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee) – page 12
Introduction		
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention – page 4-5
	6b	Explanation for choice of comparators - page 4-5
Objectives	7	Specific objectives or hypotheses – page 6, page 11

For peer review only

Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory) – page 6-11
Methods: Participants, interventions, and outcomes		
Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained – page 6
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists) – page 6-7
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered – page 8
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease) - page 8
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests) – page 9
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial – appendix
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended – page 10-11
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure) – page 8-11
Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations – page 11-12
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size – page 11-12
Methods: Assignment of interventions (for controlled trials)		
Allocation:		

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

BMJ Open: first published as 10.1136/bmjopen-2012-002314 on 4 December 2012. Downloaded from http://bmjopen.bmj.com/ on April 19, 2024 by guest. Protected by copyright.

Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions – page 8
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned – page 8
Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions – page 8
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how – page 8
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial – not applicable, no blinding
Methods: Data collection, management, and analysis		
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol – page 11
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols – page 11
Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol – page 11
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol – page 12
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses) – page 12
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation) – page 12

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

BMJ Open: first published as 10.1136/bmjopen-2012-002055 on 25 October 2012. Downloaded from http://bmjopen.bmj.com/ on April 19, 2024 by guest. Protected by copyright.

Methods: Monitoring

Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed – page 12
	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial – page 12
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct - appendix
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor – not applicable

Ethics and dissemination

Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval – page 2, 13
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators) – page 2
Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32) – page 8
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable – not applicable
Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial – page 11
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site – page 13
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators – page 13
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation – not applicable

peer review only

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions – page 13
	31b	Authorship eligibility guidelines and any intended use of professional writers – page 13
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code – page 13
Appendices		
Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates - appendix
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable- not applicable

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.

For peer review only

BMJ Open

Immunosuppression reduction when administering a booster dose of the BNT162b2 mRNA SARS-CoV-2 vaccine in kidney transplant recipients without adequate humoral response following two vaccine doses: protocol for a randomised controlled trial (BECAME study)

Journal:	*BMJ Open*
Manuscript ID	bmjopen-2021-055611.R1
Article Type:	Protocol
Date Submitted by the Author:	20-Sep-2021
Complete List of Authors:	Yahav, Dafna; Rabin Medical Center, Rozen-Zvi, Benaya; Rabin Medical Center, Mashraki, Tiki; Rabin Medical Center, Department of Nephrology and Hypertension Atamna, Alaa; Tel Aviv University, Sackler school of medicine; Rabin Medical Center, Infectious diseases unit Ben-Zvi, Haim ; Rabin Medical Center, Department of Medicine E, Beilinson Hospital Bar-Haim, Erez; Israel Institute for Biological Research, Department of Biochemistry and Molecular Genetics; IIBR Rahamimov, R.; Rabin Medical Center
Primary Subject Heading:	Infectious diseases
Secondary Subject Heading:	Immunology (including allergy), Public health, Infectious diseases
Keywords:	Renal transplantation < NEPHROLOGY, COVID-19, IMMUNOLOGY

SCHOLARONE™
Manuscripts

BMJ Open: first published as 10.1136/bmjopen-2021-055611 on 11 October 2021. Downloaded from http://bmjopen.bmj.com/ on April 9, 2024 by guest. Protected by copyright.

I, the Submitting Author has the right to grant and does grant on behalf of all authors of the Work (as defined in the below author licence), an exclusive licence and/or a non-exclusive licence for contributions from authors who are: i) UK Crown employees; ii) where BMJ has agreed a CC-BY licence shall apply, and/or iii) in accordance with the terms applicable for US Federal Government officers or employees acting as part of their official duties; on a worldwide, perpetual, irrevocable, royalty-free basis to BMJ Publishing Group Ltd ("BMJ") its licensees and where the relevant Journal is co-owned by BMJ to the co-owners of the Journal, to publish the Work in this journal and any other BMJ products and to exploit all rights, as set out in our licence.

The Submitting Author accepts and understands that any supply made under these terms is made by BMJ to the Submitting Author unless you are acting as an employee on behalf of your employer or a postgraduate student of an affiliated institution which is paying any applicable article publishing charge ("APC") for Open Access articles. Where the Submitting Author wishes to make the Work available on an Open Access basis (and intends to pay the relevant APC), the terms of reuse of such Open Access shall be governed by a Creative Commons licence – details of these licences and which Creative Commons licence will apply to this Work are set out in our licence referred to above.

Other than as permitted in any relevant BMJ Author's Self Archiving Policies, I confirm this Work has not been accepted for publication elsewhere, is not being considered for publication elsewhere and does not duplicate material already published. I confirm all authors consent to publication of this Work and authorise the granting of this licence.

For peer review only

Immunosuppression reduction when administering a booster dose of the BNT162b2 mRNA SARS-CoV-2 vaccine in kidney transplant recipients without adequate humoral response following two vaccine doses: protocol for a randomised controlled trial (BECAME study)

Dafna Yahav[1,2*], Benaya Rozen-Zvi[2,3*], Tiki Mashraki[3,4], Alaa Atamna[1,2], Haim Ben-Zvi[2,5], Erez Bar-Haim[6], Ruth Rahamimov[2,3,4]

[1] Infectious Diseases Unit, Rabin Medical Center, Beilinson Campus, Petah-Tikva, Israel

[2] Sackler Faculty of Medicine, Tel Aviv University, Tel Aviv, Israel

[3] Department of Nephrology and Hypertension, Rabin Medical Center, Beilinson Campus, Petah-Tikva, Israel

[4] Department of Transplantation, Rabin Medical Center, Beilinson Campus, Petah-Tikva, Israel

[5] Clinical Microbiology Laboratory, Rabin Medical Center, Beilinson Hospital, Petah-Tikva, Israel

[6] Department of Biochemistry and Molecular Genetics, Israel Institute for Biological Research, 74100 Ness-Ziona, Israel.

[*] BRZ and DY contributed equally for this work

Corresponding author: Prof. Dafna Yahav, Infectious Diseases Unit, Rabin Medical Center, Beilinson Hospital, 39 Jabotinsky Road, Petah-Tikva, 49100, Israel. Phone: 972-3-9377512, Fax: 972-3-9376512. E-mail: dafna.yahav@gmail.com

Keywords: kidney transplant recipients; COVID-19; vaccine; immunosuppression reduction; randomized controlled trial

Word count:

For peer review only

Abstract

Introduction: Inadequate antibody response to mRNA SARS-CoV-2 vaccination has been described among kidney transplant recipients. Immunosuppression level and specifically, use of antimetabolite in the maintenance immunosuppressive regimen, are associated with inadequate response. In light of the severe consequences of COVID-19 in solid organ transplant recipients, we believe it is justified to examine new vaccination strategies in these patients.

Methods and analysis: BECAME is a single center, open label, investigator-initiated randomised controlled, superiority trial, aiming to compare immunosuppression reduction combined with a third BNT162b2 vaccine dose versus third dose alone. The primary outcome will be seropositivity rate against SARS-CoV-2. A sample size of 154 patients was calculated for the seropositivity endpoint assuming 25% seropositivity in the control group and 50% in the intervention group. A sample of participants per arm will be also tested for T-cell response. We also plan to perform a prospective observational study, evaluating seropositivity among ~350 kidney transplant recipients consenting to receive a third vaccine dose, who are not eligible for the randomised controlled trial.

Ethics and dissemination: The trial is approved by local ethics committee of Rabin medical center (RMC-0192- 21). All participants will be required to provide written informed consent. Results of this trial will be published; trial data will be available. Protocol amendments will be submitted to the local ethics committee.

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Registration: The study is registered in clinicaltrials.gov (NCT04961229).

