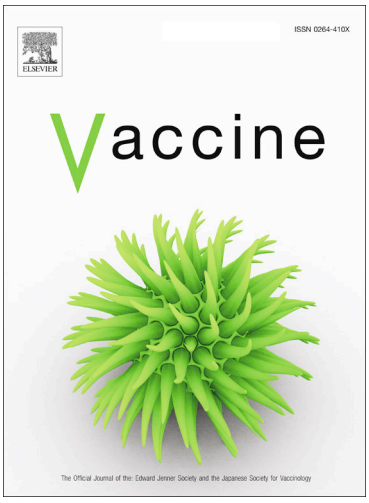
Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Journal Pre-proofs



COVID-19 booster vaccination during pregnancy enhances maternal binding and neutralizing antibody responses and transplacental antibody transfer to the newborn

Flor M. Munoz, Christine M. Posavad, Barbra A. Richardson, Martina L. Badell, Katherine E. Bunge, Mark J. Mulligan, Lalitha Parameswaran, Clifton W. Kelly, Courtney Olson-Chen, Richard M. Novak, Rebecca C. Brady, Marcela F. Pasetti, Emily A. Defranco, Jeffrey S. Gerber, Ms. Mallory C. Shriver, Mehul S. Suthar, Rhea N. Coler, Bryan J. Berube, Ms. So Hee Kim, Jeanna M. Piper, Ms. Ashley M. Miller, Cristina V. Cardemil, Kathleen M. Neuzil, Richard H. Beigi, DMID 21-0004 Study Group,

PII: S0264-410X(23)00698-9
DOI: https://doi.org/10.1016/j.vaccine.2023.06.032
Reference: JVAC 25064

To appear in: *Vaccine*

Received Date: 20 January 2023
Revised Date: 13 March 2023
Accepted Date: 6 June 2023

Please cite this article as: F.M. Munoz, C.M. Posavad, B.A. Richardson, M.L. Badell, K.E. Bunge, M.J. Mulligan, L. Parameswaran, C.W. Kelly, C. Olson-Chen, R.M. Novak, R.C. Brady, M.F. Pasetti, E.A. Defranco, J.S. Gerber, Ms. Mallory C. Shriver, M.S. Suthar, R.N. Coler, B.J. Berube, Ms. So Hee Kim, J.M. Piper, Ms. Ashley M. Miller, C.V. Cardemil, K.M. Neuzil, R.H. Beigi, DMID 21-0004 Study Group, COVID-19 booster vaccination during pregnancy enhances maternal binding and neutralizing antibody responses and transplacental antibody transfer to the newborn, *Vaccine* (2023), doi: https://doi.org/10.1016/j.vaccine.2023.06.032

COVID-19 booster vaccination during pregnancy enhances maternal binding and neutralizing antibody responses and transplacental antibody transfer to the newborn

Short Title: COVID-19 booster vaccine antibody responses during pregnancy

Flor M. MUNOZ, MD[1]; Christine M. POSAVAD, PhD[2]; Barbra A. RICHARDSON, PhD[2,3]; Martina L. BADELL, MD[4]; Katherine E. BUNGE, MD[5]; Mark J. MULLIGAN, MD[6]; Lalitha PARAMESWARAN, MD[6]; Clifton W. KELLY, MS[7]; Courtney OLSON-CHEN, MD[8]; Richard M. NOVAK, MD[9]; Rebecca C. BRADY, MD[10]; Marcela F. PASETTI, PhD[11]; Emily A. DEFRANCO, DO[10]; Jeffrey S. GERBER, MD[12]; Ms. Mallory C. SHRIVER, MSc[11]; Mehul S. SUTHAR, PhD[13]; Rhea N. COLER, PhD[14]; Bryan J. BERUBE, PhD[14]; Ms. So Hee KIM[7]; Jeanna M. PIPER, MD[15]; Ms. Ashley M. MILLER, MA[16]; Cristina V. CARDEMIL, MD[15]; Kathleen M. NEUZIL, MD[11]; Richard H. BEIGI, MD[5]; DMID 21-0004 Study Group[17]

[1]Departments of Pediatrics and Molecular Virology & Microbiology, Baylor College of Medicine, and Texas Children's Hospital, Houston, TX 77030, florm@bcm.edu

[2]Vaccine and Infectious Disease Division, Fred Hutchinson Cancer Center; Department of Laboratory Medicine and Pathology, University of Washington, Seattle, WA 98109, cposavad@fredhutch.org

[3]Departments of Biostatistics and Global Health, University of Washington, Vaccine and Infectious Disease and Public Health Sciences Divisions, Fred Hutchinson Cancer Center, Seattle, WA 98109, barbrar@uw.edu

[4]Department of Gynecology and Obstetrics, Division of Maternal Fetal Medicine, Emory University Hospital Midtown Perinatal Center, Atlanta, GA 30308, mbadell@emory.edu

[5]Department of Obstetrics, Gynecology and Reproductive Sciences, UPMC Magee-Womens Hospital, Pittsburgh, PA 15213, bungke@upmc.edu, beigrh@upmc.edu

[6]NYU Langone Vaccine Center and Division of Infectious Diseases and Immunology, Department of Medicine, NYU Grossman School of Medicine, New York, NY 10016, mark.mulligan@nyulangone.org, lalitha.parameswaran@nyulangone.org

[7]Statistical Center for HIV/AIDS Research and Prevention (SCHARP), Fred Hutchinson Cancer Center, Seattle, WA 98109, cwkelly@scharp.org, skim234@scharp.org

[8]Department of Obstetrics and Gynecology, University of Rochester, Rochester, NY 14642, courtney_olson-chen@urmc.rochester.edu

[9]Division of Infectious Diseases, University of Illinois, Chicago, IL 60612, rmnovak@uic.edu