Article summary

Strengths and limitations of this study

- This randomised controlled trial addresses a question of crucial importance for organ transplant recipients during the COVID-19 pandemic

- Allocation concealment will reduce the risk of bias, though blinding will not be possible

- Antibody level measures for all participants at several timepoints, and partial sampling for T-cell response will provide an overview on protectivity of vaccine

- Currently, no neutralizing antibody testing is planned, limiting the evaluation of protective effect of the vaccine

For peer review only

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Introduction

COVID-19 outbreak has great impact on solid organ transplant recipients. Mortality rates among kidney transplant recipients have been reported between 13 to 50%, with high rates of complications, including acute kidney injury (AKI) in 30-89% of hospitalized patients. (1) Severe consequences of COVID-19 were also demonstrated among vaccinated kidney transplant recipient who were infected with SARS-CoV-2, with considerable mortality. (2)

The Pfizer mRNA-based BNT162b2 vaccine, the first vaccine approved by the FDA against SARS-CoV-2 infection, has been delivered to over 5 million people in Israel since December 2020. Immunocompromised patients were excluded from the phase III trial evaluating this vaccine, and thus, the efficacy and safety of the vaccine in this patient population are currently not well studied. (3) A large study from Israel has validated the effectiveness of this vaccine in the general population, (4) however the number of vaccinated solid organ transplant recipients in this cohort (n=435) was too small for performing evaluation of vaccine effectiveness for this sub-group. (5)

The American Society of Transplantation and other transplantation societies in the world have recommended vaccinating transplant candidates and recipients against SARS-CoV-2 despite lack of data regarding efficacy in these populations, based on encouraging clinical results in other populations. (6)

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Early phase 1/2 studies showed that BNT162b2 elicited strong antibody response in healthy adults. The titer of the neutralizing antibodies increased with dose and also increased after the second injection in comparison to the first. (7,8) High rates of antibody response to two doses of the vaccine were also documented in healthy population, accompanied by a distinct Th1 type T cell response. (9) While the role of neutralizing antibodies in protection from SARS-CoV-2 was demonstrated, it is expected that a steady T cell response has a central role against SARS-CoV-2 infection. (10) Solid organ recipient are expected to gain lower immune response to vaccinations with varying effectiveness between different vaccines and different transplanted organ populations. (11) Broad impairments in both humoral and cellular response to mRNA vaccines have been reported in kidney transplant recipients. Several studies from Israel demonstrated low rates of antibody response to the BNT162b2 vaccine among solid organ transplant recipients, including 36% seropositivity among 308 kidney transplant recipients 2-4 weeks after the second vaccine dose; (12) 47% seropositivity among 80 liver transplant recipients; (13) 49% among 37 heart transplant recipients; (14) and 18% among 168 lung transplant recipients. (15) Most demonstrated an association between mycophenolic acid dose and calcineurin inhibitors (CNI) blood levels and antibody response. A large study evaluating response to either BNT162b2 vaccine or the mRNA-1273 (Moderna) vaccine among 658 organ transplant recipients also demonstrated low seropositivity rates of 54% a median of 29 days after two vaccine doses. (16) In addition, diminished generation of plasmablasts and memory B cells in response to mRNA vaccine among kidney transplant recipients were reported. (17) Impairments in T cell response were also described, with high rates of spike-specific T helper cell response, however reduced magnitude of response, as well as limited effector cytokine

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

production. (18,19) Recent studies demonstrate improve humoral and cellular responses following third booster mRNA vaccine dose among transplant recipients. Yet, a considerable portion of these patients remained seronegative. (20–22) We plan a randomised controlled trial aiming to evaluate whether a third booster dose of mRNA SARS-CoV-2 vaccine BNT162b2, with or without immunosuppression reduction, improves the humoral response in kidney transplant recipients.

Methods and analysis

Study hypothesis and aims

We aim to evaluate the effect of a third mRNA vaccine dose with and without immunosuppression reduction on rates of seropositivity among kidney transplant recipients. We hypothesize that immunosuppression reduction combined with a third dose will demonstrate superiority over a third dose alone in terms of seropositivity rates.

Study design:

The study is a single center, randomised controlled, superiority, open-label trial, with an observational cohort as below:

- Randomised controlled trial (2 arms):

Third dose of BNT162b2 vaccine with or without reduction of mycophenolic acid dose (see below).

Observational arm: a third vaccine dose with no change in immunosuppression for patients that are excluded from the randomised trial (see exclusion criteria)

Setting

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

The study will be conducted at Rabin Medical Center in Israel, in the transplantation follow-up clinic.

Study population:

We will include in both the RCT and observational study adult (age≥18 years) kidney transplant recipients that received two doses of BNT162b2 vaccine at least 3 weeks prior to enrollment, and were seronegative (IgG against the spike protein of SARS-CoV-2 below 50 AU/ml) at least two weeks after the second vaccine dose.

Additional inclusion criteria for the RCT:

- Recipients treated with three anti-rejection medications including: prednisone, tacrolimus, mycophenolate mofetil or mycophenolic acid.

Patients with any dosage of mycophenolic acid, mycophenolate mofetil, and prednisone will be eligible for inclusion. Regarding tacrolimus, trough blood levels 5-10 nGr/ml will be required for inclusion. Lower or higher doses will have to be adjusted before re-considering for inclusion.

Exclusion criteria for both RCT and observational part:

- Past infection with SARS-CoV-2
- Pregnancy
- Age below 18 years
- Active infection

Additional exclusion criteria for RCT only:

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

- Recipients at a high risk for acute or chronic humoral rejection including:

  - Recipients with positive panel-reactive antibody (PRA) (any positive value) at any time before or after transplantation
  - Recipients that had an acute rejection in the last year
  - Recipients less than 6 months after transplantation
  - Recipients that are considered at high risk for rejection according to the primary care nephrologist
  - Recipients taking less than 3 anti-rejection medications
  - Recipients currently treated with mTOR inhibitors (everolimus, sirolimus) and/or azathioprine
  - Recipients treated with plasmapheresis in the previous 3 months
  - Recipients treated with eculizumab in the last year
  - Recipient treated with IVIG in the previous 3 months
  - Recipient treated with rituximab in the previous 6 months

Patients can withdraw from the study at any time.

Patient randomisation

Patients will be randomised to immunosuppression reduction versus no immunosuppression reduction in a 1:1 ratio. Randomisation will be performed using a computer-generated list of random numbers that will be allocated centrally through a web site.

Interventions:

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

All recipients more than 6 months post transplantation and at least 3 weeks following second vaccine dose will be approached and invited to a first study visit.

At first visit:

- Signed informed consent will be obtained from participants willing to participate by study investigators who routinely work in the transplantation clinic.

- Anti-spike antibody response will be assessed using SARS-CoV-2 IgG II Quant (Abbott©) assay. (23) Participants who have a documented seronegative test in the last 6 weeks will not be tested again.

- Tacrolimus levels will be obtained

Participants will be invited for an additional visit once negative serology result will be reported, within 7 days of test collection. At this second visit all participants who gave informed consent to participate in either the prospective non-randomised study or RCT will receive a single vaccine dose.

In addition, participants in the RCT will be randomised into two groups:

1) Third booster dose of BNT162b2 (one standard dose) with no change in immunosuppression protocol

2) Third booster dose of BNT162b2 (one standard dose) with immunosuppression reduction according to protocol (mycophenolic temporary cessation 4 days before (5 half-lives) and one week (expected antibody response) after vaccination (to allow for antibody response).

Patients who will test seronegative will be informed by the study coordinator by phone in which study arm they will be participating and receive instructions for

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

immunosuppression reduction both during the phone call and by written instructions provided to each patient during the first visit (see Appendix). Participants in the observational study will receive a third vaccine standard dose, without any change in immunosuppression (beyond routine care)

For all groups:

- Antibodies titer against spike protein will be evaluated at 2 weeks and 3, 6, 12 months after the third vaccine dose

- T-cell response will be evaluated for a small, randomly selected, subset of patients in each group (estimated 20 patients per arm) before booster dose, at 2 weeks after booster dose, and at 3 months. For T cell response quantification, peripheral blood mononuclear cell (PBMC) will be stimulated for 24 hours with spike protein and secreted interferon-gamma (IFNg) will be measured by ELISA (using the SARS-CoV-2 IFNg release assay EUROIMMUN, Lübeck, Germany). (24)

- Follow-up for adverse events, rejection, or SARS-CoV-2 infection will be performed at 2 weeks and at three, 6 and 12 months post third vaccination dose

Outcomes:

The primary outcome of both RCT and observational study will be humoral response against SARS-CoV-2, defined as anti-spike protein titer above 50 AU/ml at 2 weeks post vaccination

Secondary outcomes

- Humoral response against SARS-CoV-2 at 3-, 6-, and 12-months post vaccination

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

- Log transformed titer of anti S protein at 2 weeks and 3, 6, and 12 months
- Log transformed change in anti-spike protein titer at 2 weeks and 3, 6, and 12 months
- Adverse events to booster dose at 2 weeks post vaccine. Severity of adverse events will be assessed using CTCAE v4.0 criteria
- Acute rejection of the allograft at 2 weeks, 3,6, and 12 months (documented by biopsy)
- SARS-CoV-2 infection during the follow up period (until 12 months following vaccine)
- Other viral reactivation during the follow up period (VZV, CMV), tested according to clinical suspicion)
- Number of hospitalizations until 12 months

Due to technical limitations, we do not plan biobanking in this study.

Monitoring outcomes:

Participants in both RCT and observational study will be invited for a clinic visit at 2 weeks, 3, 6, and 12 months at the post-transplant follow up clinic at Rabin Medical Center. During their visit they will be questioned for adverse events (see Appendix for questionnaire); blood and urine tests will be checked as in the usual post-transplant follow ups for creatinine levels, any increase in creatinine of more than 20% will be promptly followed by kidney biopsy (after excluding other reasone), urine protein or microalbumin, tacrolimus levels and anti-spike protein antibodies. Data will be entered to the REDCap software, and the primary investigator will be responsible to check for timely data entry, missing data and suspected faulty data.

Predefined subgroup analysis:

- Recipients 65 years and older

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Sample size:

The trial is designed to demonstrate superiority of immunosuppression reduction in terms of seropositivity rates. For the seropositivity endpoint we calculated a sample size of 77 participants per arm, assuming a 25% seropositivity rate in the control group and 50% in the intervention group, (22) with a 1-sided hypothesis with 2.5% α-risk and 90% power.

For the observational study we assume ~350 participants.

Stopping rules:

We plan to assess for acute rejection episodes following first 20 participants allocated to the immunosuppression reduction arm. If an acute rejection episode will be demonstrated within 2 weeks follow up from the booster dose – the study will be terminated. Acute rejection will be defined as 20% increase from baseline creatinine with no other plausible explanation. Need for histological proof of rejection will be discussed case by case by an independent safety monitoring committee. In addition, interim analyses will be performed after the recruitment of each 50 participants. At each analysis, rejection episodes will be evaluated and rates of seropositivity will be assessed. An independent safety monitoring committee of three nephrologists or ID physicians, specialist in managing transplant patients, will meet after each interim analysis to decide whether the study should be terminated.

Limitations

This study is an investigator initiated study, addressing a practical clinical need. However, it has several limitations. First, the study is open label, however we do not expect that to influence the primary laboratory outcome. Second, the main limitation would be the limited analysis of T-cell response (planned for only~40 participants),

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

and the absence of neutralizing antibody test. Nevertheless, positive correlation has been reported between anti-spike antibodies and neutralizing antibodies. (25)

Statistical analysis

Fisher's exact test will be performed for the proportion of participants with seropositivity, and comparisons of anti-S antibody titers will be tested with the Mann-Whitney U test, and 95% confidence intervals (95% CI) will be calculated. IgG concentrations below threshold of detection will be given the lowest detectable value multiplied by 0.5. A p-value < 0.05 will be considered statistically significant. All data will be analyzed using SPSS (Version 27).

Patient and Public Involvement

No patient involved

Ethics and dissemination

In recent studies, no serious adverse events among transplant recipients receiving two mRNA vaccine doses were found. (12–15) Temporary reduction of immunosuppression during sepsis in kidney transplant recipients was not associated with an increased risk of rejection or long-term graft failure. (26) The trial is approved by local ethics committee of Rabin medical center (RMC-0192- 21). All participants will be required to provide written informed consent.

This trial's results will be made available through publication. Patient data (de-identified) will be available for other researchers by request. This will be considered for researchers presenting a methodologically adequate protocol, ethical approval, and signing data transfer agreement.

Study timeline

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Patients will be recruited during September-November 2021, follow up will be for 12 months. Analysis and writing of the manuscript are planned until June 2023. Current status of the study – still not recruiting.

Author contributions: All authors contributed to conception, design, trial management and planned data analysis. BRZ, RR, and DY contributed to the conception of the trial; BRZ, RR, DY, AA, EBH, and TM contributed to the trial design; BRZ, RR, DY, HBZ and AA contributed to trial database and randomisation site design. BRZ, DY and RR wrote the first draft of the manuscript. All authors revised the protocol and approved the final manuscript.

Funding: no external funding. The vaccine doses were provided by the Israeli ministry of health.

Competing interests – all authors declare no competing interests

For peer review only

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

References:

1. Azzi Y, Bartash R, Scalea J, Loarte-Campos P, Akalin E. COVID-19 and Solid Organ Transplantation: A Review Article. Transplantation. 2021 Jan 1;105(1):37–55.

2. Tau N, Yahav D, Schneider S, Rozen-Zvi B, Sneineh MA, Rahamimov R. Severe consequences of COVID-19 infection among vaccinated kidney transplant recipients. American Journal of Transplantation [Internet]. [cited 2021 May 28];n/a(n/a). Available from: https://onlinelibrary.wiley.com/doi/abs/10.1111/ajt.16700

3. 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 Dec 31;383(27):2603–15.

4. Dagan N, Barda N, Kepten E, Miron O, Perchik S, Katz MA, et al. BNT162b2 mRNA Covid-19 Vaccine in a Nationwide Mass Vaccination Setting. N Engl J Med. 2021 Feb 24;

5. Roest S, Hoek RAS, Manintveld OC. BNT162b2 mRNA Covid-19 Vaccine in a Nationwide Mass Vaccination Setting. N Engl J Med. 2021 Apr 21;384(20).

6. COVID-19 Vaccine Information for Transplant Recipients [Internet]. American Society of Transplantation. 2021 [cited 2021 May 15]. Available from: https://www.myast.org/covid-19-vaccine-information-transplant-recipients

7. 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 Dec 17;383(25):2439–50.

8. Sahin U, Muik A, Derhovanessian E, Vogler I, Kranz LM, Vormehr M, et al. COVID-19 vaccine BNT162b1 elicits human antibody and TH1 T cell responses. Nature. 2020 Oct;586(7830):594–9.

9. Favresse J, Bayart J-L, Mullier F, Dogné J-M, Closset M, Douxfils J. Early antibody response in healthcare professionals after two doses of SARS-CoV-2 mRNA vaccine (BNT162b2). Clin Microbiol Infect. 2021 May 8;

10. Jin P, Li J, Pan H, Wu Y, Zhu F. Immunological surrogate endpoints of COVID-2019 vaccines: the evidence we have versus the evidence we need. Signal Transduction and Targeted Therapy. 2021 Feb 2;6(1):1–6.

11. Eckerle I, Rosenberger KD, Zwahlen M, Junghanss T. Serologic vaccination response after solid organ transplantation: a systematic review. PLoS One. 2013;8(2):e56974.