[10]Cincinnati Children's Hospital Medical Center, Division of Infectious Diseases, University of Cincinnati College of Medicine, Cincinnati, OH 45229, rebecca.brady@cchmc.org, defranee@ucmail.uc.edu

[11]Center for Vaccine Development and Global Health, University of Maryland School of Medicine, Baltimore, MD 21201, mpasetti@som.umaryland.edu, mallory.shriver@som.umaryland.edu, kneuzil@som.umaryland.edu

[12]Division of Infectious Diseases, Children's Hospital of Philadelphia, University of Pennsylvania Perelman School of Medicine, Philadelphia, PA 19146, gerberj@chop.edu

[13]Emory Vaccine Center, Yerkes National Primate Research Center; Department of Pediatrics; Department of Microbiology and Immunology, Emory School of Medicine, Emory University, Atlanta, GA 30322, mehul.s.suthar@emory.edu

[14]Seattle Children's Research Institute, Center for Global Infectious Disease Research, Seattle, WA 98109, Rhea.Coler@seattlechildrens.org, Bryan.Berube@seattlechildrens.org

[15]National Institute of Allergy and Infectious Diseases, National Institutes of Health, Rockville, MD 20850, piperj@niaid.nih.gov, cristina.cardemil@nih.gov

[16]FHI 360, Durham, NC 27701, amiller@fhi360.org

[17]DMID 21-0004 Study Group

Corresponding Author:

Flor M. Munoz, MD

Baylor College of Medicine

Texas Children's Hospital

1102 Bates Ave, Suite 1150

Houston, TX 77030

Tel: 832-824-4371

Fax: 832-825-4347

Email: florm@bcm.edu

Source of Funding

Supported by the Infectious Diseases Clinical Research Consortium (IDCRC) through the National Institute of Allergy and Infectious Diseases, part of the National Institutes of Health (NIH), under award numbers UM1AI148684, UM1AI148575, UM1AI148372, UM1AI148452, UM1AI148576, UM1AI148574, UM1AI148450, UM1AI148685, UM1AI148689, and UM1AI148373. The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health.

Clinical Trial Information

This manuscript reflects Protocol Version 5.0, December 13, 2021; DMID 21-0004, ClinicalTrials.gov number NCT05031468. Recruitment began on July 6, 2021 and completed on January 31, 2022. https://clinicaltrials.gov/ct2/show/NCT05031468

JVAC-D-23-00123-R1

COVID-19 booster vaccination during pregnancy enhances maternal binding and neutralizing antibody responses and transplacental antibody transfer to the newborn

Short Title: COVID-19 booster vaccine antibody responses during pregnancy

Flor M. MUNOZ, MD[1]; Christine M. POSAVAD, PhD[2]; Barbra A. RICHARDSON, PhD[2,3]; Martina L. BADELL, MD[4]; Katherine E. BUNGE, MD[5]; Mark J. MULLIGAN, MD[6]; Lalitha PARAMESWARAN, MD[6]; Clifton W. KELLY, MS[7]; Courtney OLSON-CHEN, MD[8]; Richard M. NOVAK, MD[9]; Rebecca C. BRADY, MD[10]; Marcela F. PASETTI, PhD[11]; Emily A. DEFRANCO, DO[10]; Jeffrey S. GERBER, MD[12]; Ms. Mallory C. SHRIVER, MSc[11]; Mehul S. SUTHAR, PhD[13]; Rhea N. COLER, PhD[14]; Bryan J. BERUBE, PhD[14]; Ms. So Hee KIM[7]; Jeanna M. PIPER, MD[15]; Ms. Ashley M. MILLER, MA[16]; Cristina V. CARDEMIL, MD[15]; Kathleen M. NEUZIL, MD[11]; Richard H. BEIGI, MD[5]; DMID 21-0004 Study Group[17]

[1]Departments of Pediatrics and Molecular Virology & Microbiology, Baylor College of Medicine, and Texas Children's Hospital, Houston, TX 77030, florm@bcm.edu

[2]Vaccine and Infectious Disease Division, Fred Hutchinson Cancer Center; Department of Laboratory Medicine and Pathology, University of Washington, Seattle, WA 98109, cposavad@fredhutch.org

[3]Departments of Biostatistics and Global Health, University of Washington, Vaccine and Infectious Disease and Public Health Sciences Divisions, Fred Hutchinson Cancer Center, Seattle, WA 98109, barbrar@uw.edu

[4]Department of Gynecology and Obstetrics, Division of Maternal Fetal Medicine, Emory University Hospital Midtown Perinatal Center, Atlanta, GA 30308, mbadell@emory.edu

[5]Department of Obstetrics, Gynecology and Reproductive Sciences, UPMC Magee-Womens Hospital, Pittsburgh, PA 15213, bungke@upmc.edu, beigrh@upmc.edu

[6]NYU Langone Vaccine Center and Division of Infectious Diseases and Immunology, Department of Medicine, NYU Grossman School of Medicine, New York, NY 10016, mark.mulligan@nyulangone.org, lalitha.parameswaran@nyulangone.org

[7]Statistical Center for HIV/AIDS Research and Prevention (SCHARP), Fred Hutchinson Cancer Center, Seattle, WA 98109, cwkelly@scharp.org, skim234@scharp.org

[8]Department of Obstetrics and Gynecology, University of Rochester, Rochester, NY 14642, courtney_olson-chen@urmc.rochester.edu

[9]Division of Infectious Diseases, University of Illinois, Chicago, IL 60612, rmnovak@uic.edu

[10]Cincinnati Children's Hospital Medical Center, Division of Infectious Diseases, University of Cincinnati College of Medicine, Cincinnati, OH 45229, rebecca.brady@cchmc.org, defranee@ucmail.uc.edu