12. Rozen-Zvi B, Yahav D, Agur T, Zingerman B, Ben-Zvi H, Atamna A, et al. Antibody response to mRNA SARS-CoV-2 vaccine among kidney transplant recipients - Prospective cohort study. Clin Microbiol Infect. 2021 May 3;

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

13. Rabinowich L, Grupper A, Baruch R, Ben-Yehoyada M, Halperin T, Turner D, et al. Low immunogenicity to SARS-CoV-2 vaccination among liver transplant recipients. J Hepatol. 2021 Apr 20;

14. Itzhaki Ben Zadok O, Shaul AA, Ben-Avraham B, Yaari V, Ben Zvi H, Shostak Y, et al. Immunogenicity of the BNT162b2 mRNA Vaccine in Heart Transplanted Patients-A Prospective Cohort Study. Eur J Heart Fail. 2021 Apr 29;

15. Shostak Y, Shafran N, Heching M, Rosengarten D, Shtraichman O, Shitenberg D, et al. Early humoral response among lung transplant recipients vaccinated with BNT162b2 vaccine. Lancet Respir Med. 2021 May 5;

16. Boyarsky BJ, Werbel WA, Avery RK, Tobian AAR, Massie AB, Segev DL, et al. Antibody Response to 2-Dose SARS-CoV-2 mRNA Vaccine Series in Solid Organ Transplant Recipients. JAMA. 2021 May 5;

17. Rincon-Arevalo H, Choi M, Stefanski A-L, Halleck F, Weber U, Szelinski F, et al. Impaired humoral immunity to SARS-CoV-2 BNT162b2 vaccine in kidney transplant recipients and dialysis patients. Sci Immunol. 2021 Jun 15;6(60):eabj1031.

18. Sattler A, Schrezenmeier E, Weber UA, Potekhin A, Bachmann F, Straub-Hohenbleicher H, et al. Impaired humoral and cellular immunity after SARS-CoV-2 BNT162b2 (tozinameran) prime-boost vaccination in kidney transplant recipients. J Clin Invest. 2021 Jul 15;131(14):150175.

19. Stumpf J, Siepmann T, Lindner T, Karger C, Schwöbel J, Anders L, et al. Humoral and cellular immunity to SARS-CoV-2 vaccination in renal transplant versus dialysis patients: A prospective, multicenter observational study using mRNA-1273 or BNT162b2 mRNA vaccine. The Lancet Regional Health – Europe [Internet]. 2021 Jul 22 [cited 2021 Aug 31];0(0). Available from: https://www.thelancet.com/journals/lanepe/article/PIIS2666-7762(21)00155-1/abstract

20. Stumpf J, Tonnus W, Paliege A, Rettig R, Steglich A, Gembardt F, et al. Cellular And Humoral Immune Responses after Three Doses of BNT162b2 mRNA SARS-Cov-2 Vaccine in Kidney Transplant. Transplantation. 2021 Jul 22;

21. Kamar N, Abravanel F, Marion O, Couat C, Izopet J, Del Bello A. Three Doses of an mRNA Covid-19 Vaccine in Solid-Organ Transplant Recipients. New England Journal of Medicine. 2021 Aug 12;385(7):661–2.

22. Hall VG, Ferreira VH, Ku T, Ierullo M, Majchrzak-Kita B, Chaparro C, et al. Randomized Trial of a Third Dose of mRNA-1273 Vaccine in Transplant Recipients. New England Journal of Medicine. 2021 Aug 11;0(0):null.

23. Abbott laboratories. SARS-CoV-2 IgG II Quant [Internet]. 2020 [cited 2021 Feb 24]. Available from:

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

https://www.corelaboratory.abbott/int/en/offerings/segments/infectious-disease/sars-cov-2

24. Huzly D, Panning M, Smely F, Enders M, Komp J, Steinmann D. Validation and performance evaluation of a novel interferon-γ release assay for the detection of SARS-CoV-2 specific T-cell response [Internet]. 2021 Jul [cited 2021 Sep 2] p. 2021.07.17.21260316. Available from: https://www.medrxiv.org/content/10.1101/2021.07.17.21260316v1

25. Trougakos IP, Terpos E, Zirou C, Sklirou AD, Apostolakou F, Gumeni S, et al. Comparative kinetics of SARS-CoV-2 anti-spike protein RBD IgGs and neutralizing antibodies in convalescent and naïve recipients of the BNT162b2 mRNA vaccine versus COVID-19 patients. BMC Medicine. 2021 Aug 23;19(1):208.

26. Yahav D, Sulimani O, Green H, Margalit I, Ben-Zvi H, Mor E, et al. Immunosuppression reduction in kidney transplant recipients during bacterial infection-A retrospective study. Clin Transplant. 2019 Oct;33(10):e13707.

For peer review only

Appendix

Questionnaire for adverse events

Did you have any of these symptoms after the first vaccine injection?

1st vaccine date:	yes	No	Severity
Local pain on injection site			
Muscle pain			
headache			
Fatigue			
Allergic reaction			
Fever			
Lymph node enlargement			
Need of medical treatment			
Need of hospitalization			

Did you have any of these symptoms after the second vaccine injection?

2nd vaccine date:	yes	No	Severity
Local pain on injection site			
Muscle pain			
headache			
Fatigue			
Allergic reaction			
Fever			
Lymph node enlargement			
Need of medical treatment			
Need of hospitalization			

For peer review only

Instructions for study participant - immunosuppression management

Dear participant,

You participate in this randomised controlled trial to evaluate the safety and effectiveness of a booster (third) dose of mRNA SARS-CoV-2 BNT162b2 vaccine with or without immunosuppression reduction.

In this study you will be randomised by a computer to be in one of two arms of the study. Following randomization, you will be informed by the study coordinator, who is a nurse in the transplantation clinic, to which arm you belong:

Arm 1 – immunosuppression continuation: continue with your medications as usual before and after the third vaccine injection date.

Arm 2 - immunosuppression reduction: stop taking Myfortic or Cellcept from 4 days before your scheduled vaccine date and 7 days after. Please continue with all your other medications as usual. 4 days before the vaccine – if the vaccine is on Thursday, you stop Myfortic or Cellcept on Sunday and resume on next Thursday; if the vaccine is on Wednesday, you stop Myfortic or Cellcept on Saturday and resume on next Wednesday.

If you have any questions, don't hesitate to contact the study team and/or the transplantation clinic.

For peer review only

SPIRIT
STANDARD PROTOCOL ITEMS: RECOMMENDATIONS FOR INTERVENTIONAL TRIALS

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description
Administrative information		
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym – page 1
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry – page 1,3
	2b	All items from the World Health Organization Trial Registration Data Set – the trial we be registered in the NIH registration data set and the Israeli ministry of health data set – page 3
Protocol version	3	Date and version identifier – 10-Sep-2021, Version 2
Funding	4	Sources and types of financial, material, and other support – page 14
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors – page 1, page 14
	5b	Name and contact information for the trial sponsor – no sponsor
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities – not applicable
	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee) – page 13
Introduction		
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention – page 4-6
	6b	Explanation for choice of comparators - page 4-6
Objectives	7	Specific objectives or hypotheses – page 6

For peer review only

Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory) – page 6-11

Methods: Participants, interventions, and outcomes

Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained – page 6
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists) – page 6-7
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered – page 8
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease) - page 8
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests) – page 9
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial – appendix
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended – page 10-11
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure) – page 8-11
Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations – page 12
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size – page 12-13

Methods: Assignment of interventions (for controlled trials)

Allocation:

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

BMJ Open: first published as 10.1136/bmjopen-2012-001570 on 8 August 2012. Downloaded from http://bmjopen.bmj.com/ on April 19, 2024 by guest. Protected by copyright.

Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions – page 8
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned – page 8
Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions – page 8
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how – page 8
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial – not applicable, no blinding
Methods: Data collection, management, and analysis		
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol – page 11
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols – page 11
Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol – page 11
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol – page 12-13
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses) – page 12-13
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation) – page 12-13

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

BMJ Open: first published as 10.1136/bmjopen-2012-002055 on 25 October 2012. Downloaded from http://bmjopen.bmj.com/ on April 19, 2024 by guest. Protected by copyright.

Methods: Monitoring

Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed – page 12
	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial – page 12
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct - appendix
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor – not applicable

Ethics and dissemination

Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval – page 2, 13
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators) – page 2
Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32) – page 8
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable – not applicable
Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial – page 11
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site – page 13
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators – page 13
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation – not applicable

peer review only

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions – page 13
	31b	Authorship eligibility guidelines and any intended use of professional writers – page 13
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code – page 13
Appendices		
Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates - appendix
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable- not applicable

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.

For peer review only

BMJ Open

Immunosuppression reduction when administering a booster dose of the BNT162b2 mRNA SARS-CoV-2 vaccine in kidney transplant recipients without adequate humoral response following two vaccine doses: protocol for a randomised controlled trial (BECAME study)

Journal:	*BMJ Open*
Manuscript ID	bmjopen-2021-055611.R2
Article Type:	Protocol
Date Submitted by the Author:	28-Sep-2021
Complete List of Authors:	Yahav, Dafna; Rabin Medical Center, Rozen-Zvi, Benaya; Rabin Medical Center, Mashraki, Tiki; Rabin Medical Center, Department of Nephrology and Hypertension Atamna, Alaa; Tel Aviv University, Sackler school of medicine; Rabin Medical Center, Infectious diseases unit Ben-Zvi, Haim ; Rabin Medical Center, Department of Medicine E, Beilinson Hospital Bar-Haim, Erez; Israel Institute for Biological Research, Department of Biochemistry and Molecular Genetics; IIBR Rahamimov, R.; Rabin Medical Center
Primary Subject Heading:	Infectious diseases
Secondary Subject Heading:	Immunology (including allergy), Public health, Infectious diseases
Keywords:	Renal transplantation < NEPHROLOGY, COVID-19, IMMUNOLOGY

SCHOLARONE™
Manuscripts

BMJ Open: first published as 10.1136/bmjopen-2021-055611 on 11 October 2021. Downloaded from http://bmjopen.bmj.com/ on April 9, 2024 by guest. Protected by copyright.

I, the Submitting Author has the right to grant and does grant on behalf of all authors of the Work (as defined in the below author licence), an exclusive licence and/or a non-exclusive licence for contributions from authors who are: i) UK Crown employees; ii) where BMJ has agreed a CC-BY licence shall apply, and/or iii) in accordance with the terms applicable for US Federal Government officers or employees acting as part of their official duties; on a worldwide, perpetual, irrevocable, royalty-free basis to BMJ Publishing Group Ltd ("BMJ") its licensees and where the relevant Journal is co-owned by BMJ to the co-owners of the Journal, to publish the Work in this journal and any other BMJ products and to exploit all rights, as set out in our licence.

The Submitting Author accepts and understands that any supply made under these terms is made by BMJ to the Submitting Author unless you are acting as an employee on behalf of your employer or a postgraduate student of an affiliated institution which is paying any applicable article publishing charge ("APC") for Open Access articles. Where the Submitting Author wishes to make the Work available on an Open Access basis (and intends to pay the relevant APC), the terms of reuse of such Open Access shall be governed by a Creative Commons licence – details of these licences and which Creative Commons licence will apply to this Work are set out in our licence referred to above.

Other than as permitted in any relevant BMJ Author's Self Archiving Policies, I confirm this Work has not been accepted for publication elsewhere, is not being considered for publication elsewhere and does not duplicate material already published. I confirm all authors consent to publication of this Work and authorise the granting of this licence.

For peer review only

Immunosuppression reduction when administering a booster dose of the BNT162b2 mRNA SARS-CoV-2 vaccine in kidney transplant recipients without adequate humoral response following two vaccine doses: protocol for a randomised controlled trial (BECAME study)

Dafna Yahav[1,2*], Benaya Rozen-Zvi[2,3*], Tiki Mashraki[3,4], Alaa Atamna[1,2], Haim Ben-Zvi[2,5], Erez Bar-Haim[6], Ruth Rahamimov[2,3,4]

[1] Infectious Diseases Unit, Rabin Medical Center, Beilinson Campus, Petah-Tikva, Israel

[2] Sackler Faculty of Medicine, Tel Aviv University, Tel Aviv, Israel

[3] Department of Nephrology and Hypertension, Rabin Medical Center, Beilinson Campus, Petah-Tikva, Israel

[4] Department of Transplantation, Rabin Medical Center, Beilinson Campus, Petah-Tikva, Israel

[5] Clinical Microbiology Laboratory, Rabin Medical Center, Beilinson Hospital, Petah-Tikva, Israel

[6] Department of Biochemistry and Molecular Genetics, Israel Institute for Biological Research, 74100 Ness-Ziona, Israel.

[*] BRZ and DY contributed equally for this work

Corresponding author: Prof. Dafna Yahav, Infectious Diseases Unit, Rabin Medical Center, Beilinson Hospital, 39 Jabotinsky Road, Petah-Tikva, 49100, Israel. Phone: 972-3-9377512, Fax: 972-3-9376512. E-mail: dafna.yahav@gmail.com

Keywords: kidney transplant recipients; COVID-19; vaccine; immunosuppression reduction; randomized controlled trial

Word count:

For peer review only

Abstract

Introduction: Inadequate antibody response to mRNA SARS-CoV-2 vaccination has been described among kidney transplant recipients. Immunosuppression level and specifically, use of antimetabolite in the maintenance immunosuppressive regimen, are associated with inadequate response. In light of the severe consequences of COVID-19 in solid organ transplant recipients, we believe it is justified to examine new vaccination strategies in these patients.

Methods and analysis: BECAME is a single center, open label, investigator-initiated randomised controlled, superiority trial, aiming to compare immunosuppression reduction combined with a third BNT162b2 vaccine dose versus third dose alone. The primary outcome will be seropositivity rate against SARS-CoV-2. A sample size of 154 patients was calculated for the seropositivity endpoint assuming 25% seropositivity in the control group and 50% in the intervention group. A sample of participants per arm will be also tested for T-cell response. We also plan to perform a prospective observational study, evaluating seropositivity among ~350 kidney transplant recipients consenting to receive a third vaccine dose, who are not eligible for the randomised controlled trial.

Ethics and dissemination: The trial is approved by local ethics committee of Rabin medical center (RMC-0192- 21). All participants will be required to provide written informed consent. Results of this trial will be published; trial data will be available. Protocol amendments will be submitted to the local ethics committee.

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Registration: The study is registered in clinicaltrials.gov (NCT04961229).

Article summary

Strengths and limitations of this study

- This randomised controlled trial addresses a question of crucial importance for organ transplant recipients during the COVID-19 pandemic

- Allocation concealment will reduce the risk of bias, though blinding will not be possible

- Antibody level measures for all participants at several timepoints, and partial sampling for T-cell response will provide an overview on protectivity of vaccine

- Currently, no neutralizing antibody testing is planned, limiting the evaluation of protective effect of the vaccine

- The study is open-label, however the primary outcome is an objective laboratory test result

For peer review only

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Introduction

COVID-19 outbreak has great impact on solid organ transplant recipients. Mortality rates among kidney transplant recipients have been reported between 13 to 50%, with high rates of complications, including acute kidney injury (AKI) in 30-89% of hospitalized patients. (1) Severe consequences of COVID-19 were also demonstrated among vaccinated kidney transplant recipient who were infected with SARS-CoV-2, with considerable mortality. (2)

The Pfizer mRNA-based BNT162b2 vaccine, the first vaccine approved by the FDA against SARS-CoV-2 infection, has been delivered to over 5 million people in Israel since December 2020. Immunocompromised patients were excluded from the phase III trial evaluating this vaccine, and thus, the efficacy and safety of the vaccine in this patient population are currently not well studied. (3) A large study from Israel has validated the effectiveness of this vaccine in the general population, (4) however the number of vaccinated solid organ transplant recipients in this cohort (n=435) was too small for performing evaluation of vaccine effectiveness for this sub-group. (5)

The American Society of Transplantation and other transplantation societies in the world have recommended vaccinating transplant candidates and recipients against

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

SARS-CoV-2 despite lack of data regarding efficacy in these populations, based on encouraging clinical results in other populations. (6)