[11]Center for Vaccine Development and Global Health, University of Maryland School of Medicine, Baltimore, MD 21201, mpasetti@som.umaryland.edu, mallory.shriver@som.umaryland.edu, kneuzil@som.umaryland.edu

[12]Division of Infectious Diseases, Children's Hospital of Philadelphia, University of Pennsylvania Perelman School of Medicine, Philadelphia, PA 19146, gerberj@chop.edu

[13]Emory Vaccine Center, Yerkes National Primate Research Center; Department of Pediatrics; Department of Microbiology and Immunology, Emory School of Medicine, Emory University, Atlanta, GA 30322, mehul.s.suthar@emory.edu

[14]Seattle Children's Research Institute, Center for Global Infectious Disease Research, Seattle, WA 98109, Rhea.Coler@seattlechildrens.org, Bryan.Berube@seattlechildrens.org

[15]National Institute of Allergy and Infectious Diseases, National Institutes of Health, Rockville, MD 20850, piperj@niaid.nih.gov, cristina.cardemil@nih.gov

[16]FHI 360, Durham, NC 27701, amiller@fhi360.org

[17]DMID 21-0004 Study Group

Source of Funding

Supported by the Infectious Diseases Clinical Research Consortium (IDCRC) through the National Institute of Allergy and Infectious Diseases, part of the National Institutes of Health (NIH), under award numbers UM1AI148684, UM1AI148575, UM1AI148372, UM1AI148452, UM1AI148576, UM1AI148574, UM1AI148450, UM1AI148685, UM1AI148689, and UM1AI148373. The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health.

Clinical Trial Information

This manuscript reflects Protocol Version 5.0, December 13, 2021; DMID 21-0004, ClinicalTrials.gov number NCT05031468. Recruitment began on July 6, 2021 and completed on January 31, 2022. https://clinicaltrials.gov/ct2/show/NCT05031468

Corresponding Author:

Flor M. Munoz, MD

Baylor College of Medicine

Texas Children's Hospital

1102 Bates Ave, Suite 1150

Houston, TX 77030

Tel: 832-824-4371

Fax: 832-825-4347

Email: florm@bcm.edu

Highlights

183

184 • Data are needed to understand responses to primary and booster COVID-19 vaccinations
185 during pregnancy.
186 • COVID-19 mRNA vaccines during pregnancy elicited robust binding and neutralizing
187 antibody responses in mothers and newborns.
188 • Booster vaccination during pregnancy elicited significantly higher antibody levels in
189 mothers at delivery and cord blood than 2-dose primary vaccination, including against the
190 Delta and Omicron BA.1 variants.
191 • COVID-19 vaccines, including booster doses, should continue to be strongly
192 recommended during pregnancy.

Abstract:

The immune response to COVID-19 booster vaccinations during pregnancy for mothers and their newborns and the functional response of vaccine-induced antibodies against Omicron variants are not well characterized. We conducted a prospective, multicenter cohort study of participants vaccinated during pregnancy with primary or booster mRNA COVID-19 vaccines from July 2021 to January 2022 at 9 academic sites. We determined SARS-CoV-2 binding and live virus and pseudovirus neutralizing antibody (nAb) titers pre- and post-vaccination, and at delivery for both maternal and infant participants. Immune responses to ancestral and Omicron BA.1 SARS-CoV-2 strains were compared between primary and booster vaccine recipients in maternal sera at delivery and in cord blood, after adjusting for days since last vaccination.

A total of 240 participants received either Pfizer or Moderna mRNA vaccine during pregnancy (primary 2-dose series:167; booster dose:73). Booster vaccination resulted in significantly higher binding and nAb titers, including to the Omicron BA.1 variant, in maternal serum at delivery and in cord blood compared to a primary 2-dose series (range 0.44 to 0.88 $\log_{10}$ higher, $p<0.0001$ for all comparisons). Live virus nAb to Omicron BA.1 were present at delivery in 9% (GMT ID50 12.7) of Pfizer and 22% (GMT ID50 14.7) of Moderna primary series recipients, and in 73% (GMT ID50 60.2) of mRNA boosted participants ($p<0.0001$), although titers were significantly lower than to the D614G strain. Transplacental antibody transfer was efficient for all regimens with median transfer ratio range: 1.55-1.77 for IgG, 1.00-1.78 for live virus nAb and 1.79-2.36 for pseudovirus nAb. COVID-19 mRNA vaccination during pregnancy elicited robust immune responses in mothers and efficient transplacental antibody transfer to the newborn. A booster dose during pregnancy significantly increased maternal and cord blood binding and neutralizing antibody levels, including against Omicron BA.1. Findings support the use of a booster dose of COVID-19 vaccine during pregnancy.

Keywords: SARS-CoV-2, COVID-19, booster vaccination, pregnancy, neutralizing antibodies, transplacental antibody, newborn

List of Abbreviations

ACOG: American College of Obstetricians and Gynecologists

BAU/mL: Binding Antibody Units

BA.1: Omicron Variant

CDC: Centers for Disease Control and Prevention

CoVPN: COVID Prevention Network

DMID: Division of Microbiology and Infectious Diseases

DSMB: Data Safety monitoring Board

FRNT: Focus reduction neutralization titer

GMT: Geometric mean titer

IDCRC: Infectious Diseases Clinical Research Consortium

IgG: Immunoglobulin G

IQRs: Interquartile ranges

MSD: Meso Scale Discovery

nAb: Neutralizing antibodies

NIH: National Institutes of Health

RBD: Receptor binding domain

Wuhan-Hu-1: SARS-CoV-2 Spike protein

Spike: Full-length spike

U.S.: United States

VTEU: Vaccine Treatment and Evaluation Unit

WHO: World Health Organization

Introduction

Pregnant individuals are at increased risk of severe disease and obstetric complications after SARS-CoV-2 infection.[1,2,3,4] With the emergence of Omicron variants in late 2021, it has become apparent that infants younger than 6 months of age who become infected with SARS-CoV-2 are also at increased risk of hospitalization.[5,6] During the Omicron (BA.1, BA.4 and BA.5) variant waves, COVID-19 hospitalization rates for infants 0 through 5 months of age increased above rates in older children, adolescents and adults <65years old.[6] This is likely due to immunity in older age groups increasing through vaccination and prior infection, while young infants remain immunologically naïve and not eligible for vaccination until 6 months of age.