Early phase 1/2 studies showed that BNT162b2 elicited strong antibody response in healthy adults. The titer of the neutralizing antibodies increased with dose and also increased after the second injection in comparison to the first. (7,8) High rates of antibody response to two doses of the vaccine were also documented in healthy population, accompanied by a distinct Th1 type T cell response. (9) While the role of neutralizing antibodies in protection from SARS-CoV-2 was demonstrated, it is expected that a steady T cell response has a central role against SARS-CoV-2 infection. (10) Solid organ recipient are expected to gain lower immune response to vaccinations with varying effectiveness between different vaccines and different transplanted organ populations. (11) Broad impairments in both humoral and cellular response to mRNA vaccines have been reported in kidney transplant recipients. Several studies from Israel demonstrated low rates of antibody response to the BNT162b2 vaccine among solid organ transplant recipients, including 36% seropositivity among 308 kidney transplant recipients 2-4 weeks after the second vaccine dose; (12) 47% seropositivity among 80 liver transplant recipients; (13) 49% among 37 heart transplant recipients; (14) and 18% among 168 lung transplant recipients. (15) Most demonstrated an association between mycophenolic acid dose and calcineurin inhibitors (CNI) blood levels and antibody response. A large study evaluating response to either BNT162b2 vaccine or the mRNA-1273 (Moderna) vaccine among 658 organ transplant recipients also demonstrated low seropositivity rates of 54% a median of 29 days after two vaccine doses. (16) In addition, diminished generation of plasmablasts and memory B cells in response to mRNA vaccine among kidney transplant recipients were reported. (17) Impairments in T cell

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

response were also described, with high rates of spike-specific T helper cell response, however reduced magnitude of response, as well as limited effector cytokine production. (18,19) Recent studies demonstrate improve humoral and cellular responses following third booster mRNA vaccine dose among transplant recipients. Yet, a considerable portion of these patients remained seronegative. (20–22) We plan a randomised controlled trial aiming to evaluate whether a third booster dose of mRNA SARS-CoV-2 vaccine BNT162b2, with or without immunosuppression reduction, improves the humoral response in kidney transplant recipients.

Methods and analysis

Study hypothesis and aims

We aim to evaluate the effect of a third mRNA vaccine dose with and without immunosuppression reduction on rates of seropositivity among kidney transplant recipients. We hypothesize that immunosuppression reduction combined with a third dose will demonstrate superiority over a third dose alone in terms of seropositivity rates.

Study design:

The study is a single center, randomised controlled, superiority, open-label trial, with an observational cohort as below:

- Randomised controlled trial (2 arms):

Third dose of BNT162b2 vaccine with or without reduction of mycophenolic acid dose (see below).

Observational arm: a third vaccine dose with no change in immunosuppression for patients that are excluded from the randomised trial (see exclusion criteria)

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Setting

The study will be conducted at Rabin Medical Center in Israel, in the transplantation follow-up clinic.

Study population:

We will include in both the RCT and observational study adult (age≥18 years) kidney transplant recipients that received two doses of BNT162b2 vaccine at least 3 weeks prior to enrollment, and were seronegative (IgG against the spike protein of SARS-CoV-2 below 50 AU/ml) at least two weeks after the second vaccine dose.

Additional inclusion criteria for the RCT:

- Recipients treated with three anti-rejection medications including: prednisone, tacrolimus, mycophenolate mofetil or mycophenolic acid.

Patients with any dosage of mycophenolic acid, mycophenolate mofetil, and prednisone will be eligible for inclusion. Regarding tacrolimus, trough blood levels 5-10 nGr/ml will be required for inclusion. Lower or higher doses will have to be adjusted before re-considering for inclusion.

Exclusion criteria for both RCT and observational part:

- Past infection with SARS-CoV-2
- Pregnancy
- Age below 18 years
- Active infection

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Additional exclusion criteria for RCT only:

- Recipients at a high risk for acute or chronic humoral rejection including:

  - Recipients with positive panel-reactive antibody (PRA) (any positive value) at any time before or after transplantation
  - Recipients that had an acute rejection in the last year
  - Recipients less than 6 months after transplantation
  - Recipients that are considered at high risk for rejection according to the primary care nephrologist
  - Recipients taking less than 3 anti-rejection medications
  - Recipients currently treated with mTOR inhibitors (everolimus, sirolimus) and/or azathioprine
  - Recipients treated with plasmapheresis in the previous 3 months
  - Recipients treated with eculizumab in the last year
  - Recipient treated with IVIG in the previous 3 months
  - Recipient treated with rituximab in the previous 6 months

Patients can withdraw from the study at any time.

Patient randomisation

Patients will be randomised to immunosuppression reduction versus no immunosuppression reduction in a 1:1 ratio. Randomisation will be performed using a computer-generated list of random numbers that will be allocated centrally through a web site.

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Interventions:

All recipients more than 6 months post transplantation and at least 3 weeks following second vaccine dose will be approached and invited to a first study visit (for trial flow see Appendix part A).

At first visit:

- Signed informed consent will be obtained from participants willing to participate by study investigators who routinely work in the transplantation clinic.

- Anti-spike antibody response will be assessed using SARS-CoV-2 IgG II Quant (Abbott©) assay. (23) Participants who have a documented seronegative test in the last 6 weeks will not be tested again.

- Tacrolimus levels will be obtained

Participants will be invited for an additional visit once negative serology result will be reported, within 7 days of test collection. At this second visit all participants who gave informed consent to participate in either the prospective non-randomised study or RCT will receive a single vaccine dose.

In addition, participants in the RCT will be randomised into two groups:

1) Third booster dose of BNT162b2 (one standard dose) with no change in immunosuppression protocol

2) Third booster dose of BNT162b2 (one standard dose) with immunosuppression reduction according to protocol (mycophenolic temporary cessation 4 days before (5 half-lives) and one week (expected antibody response) after vaccination (to allow for antibody response).

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Patients who will test seronegative will be informed by the study coordinator by phone in which study arm they will be participating and receive instructions for immunosuppression reduction both during the phone call and by written instructions provided to each patient during the first visit (see Appendix part B). Participants in the observational study will receive a third vaccine standard dose, without any change in immunosuppression (beyond routine care)

For all groups:

- Antibodies titer against spike protein will be evaluated at 2 weeks and 3, 6, 12 months after the third vaccine dose

- T-cell response will be evaluated for a small, randomly selected, subset of patients in each group (estimated 20 patients per arm) before booster dose, at 2 weeks after booster dose, and at 3 months. For T cell response quantification, peripheral blood mononuclear cell (PBMC) will be stimulated for 24 hours with spike protein and secreted interferon-gamma (IFNg) will be measured by ELISA (using the SARS-CoV-2 IFNg release assay EUROIMMUN, Lübeck, Germany). (24)

- Follow-up for adverse events, rejection, or SARS-CoV-2 infection will be performed at 2 weeks and at three, 6 and 12 months post third vaccination dose

Outcomes:

The primary outcome of both RCT and observational study will be humoral response against SARS-CoV-2, defined as anti-spike protein titer above 50 AU/ml at 2 weeks post vaccination

Secondary outcomes

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

- Humoral response against SARS-CoV-2 at 3-, 6-, and 12-months post vaccination
- Humoral response of > 4160 AU/mL, corresponding with antibody neutralization (25,26)
- Log transformed titer of anti S protein at 2 weeks and 3, 6, and 12 months
- Log transformed change in anti-spike protein titer at 2 weeks and 3, 6, and 12 months
- Adverse events to booster dose at 2 weeks post vaccine. Severity of adverse events will be assessed using CTCAE v4.0 criteria
- Acute rejection of the allograft at 2 weeks, 3,6, and 12 months (documented by biopsy)
- SARS-CoV-2 infection during the follow up period (until 12 months following vaccine)
- Other viral reactivation during the follow up period (VZV, CMV), tested according to clinical suspicion)
- Number of hospitalizations until 12 months

Due to technical limitations, we do not plan biobanking in this study.