Importantly, COVID-19 vaccination during pregnancy is critical to mitigate the burden of disease for mothers and simultaneously represents the best approach to address this gap in protection for their infants.[7,8,9,10,11] Vaccine-induced antibodies transferred transplacentally to the infant reduces the risk of severe COVID-19 disease and hospitalization in infants in the first months of life.[12,13] In October 2021, pregnant individuals became eligible for booster vaccinations in the United States, yet the response to a booster dose and how it translates into neonatal antibody transfer and potential maternal and infant protection has not been well characterized.[14]

In this prospective cohort study, we measured the binding and neutralizing antibody responses to COVID-19 mRNA vaccines in pregnant participants and antibody levels in cord blood. We report the effect of primary series versus booster vaccination in pregnant mothers and on transplacental antibody levels in the newborn, and describe the functional immune response to Omicron in these groups.

Materials and Methods

This United States (U.S.)-based multicenter cohort study enrolled pregnant participants with and without medical comorbidities from July 6, 2021 to January 31, 2022. Eligible participants received a primary 2-dose series of Pfizer-BioNTech (Pfizer) or Moderna mRNA-1273 (Moderna) vaccine, or a monovalent booster dose of either vaccine, at any time during pregnancy as per current recommendations. Sera for antibody assays were derived from maternal blood collected pre- and post-vaccination (from 2 weeks post-vaccination to delivery), and maternal and cord blood collected at delivery. Maternal history of SARS-CoV-2 infection was collected at enrollment and at each study visit. Follow-up to 12 months post-delivery is ongoing and results will be reported separately. Detailed protocol and study procedures are described elsewhere (DMID 21-0004).[15]

Immunogenicity

Binding immunoglobulin G (IgG) levels to full-length Spike (Spike) and to the receptor binding domain (RBD) of Spike evaluated using the validated Meso Scale Discovery (MSD) V-PLEX® SARS-CoV-2 Panel 2 IgG assay (MSD #K15383U)[16] were bridged to international standards and reported as Binding Antibody Units (BAU/mL). SARS-CoV-2 neutralizing antibody (nAb) titers were evaluated by a pseudovirus neutralizing assay using a replication-incompetent lentivirus coding for luciferase and containing the SARS-CoV-2 Spike protein (Wuhan-Hu-1) in the viral

envelope (expressed as an IC50 value indicating the sample antibody titer capable of inhibiting viral entry and replication by 50%)[17], and a live virus focus reduction neutralization titer (FRNT) assay with viruses representing SARS-CoV-2 Spike mutation D614G and Delta and Omicron BA.1 variants [expressed as the serum inhibitory dilution required to achieve 50% neutralization (ID50)].[18] Detailed assay methods are in Supplementary Materials. Transplacental antibody transfer was evaluated by calculating the ratio of specific antibody levels in maternal and cord blood sera at the time of delivery.

Statistical Analysis

Medians and interquartile ranges (IQRs) for binding IgG, IC50 for pseudovirus nAb levels, and ID50 for live virus nAb levels were summarized by study visit and vaccine type. Differences in antibody levels between groups at delivery were tested using regression analyses controlling for days since last vaccine dose and prior self-reported SARS-CoV-2 infection or N-protein positive at delivery, as well as sensitivity analyses that were restricted to participants with vaccination in the same time interval between last vaccination and delivery.

Patient and Public Involvement

Patients or the public were not involved in the design, conduct, reporting, or dissemination plans of this research.

Results

This analysis describes 240 pregnant participants who gave birth and their newborns: 100 Pfizer (102 infants) and 67 Moderna (68 infants) 2-dose vaccine recipients, and 73 booster dose participants (75 infants) (Table 1). Booster doses were mostly homologous with the primary series (80.8%). The median age of participants was 34 years (range, 22-51). Participants completed their primary 2-dose series at a median of 17.1 weeks of gestation, while booster vaccination was received at a median of 28.6 weeks of gestation. Post-vaccination sera were collected at a median of 18.7 (range: 1.6-33.3) weeks following completion of the 2-dose series and 6.0 (range: 1.1-19.9) weeks following the booster dose. The interval (median weeks) between last vaccine dose and delivery was shorter for booster dose recipients (10.4) than primary 2-dose recipients (21.7). Overall, 14.4% of primary 2-dose recipients and 17.8% of booster dose recipients had self-reported SARS-CoV-2 infection or were N-protein positive up to delivery.

SARS-CoV-2 binding antibodies

Serum binding IgG to Spike and RBD were detected in all primary 2-dose and booster dose recipients at the post-vaccination and delivery visits, and in all cord blood samples (Figure 1). Significantly higher antibody levels were measured post-vaccination and at delivery in participants who received a booster vaccination during pregnancy compared to those who received only a primary 2-dose series (Table 2). At delivery, the geometric mean titer (GMT) of IgG to Spike in booster vaccine recipients was 2,201 BAU/mL (n=73), 9.3-fold higher than in those receiving two doses of Pfizer (GMT 236 BAU/mL, n=100), and 4.6-fold higher than in those receiving two doses of Moderna (479 BAU/mL, n=67) vaccines (Figure 1). Booster vaccination also elicited significantly higher levels of Spike IgG in cord blood, where the GMT

was 3,290 BAU/mL, 8.9-fold and 4.2-fold higher than in cord blood from those vaccinated with two doses of Pfizer (GMT 369 BAU/mL) or Moderna (GMT 792 BAU/mL), respectively (Figure 1). Similar trends were observed for RBD IgG in cord blood and at the post-vaccination visit to both Spike and RBD IgG (Figure 1, Table 2).

Overall, the booster group and their infants had ~0.6 $\log_{10}$ higher Spike and RBD IgG levels at delivery compared to the combined primary mRNA vaccine group (Pfizer and Moderna) after adjusting for days since last vaccination ($p<0.0001$) (Table 2). Sensitivity analyses showed similar results (data not shown).

SARS-CoV-2 neutralizing antibodies

Significantly higher live virus nAb titers to D614G were measured post-vaccination in pregnant participants who received a booster (GMT ID50 630.3) compared to those receiving a primary 2-dose series (GMT ID50 62.2 for Pfizer, 192.5 for Moderna) (Figure 2). High live virus nAb titers persisted at delivery and were detectable in 100% of boosted participants (GMT ID50 446.4), compared to 68% (GMT ID50 49.9) and 96% (GMT ID50 179.5) of participants receiving 2 doses of Pfizer or Moderna, respectively (Figure 2, Table 2, Table 3). While live virus nAb titers to Omicron BA.1 were present in only 9% (GMT ID50 12.7) of Pfizer and 22% (GMT ID50 14.7) of Moderna dosed participants at delivery, 73% (GMT ID50 60.2) of boosted participants had detectable live virus nAb titers to Omicron BA.1 ($p<0.0001$). Live virus nAb activity against Delta was intermediate between D614G and Omicron BA.1. Similarly, pseudovirus nAb titers were significantly higher post-vaccination and at delivery in those who received a booster (GMT IC50 708 and 513, respectively) compared to those who received a 2-dose series (GMT IC50 108 and 78, respectively, for Pfizer; 154 and 122, respectively, for Moderna) (Figure 1, Table 2, Table 3).

Live virus nAb titers to D614G were also significantly higher in cord blood in the booster group (GMT ID50 742.5) compared to participants receiving 2 doses of Pfizer (GMT ID50 77.1) or Moderna (GMT ID50 134.7) ($p<0.0001$) (Figure 2, Table 2, Table 3). Notably, live virus nAb titers to Omicron BA.1 were significantly higher in cord blood from the booster group (88% response rate, GMT ID50 109.2) compared to those receiving 2 doses of Pfizer (14% response rate, GMT ID50 13.3) or Moderna (22% response rate, GMT ID50 14.6) ($p<0.0001$). Sensitivity analyses showed similar results (data not shown). Significantly higher pseudovirus nAb titers were also observed in cord blood in the booster group (GMT IC50 941) compared to those receiving 2 doses of Pfizer (GMT IC50 159) or Moderna (GMT IC50 359) (Figure 1, Table 2, Table 3).

Transplacental antibody transfer

Efficient transplacental transfer (ratio ≥ 1.0) was observed with both primary and booster vaccination during pregnancy, with median antibody transfer ratios between 1.55 and 1.77 for binding IgG, between 1.00 and 1.78 for live virus nAb, and between 1.79 and 2.36 for pseudovirus nAb (Table 4).

Discussion

In this large, multicenter prospective cohort study, robust antibody responses to mRNA COVID-19 vaccines were detected in pregnant participants immunized across all gestational ages. The substantial increase in binding and neutralizing antibody titers measured in mothers and newborns at the time of delivery after a booster vaccination is a key finding which strongly supports the administration of booster doses during pregnancy. In addition to the D614G vaccine strain, this finding was also observed in the nAb response to Delta and Omicron BA.1 where levels were significantly higher in booster recipients at delivery and in cord blood compared to those receiving a primary 2-dose series only. This booster effect is particularly relevant given the persistence of Omicron subvariants in current phases of the pandemic. However, the nAb levels to Omicron BA.1 were significantly lower than to the vaccine-matched D614G variant, as expected and observed in non-pregnant populations.[19]

Additionally, as reported by other investigators, maternal binding IgG antibodies against both Spike and RBD SARS-CoV-2 proteins were efficiently transferred across the placenta and concentrated in the infant.[8,20.21] This latter finding is particularly important given high hospitalization rates among infants <6 months old during the Omicron BA.1 and BA.5 surges.[5,6] Transplacental antibody transfer is the key component of newborn protection from SARS-CoV-2 infection, and parallels demonstrated neonatal protection from other respiratory pathogens such as influenza and pertussis.[22,23] A recent study in Israel showed that IgG antibody titers in infants in the first few weeks of life correlated with SARS-CoV-2 IgG levels at birth.[23] Additionally, vaccine effectiveness studies have shown a reduction in hospitalization risk for the infant in the first few months of life following maternal vaccination during pregnancy.[12] Achieving higher antibody titers at birth could therefore provide protection against disease in the infant for a period of time until active vaccination. While an absolute correlate of protection is unknown, our study's findings taken in the context of these other studies supports the likelihood of infant protection during a period of high vulnerability and current gap in vaccine eligibility for infants less than 6 months old.

Our study findings demonstrate that both maternal and infant protection can be enhanced with booster vaccination during pregnancy. Similar to our results, Kugelman et al. reported significantly higher, IgG responses in pregnant women in Israel who received a booster dose of the Pfizer-BioNTech mRNA vaccine compared to a historical control group of pregnant women who received only two doses of vaccine in the same gestational age window recipients.[24] Our study extends these findings to include pseudo- and live nAb data, which confirm the functional activity and potential protective effect of these antibodies in the newborn.