Monitoring outcomes:

Participants in both RCT and observational study will be invited for a clinic visit at 2 weeks, 3, 6, and 12 months at the post-transplant follow up clinic at Rabin Medical Center. During their visit they will be questioned for adverse events (see Appendix part C for questionnaire); blood and urine tests will be checked as in the usual post-transplant follow ups for creatinine levels, any increase in creatinine of more than 20% will be promptly followed by kidney biopsy (after excluding other reasons), urine protein or microalbumin, tacrolimus levels and anti-spike protein antibodies.

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Data will be entered to the REDCap software, and the primary investigator will be responsible to check for timely data entry, missing data and suspected faulty data.

Predefined subgroup analysis:

- Recipients 65 years and older

Sample size:

The trial is designed to demonstrate superiority of immunosuppression reduction in terms of seropositivity rates. For the seropositivity endpoint we calculated a sample size of 77 participants per arm, assuming a 25% seropositivity rate in the control group and 50% in the intervention group, (22) with a 1-sided hypothesis with 2.5% α-risk and 90% power.

For the observational study we assume ~350 participants.

Stopping rules:

We plan to assess for acute rejection episodes following first 20 participants allocated to the immunosuppression reduction arm. If an acute rejection episode will be demonstrated within 2 weeks follow up from the booster dose – the study will be terminated. Acute rejection will be defined as 20% increase from baseline creatinine with no other plausible explanation. Need for histological proof of rejection will be discussed case by case by an independent safety monitoring committee. In addition, interim analyses will be performed after the recruitment of each 50 participants. At each analysis, rejection episodes will be evaluated and rates of seropositivity will be assessed. An independent safety monitoring committee of three nephrologists or ID physicians, specialist in managing transplant patients, will meet after each interim analysis to decide whether the study should be terminated.

Statistical analysis

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Fisher's exact test will be performed for the proportion of participants with seropositivity, and comparisons of anti-S antibody titers will be tested with the Mann-Whitney U test, and 95% confidence intervals (95% CI) will be calculated. IgG concentrations below threshold of detection will be given the lowest detectable value multiplied by 0.5. A p-value < 0.05 will be considered statistically significant. All data will be analyzed using SPSS (Version 27).

Patient and Public Involvement

No patient involved

Ethics and dissemination

In recent studies, no serious adverse events among transplant recipients receiving two mRNA vaccine doses were found. (12–15) Temporary reduction of immunosuppression during sepsis in kidney transplant recipients was not associated with an increased risk of rejection or long-term graft failure. (27) The trial is approved by local ethics committee of Rabin medical center (RMC-0192- 21). All participants will be required to provide written informed consent.

This trial's results will be made available through publication. Patient data (de-identified) will be available for other researchers by request. This will be considered for researchers presenting a methodologically adequate protocol, ethical approval, and signing data transfer agreement.

Study timeline

Patients will be recruited during September-November 2021, follow up will be for 12 months. Analysis and writing of the manuscript are planned until June 2023. Current status of the study – still not recruiting.

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Author contributions: All authors contributed to conception, design, trial management and planned data analysis. BRZ, RR, and DY contributed to the conception of the trial; BRZ, RR, DY, AA, EBH, and TM contributed to the trial design; BRZ, RR, DY, HBZ and AA contributed to trial database and randomisation site design. BRZ, DY and RR wrote the first draft of the manuscript. All authors revised the protocol and approved the final manuscript.

Funding: no external funding. The vaccine doses were provided by the Israeli ministry of health.

Competing interests – all authors declare no competing interests

For peer review only

References:

1. Azzi Y, Bartash R, Scalea J, Loarte-Campos P, Akalin E. COVID-19 and Solid Organ Transplantation: A Review Article. Transplantation. 2021 Jan 1;105(1):37–55.

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

2. Tau N, Yahav D, Schneider S, Rozen-Zvi B, Sneineh MA, Rahamimov R. Severe consequences of COVID-19 infection among vaccinated kidney transplant recipients. American Journal of Transplantation [Internet]. [cited 2021 May 28];n/a(n/a). Available from: https://onlinelibrary.wiley.com/doi/abs/10.1111/ajt.16700

3. 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 Dec 31;383(27):2603–15.

4. Dagan N, Barda N, Kepten E, Miron O, Perchik S, Katz MA, et al. BNT162b2 mRNA Covid-19 Vaccine in a Nationwide Mass Vaccination Setting. N Engl J Med. 2021 Feb 24;

5. Roest S, Hoek RAS, Manintveld OC. BNT162b2 mRNA Covid-19 Vaccine in a Nationwide Mass Vaccination Setting. N Engl J Med. 2021 Apr 21;384(20).

6. COVID-19 Vaccine Information for Transplant Recipients [Internet]. American Society of Transplantation. 2021 [cited 2021 May 15]. Available from: https://www.myast.org/covid-19-vaccine-information-transplant-recipients

7. 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 Dec 17;383(25):2439–50.

8. Sahin U, Muik A, Derhovanessian E, Vogler I, Kranz LM, Vormehr M, et al. COVID-19 vaccine BNT162b1 elicits human antibody and TH1 T cell responses. Nature. 2020 Oct;586(7830):594–9.

9. Favresse J, Bayart J-L, Mullier F, Dogné J-M, Closset M, Douxfils J. Early antibody response in healthcare professionals after two doses of SARS-CoV-2 mRNA vaccine (BNT162b2). Clin Microbiol Infect. 2021 May 8;

10. Jin P, Li J, Pan H, Wu Y, Zhu F. Immunological surrogate endpoints of COVID-2019 vaccines: the evidence we have versus the evidence we need. Signal Transduction and Targeted Therapy. 2021 Feb 2;6(1):1–6.

11. Eckerle I, Rosenberger KD, Zwahlen M, Junghanss T. Serologic vaccination response after solid organ transplantation: a systematic review. PLoS One. 2013;8(2):e56974.

12. Rozen-Zvi B, Yahav D, Agur T, Zingerman B, Ben-Zvi H, Atamna A, et al. Antibody response to mRNA SARS-CoV-2 vaccine among kidney transplant recipients - Prospective cohort study. Clin Microbiol Infect. 2021 May 3;

13. Rabinowich L, Grupper A, Baruch R, Ben-Yehoyada M, Halperin T, Turner D, et al. Low immunogenicity to SARS-CoV-2 vaccination among liver transplant recipients. J Hepatol. 2021 Apr 20;

14. Itzhaki Ben Zadok O, Shaul AA, Ben-Avraham B, Yaari V, Ben Zvi H, Shostak Y, et al. Immunogenicity of the BNT162b2 mRNA Vaccine in Heart

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Transplanted Patients-A Prospective Cohort Study. Eur J Heart Fail. 2021 Apr 29;

15. Shostak Y, Shafran N, Heching M, Rosengarten D, Shtraichman O, Shitenberg D, et al. Early humoral response among lung transplant recipients vaccinated with BNT162b2 vaccine. Lancet Respir Med. 2021 May 5;

16. Boyarsky BJ, Werbel WA, Avery RK, Tobian AAR, Massie AB, Segev DL, et al. Antibody Response to 2-Dose SARS-CoV-2 mRNA Vaccine Series in Solid Organ Transplant Recipients. JAMA. 2021 May 5;

17. Rincon-Arevalo H, Choi M, Stefanski A-L, Halleck F, Weber U, Szelinski F, et al. Impaired humoral immunity to SARS-CoV-2 BNT162b2 vaccine in kidney transplant recipients and dialysis patients. Sci Immunol. 2021 Jun 15;6(60):eabj1031.

18. Sattler A, Schrezenmeier E, Weber UA, Potekhin A, Bachmann F, Straub-Hohenbleicher H, et al. Impaired humoral and cellular immunity after SARS-CoV-2 BNT162b2 (tozinameran) prime-boost vaccination in kidney transplant recipients. J Clin Invest. 2021 Jul 15;131(14):150175.