While some differences were observed by vaccine type, the clinical significance of these findings is unknown, and additional research is necessary to further characterize the immune responses of both mRNA and non-mRNA SARS-CoV-2 vaccines administered during pregnancy. Furthermore, efforts leading towards understanding the effect on maternal and infant immunity beyond the neonatal period and the effect of factors such as the timing of maternal vaccination before and during pregnancy should continue. The optimal timing of vaccination and waning of immunity in pregnancy deserve further evaluation, particularly given the higher risk of severe maternal disease occurring in the third trimester of gestation.[12,25,26] In a large study or maternal infant dyads (N=402) also conducted in Israel, Rottenstreich A, et al., reported substantial waning of anti-S and anti RBD-specific IgG responses in pregnant women at delivery if they were vaccinated in the first trimester compared to the second or third trimester.[25] These

observations support booster vaccinations during pregnancy, particularly among mothers who might have completed their primary series prior to or early in pregnancy.[27]

Additional research is also needed to better elucidate the immune response following co-administration with other maternal vaccines, and the influence of maternal health status. Further, it remains important to characterize maternal immune responses to mixed vaccine platform regimens, next generation vaccines formulated against different SARS-CoV-2 variants, and the effect of administering subsequent booster vaccinations, possibly during every pregnancy.

Strengths and Limitations

Our multicenter prospective cohort study included diverse populations in various geographic regions of the U.S. and utilized an adaptive design which provided a unique opportunity for real world evaluation of the safety and immunogenicity of primary and booster vaccinations during pregnancy. We conducted a systematic, protocol-driven data and sample collection process encompassing periods of high transmission of SARS-CoV-2 variants with impact and relevance to pregnant women and infants. The study was strengthened by a central laboratory assessment of immune responses to vaccine strain as well as Delta and Omicron BA.1 variants, including binding and live and pseudovirus nAb.

Given the observational design of our study, the timing of vaccination during pregnancy was not pre-specified and the timing of sera collection post-vaccination was opportunistic. However, this investigation purposefully took a real-world scenario and inclusive enrollment approach and our analyses controlled for interval between vaccination and delivery, as well as prior maternal infection, to assess immune responses at the time of delivery. Thus, these data are generalizable and help delineate the potential impact of vaccination throughout pregnancy. Our results are limited to the evaluation of mRNA vaccines given current vaccine availability and recommendations in the U.S.

Conclusions

Pregnant women are appropriately included among the risk groups targeted for a bivalent booster dose of mRNA vaccines by the Centers for Disease Control and Prevention (CDC) and American College of Obstetrics and Gynecology (ACOG).[8,14] While an absolute correlate of protection against SARS-CoV-2 infection is still unknown, increases in antibody responses in non-pregnant adults are associated with protection from symptomatic severe COVID-19.[28] Higher binding and neutralizing antibody responses to vaccine and emerging strains of SARS-CoV-2 have the potential to provide protection to both mothers and infants during a period of risk and high vulnerability. Our study supports that COVID-19 vaccination, and particularly booster doses, should be strongly recommended during pregnancy for maternal and neonatal protection.

Acknowledgements

All the authors contributed to study design and have read and approved the final manuscript. We acknowledge Jeannie A. Murray, MS for contributions to the preparation and submission of this manuscript. We would like to thank study participants for their contributions.

Ethics Approval and Consent to Participate

Ethical approval of this protocol was received on May 28, 2021 by Vanderbilt University Medicine Center IRB, a single IRB as part of an NIH-funded consortium, IDCRC (IRB #210718). Written informed consent was obtained from each participant.

Data Sharing

Data collected for the study will be made available to others as a de-identified patient data set after finalization of clinical study report at the discretion of the IDCRC. Analyses of data, including data from staged analyses, will be available for presentation at scientific meetings and publication to inform the scientific community. If preliminary analyses are considered of public health importance or relevant to inform research, development, and implementation of SARS-CoV-2 vaccine in pregnancy, results may be shared with public health officials and partners to inform the global scientific community. The study will be conducted in accordance with the NIH Public Access Policy publication and data sharing policies and regulations. To request study data once complete, contact Flor M. Munoz, florm@bcm.edu.

Conflicts of Interest

F.M.M. is an investigator of pediatric studies of COVID-19 vaccines for Pfizer and for a pediatric remdesivir study conducted by Gilead Sciences, Inc; serves as investigator on projects supported by an NIH contract for a Vaccine Treatment and Evaluation Unit (VTEU), serves as member of the Data Safety monitoring Board (DSMB) for clinical trials conducted by Pfizer, Moderna, Meissa Vaccines, Virometix, and the NIH; and is a member of the American Academy of Pediatrics Section of Infectious Diseases (SOID), the Immunization Expert Group of the American College of Obstetrics and Gynecology (ACOG), and was co-Chair of the COVAX-CEPI Maternal Immunization Working Group.

K.M.N. is a member of the World Health Organization (WHO) Strategic Advisory Group of Experts on Immunization, serves as co-investigator on an NIH contract for a Vaccine Treatment and Evaluation Unit (VTEU), serves as Co-Chair of the NIH COVID Prevention Network (CoVPN), and served as an investigator for Phase I/II Pfizer COVID-19 vaccine grant, with a grant to the institution, but no salary support.

M.J.M. conducts laboratory research and clinical trials with contract funding for vaccines or MABs vs SARS-CoV-2 with Lilly, Pfizer, and Sanofi and receives personal fees for Scientific Advisory Board service from Merck, Meissa Vaccines, Inc. and Pfizer.

M.S.S. served as an advisor for Moderna (ended December 2021) and is currently serving as an advisor for Ocugen, Inc.

B.A.R. currently holds a position on a DSMB for clinical trials at Gilead Sciences, Inc.

R.C.B. at Cincinnati Children's Hospital receives research grant support for clinical trials from PATH, Astra Zeneca and Pfizer on which she serves as co-investigator.

B.B. owns shares in HDT Bio Corp.

J.S.G. receives research funds from NIH for Moderna KidCOVE study.

R.M.N. is a paid advisor to Gilead and an investigator on NIH-funded trials of Moderna, Pfizer and Janssen vaccines.

J.R-K is a medical speaker for Abbott Nutrition with the UIC team.

A.R.F. holds research grants from Pfizer, Janssen, Merck, Cyanvac, Biofire Diagnostics and serves on the DSMB for Novavax.

N.R. receives funds to conduct industry trials from Pfizer, Merck, and Sanofi-Pasteur and serves as a safety consultant for EMMES and ICON.

R.W.F. Jr., MD has received funds to conduct industry trials from Pfizer, Moderna and Astra Zeneca, serves on advisory boards for Merck, Sanofi-Pasteur, Johnson and Johnson and Seqirus and serves on an ICON-sponsored DSMB for a C difficile study.

All authors have completed relevant conflicts of interest in the Disclosure of Potential Conflicts of Interest section of the Authorship Form.

References:

1. Allotey J, Stallings E, Bonet M, et al. Clinical manifestations, risk factors, and maternal and perinatal outcomes of coronavirus disease 2019 in pregnancy: living systematic review and meta-analysis. *BMJ*. 2020 Sep 1;370:m3320. doi: 10.1136/bmj.m3320. PMID: 32873575; PMCID: PMC7459193.
2. Zambrano LD, Ellington S, Strid P, et al. Update: Characteristics of Symptomatic Women of Reproductive Age with Laboratory-Confirmed SARS-CoV-2 Infection by Pregnancy Status — United States, January 22–October 3, 2020. *MMWR Morb Mortal Wkly Rep* 2020;69:1641–1647. doi: http://dx.doi.org/10.15585/mmwr.mm6944e3.
3. Metz TD, Clifton RG, Hughes BL, et al. Association of SARS-CoV-2 Infection With Serious Maternal Morbidity and Mortality From Obstetric Complications. *JAMA*. 2022 Feb 22;327(8):748-759. doi: 10.1001/jama.2022.1190. PMID: 35129581; PMCID: PMC8822445.
4. Vousden N, Ramakrishnan R, Bunch K, et al. Management and implications of severe COVID-19 in pregnancy in the UK: data from the UK Obstetric Surveillance System national cohort. *Acta Obstet Gynecol Scand*. 2022 Apr;101(4):461-470. doi: 10.1111/aogs.14329. Epub 2022 Feb 25. PMID: 35213734; PMCID: PMC9111211.
5. Marks KJ, Whitaker M, Agathis NT, et al. Hospitalization of Infants and Children Aged 0-4 Years with Laboratory-Confirmed COVID-19- COVID-NET, 14 States, March 2020-February 2022. *MMWR Morb Mortal Wkly Rep* 2022;71:429-436. doi: http://dx.doi.org/10.15585/mmwr.mm7111e2.
6. Hamid S, Woodworth K, Pham H, et al. COVID-19 Associated Hospitalizations Among U.S. Infants Aged <6 Months – COVID-Net, 13 States, June 2021-August 2022. *MMWR Morb Mortal Wkly Rep* 2022;71:1442-1448. doi: http://dx.doi.org/10.15585/mmwr.mm7145a3.
7. Carlsen EØ, Magnus MC, Oakley L, et al. Association of COVID-19 Vaccination During Pregnancy With Incidence of SARS-CoV-2 Infection in Infants. *JAMA Intern Med*. 2022 Jun 1. doi: 10.1001/jamainternmed.2022.2442. Epub ahead of print. PMID: 35648413.
8. Centers for Disease Control (CDC). COVID-19 Vaccines While Pregnant or Breastfeeding [Internet]. Last Updated October 20, 2022. Available from: https://www.cdc.gov/coronavirus/2019-ncov/vaccines/recommendations/pregnancy.html.
9. Stock SJ, Carruthers J, Calvert C, et al. SARS-CoV-2 infection and COVID-19 vaccination rates in pregnant women in Scotland. *Nat Med*. 2022 Mar;28(3):504-512. doi: 10.1038/s41591-021-01666-2. Epub 2022 Jan 13. Erratum in: *Nat Med*. 2022 Feb 4;: PMID: 35027756; PMCID: PMC8938271.
10. Goldshtein I, Steinberg DM, Kuint J, et al. Association of BNT162b2 COVID-19 Vaccination During Pregnancy With Neonatal and Early Infant Outcomes. *JAMA Pediatr*. 2022 May 1;176(5):470-477. doi: 10.1001/jamapediatrics.2022.0001. PMID: 35142809; PMCID: PMC8832306.