19. Stumpf J, Siepmann T, Lindner T, Karger C, Schwöbel J, Anders L, et al. Humoral and cellular immunity to SARS-CoV-2 vaccination in renal transplant versus dialysis patients: A prospective, multicenter observational study using mRNA-1273 or BNT162b2 mRNA vaccine. The Lancet Regional Health – Europe [Internet]. 2021 Jul 22 [cited 2021 Aug 31];0(0). Available from: https://www.thelancet.com/journals/lanepe/article/PIIS2666-7762(21)00155-1/abstract

20. Stumpf J, Tonnus W, Paliege A, Rettig R, Steglich A, Gembardt F, et al. Cellular And Humoral Immune Responses after Three Doses of BNT162b2 mRNA SARS-Cov-2 Vaccine in Kidney Transplant. Transplantation. 2021 Jul 22;

21. Kamar N, Abravanel F, Marion O, Couat C, Izopet J, Del Bello A. Three Doses of an mRNA Covid-19 Vaccine in Solid-Organ Transplant Recipients. New England Journal of Medicine. 2021 Aug 12;385(7):661–2.

22. Hall VG, Ferreira VH, Ku T, Ierullo M, Majchrzak-Kita B, Chaparro C, et al. Randomized Trial of a Third Dose of mRNA-1273 Vaccine in Transplant Recipients. New England Journal of Medicine. 2021 Aug 11;0(0):null.

23. Abbott laboratories. SARS-CoV-2 IgG II Quant [Internet]. 2020 [cited 2021 Feb 24]. Available from: https://www.corelaboratory.abbott/int/en/offerings/segments/infectious-disease/sars-cov-2

24. Huzly D, Panning M, Smely F, Enders M, Komp J, Steinmann D. Validation and performance evaluation of a novel interferon-γ release assay for the detection of SARS-CoV-2 specific T-cell response [Internet]. 2021 Jul [cited 2021 Sep 2] p.

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

2021.07.17.21260316. Available from: https://www.medrxiv.org/content/10.1101/2021.07.17.21260316v1

25. Ebinger JE, Fert-Bober J, Printsev I, Wu M, Sun N, Prostko JC, et al. Antibody responses to the BNT162b2 mRNA vaccine in individuals previously infected with SARS-CoV-2. Nat Med. 2021 Jun;27(6):981–4.

26. Redjoul R, Le Bouter A, Beckerich F, Fourati S, Maury S. Antibody response after second BNT162b2 dose in allogeneic HSCT recipients. Lancet. 2021 Jul 24;398(10297):298–9.

27. Yahav D, Sulimani O, Green H, Margalit I, Ben-Zvi H, Mor E, et al. Immunosuppression reduction in kidney transplant recipients during bacterial infection-A retrospective study. Clin Transplant. 2019 Oct;33(10):e13707.

For peer review only

Appendix

Part A: Trial flow

Part B: Instructions for study participant - immunosuppression management

Part C: Questionnaire for adverse events

A. Trial flow

Invitation for a first study visit:

Kidney transplant recipients > 6 months post transplant; and ≥ 3 weeks post 2nd vaccine

First visit: informed consent; anti-spike antibody test; tacrolimus levels; written instruction for the following steps provided (See Appendix part B)

Between first and second visit:

Negative serology -> randomization of participants in the randomized controlled trial (RCT) to no intervention or immunosuppression reduction -> study coordinator instructions for RCT participants by phone*

Second visit: vaccination for all participants (observational and RCT); further instructions for RCT participants on the intervention

Follow up since vaccination:

At 2 weeks: serology all, T-cell assay for subset, adverse events, other evaluations **

At 3 months: serology all, T-cell assay for subset, other evaluations **

At 6 months: serology all, T-cell assay for subset, other evaluations **

At 12 months: serology all, T-cell assay for subset, other evaluations **

For peer review only

* No intervention arm – do not change immunosuppressive regimen;

immunosuppression reduction arm – stop mycophenolic 4 days before second visit (vaccination) and resume one week after vaccination

** Evaluation for kidney rejection (creatinine at each visit) and SARS-CoV-2 infection (patient interview and data from computerized records)

B. Instructions for study participant - immunosuppression management

Dear participant,

You participate in this randomised controlled trial to evaluate the safety and effectiveness of a booster (third) dose of mRNA SARS-CoV-2 BNT162b2 vaccine with or without immunosuppression reduction.

In this study you will be randomised by a computer to be in one of two arms of the study. Following randomization, you will be informed by the study coordinator, who is a nurse in the transplantation clinic, to which arm you belong:

Arm 1 – immunosuppression continuation: continue with your medications as usual before and after the third vaccine injection date.

Arm 2 - immunosuppression reduction: stop taking Myfortic or Cellcept from 4 days before your scheduled vaccine date and 7 days after. Please continue with all your other medications as usual. 4 days before the vaccine – if the vaccine is on Thursday, you stop Myfortic or Cellcept on Sunday and resume on next Thursday; if the vaccine is on Wednesday, you stop Myfortic or Cellcept on Saturday and resume on next Wednesday.

If you have any questions, don't hesitate to contact the study team and/or the transplantation clinic.

For peer review only

C. Questionnaire for adverse events

Did you have any of these symptoms after the first vaccine injection?

1st vaccine date:	yes	No	Severity
Local pain on injection site			
Muscle pain			
headache			
Fatigue			
Allergic reaction			
Fever			
Lymph node enlargement			
Need of medical treatment			
Need of hospitalization			

Did you have any of these symptoms after the second vaccine injection?

2nd vaccine date:	yes	No	Severity
Local pain on injection site			
Muscle pain			
headache			
Fatigue			
Allergic reaction			
Fever			
Lymph node enlargement			
Need of medical treatment			
Need of hospitalization			

For peer review only

For peer review only

SPIRIT
STANDARD PROTOCOL ITEMS: RECOMMENDATIONS FOR INTERVENTIONAL TRIALS

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description
Administrative information		
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym – page 1
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry – page 1,3
	2b	All items from the World Health Organization Trial Registration Data Set – the trial we be registered in the NIH registration data set and the Israeli ministry of health data set – page 3
Protocol version	3	Date and version identifier – 10-Sep-2021, Version 2
Funding	4	Sources and types of financial, material, and other support – page 14
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors – page 1, page 14
	5b	Name and contact information for the trial sponsor – no sponsor
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities – not applicable
	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee) – page 13
Introduction		
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention – page 4-6
	6b	Explanation for choice of comparators - page 4-6
Objectives	7	Specific objectives or hypotheses – page 6

For peer review only

Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory) – page 6-11
Methods: Participants, interventions, and outcomes		
Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained – page 6
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists) – page 6-7
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered – page 8
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease) - page 8
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests) – page 9
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial – appendix
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended – page 10-11
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure) – page 8-11
Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations – page 12
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size – page 12-13
Methods: Assignment of interventions (for controlled trials)		
Allocation:		

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

BMJ Open: first published as 10.1136/bmjopen-2012-001570 on 8 August 2012. Downloaded from http://bmjopen.bmj.com/ on April 19, 2024 by guest. Protected by copyright.

Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions – page 8
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned – page 8
Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions – page 8
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how – page 8
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial – not applicable, no blinding
Methods: Data collection, management, and analysis		
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol – page 11
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols – page 11
Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol – page 11
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol – page 12-13
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses) – page 12-13
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation) – page 12-13

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

BMJ Open: first published as 10.1136/bmjopen-2012-002055 on 25 October 2012. Downloaded from http://bmjopen.bmj.com/ on April 19, 2024 by guest. Protected by copyright.

Methods: Monitoring

Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed – page 12
	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial – page 12
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct - appendix
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor – not applicable

Ethics and dissemination

Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval – page 2, 13
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators) – page 2
Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32) – page 8
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable – not applicable
Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial – page 11
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site – page 13
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators – page 13
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation – not applicable

peer review only

For peer review only - http://bmjopen.bmj.com/site/about/guidelines.xhtml

Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions – page 13
	31b	Authorship eligibility guidelines and any intended use of professional writers – page 13
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code – page 13
Appendices		
Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates - appendix
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable- not applicable

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.

For peer review only