11. Dagan N, Barda N, Biron-Shental T, et al. Effectiveness of the BNT162b2 mRNA COVID-19 vaccine in pregnancy. *Nat Med*. 2021 Oct;27(10):1693-1695. doi: 10.1038/s41591-021-01490-8. Epub 2021 Sep 7. PMID: 34493859.
12. Halasa NB, Olson SM, Staat MA, et al. Maternal Vaccination and Risk of Hospitalization for Covid-19 among Infants. *N Engl J Med*. 2022 Jul 14;387(2):109-119. doi: 10.1056/NEJMoa2204399. Epub 2022 Jun 22. PMID: 35731908; PMCID: PMC9342588.
13. Zerbo O, Ray GT, Fireman B, et al. Maternal SARS-CoV-2 Vaccination and Infant Protection Against SARS-CoV-2 During the First 6 Months of Life. *Res Sq* [Preprint]. 2022 Oct 18:rs.3.rs-2143552. doi: 10.21203/rs.3.rs-2143552/v1. PMID: 36299419; PMCID: PMC9603829.
14. The American College of Obstetricians and Gynecologists (ACOG). COVID-19 Vaccination Considerations for Obstetric-Gynecologic Care [Internet]. December 2020. Last Updated September 20, 2022. Available from: https://www.acog.org/clinical/clinical-guidance/practice-advisory/articles/2020/12/covid-19-vaccination-considerations-for-obstetric-gynecologic-care.
15. Munoz FM, Beigi RH, Posavad CM, et al. Multi-site observational maternal and infant COVID-19 vaccine study (MOMI-vax): a study protocol. *BMC Pregnancy Childbirth*. 2022 May 12;22(1):402. doi: 10.1186/s12884-022-04500-w. PMID: 35550037; PMCID: PMC9096328.
16. Gilbert PB, Montefiori DC, McDermott AB, et al. Immune correlates analysis of the mRNA-1273 COVID-19 vaccine efficacy clinical trial. *Science*. 2022 Jan 7;375(6576):43-50. doi: 10.1126/science.abm3425. Epub 2021 Nov 23. PMID: 34812653; PMCID: PMC9017870.
17. Larsen SE, Berube BJ, Pecor T, et al. Qualification of ELISA and neutralization methodologies to measure SARS-CoV-2 humoral immunity using human clinical samples. *J Immunol Methods*. 2021 Dec;499:113160. doi: 10.1016/j.jim.2021.113160. Epub 2021 Sep 30. PMID: 34599915; PMCID: PMC8481082
18. Edara VV, Pinsky BA, Suthar MS, et al. Infection and Vaccine-Induced Neutralizing-Antibody Responses to the SARS-CoV-2 B.1.617 Variants. *N Engl J Med*. Aug 12 2021;385(7):664-666. doi:10.1056/NEJMc2107799.
19. Belik M, Jalkanen P, Lundberg R, et al. Comparative analysis of COVID-19 vaccine responses and third booster dose-induced neutralizing antibodies against Delta and Omicron variants. *Nat Commun*. 2022 May 5;13(1):2476. doi: 10.1038/s41467-022-30162-5. PMID: 35513437; PMCID: PMC9072399.
20. Rottenstreich A, Zarbiv G, Oiknine-Djian E, Zigron R, Wolf DG, Porat S. Efficient Maternofetal Transplacental Transfer of Anti- Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) Spike Antibodies After Antenatal SARS-CoV-2 BNT162b2 Messenger RNA Vaccination. *Clin Infect Dis*. 2021 Nov 16;73(10):1909-1912. doi: 10.1093/cid/ciab266. PMID: 33822014; PMCID: PMC8083549.
21. Kugelman N, Nahshon C, Shaked-Mishan P, et al. Maternal and Neonatal SARS-CoV-2 Immunoglobulin G Antibody Levels at Delivery After Receipt of the BNT162b2

Messenger RNA COVID-19 Vaccine During the Second Trimester of Pregnancy. *JAMA Pediatr*. 2022 Mar 1;176(3):290-295. doi: 10.1001/jamapediatrics.2021.5683. PMID: 34932066; PMCID: PMC8693209.

22. Omer SB. Maternal Immunization. *N Engl J Med*. 2017 Mar 30;376(13):1256-1267. doi: 10.1056/NEJMra1509044. PMID: 28355514.

23. Kugelman N, Nahshon C, Shaked-Mishan P, et al. SARS-CoV-2 immunoglobulin G antibody levels in infants following messenger RNA COVID-19 vaccination during pregnancy. *Am J Obstet Gynecol*. 2022 Dec;227(6):911-913. doi: 10.1016/j.ajog.2022.07.016. Epub 2022 Jul 19. PMID: 35863460; PMCID: PMC9293368.

24. Kugelman N, Nahshon C, Shaked-Mishan P, et al. Maternal and Neonatal Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) Immunoglobulin G Levels After the Pfizer-BioNTech Booster Dose for Coronavirus Disease 2019 (COVID-19) Vaccination During the Second Trimester of Pregnancy. *Obstet Gynecol*. 2022 Aug 1;140(2):187-193. doi: 10.1097/AOG.0000000000004867. Epub 2022 May 27. PMID: 35852268.

25. Rottenstreich A, Zarbiv G, Oiknine-Djian E, et al. Timing of SARS-CoV-2 vaccination during the third trimester of pregnancy and transplacental antibody transfer: a prospective cohort study. *Clin Microbiol Infect*. 2022 Mar;28(3):419-425. doi: 10.1016/j.cmi.2021.10.003. Epub 2021 Nov 3. PMID: 34740773; PMCID: PMC8563509.

26. Rottenstreich A, Zarbiv G, Oiknine-Djian E, et al. The Effect of Gestational Age at BNT162b2 mRNA Vaccination on Maternal and Neonatal Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) Antibody Levels. *Clin Infect Dis*. 2022 Aug 24;75(1):e603-e610. doi: 10.1093/cid/ciac135. PMID: 35171998; PMCID: PMC8903394.

27. Faust JS, Rasmussen SA, Jamieson DJ. Pregnancy should be a condition eligible for additional doses of COVID-19 messenger RNA vaccines. *Am J Obstet Gynecol MFM*. 2022 Nov 9;5(2):100801. doi: 10.1016/j.ajogmf.2022.100801. Epub ahead of print. PMID: 36371035; PMCID: PMC9645060.

28. DeSilva MB, Mitchell PK, Klein NP, et al. Protection of 2 and 3 mRNA Vaccine Doses Against Severe Outcomes Among Adults Hospitalized with COVID-19 - VISION Network, August 2021 - March 2022. *J Infect Dis*. 2022 Nov 23:jiac458. doi: 10.1093/infdis/jiac458. Epub ahead of print. PMID: 36415904.

612 **Table 1. Study Participant Characteristics by Group Assigned at Enrollment**

613

Maternal Characteristic	**Pfizer-BioNTech n=100 Median (IQR) % (n)**	**Moderna n=67 Median (IQR) % (n)**	**Booster n=73 Median (IQR) % (n)**
Age, years	35 (31, 37)	34 (31, 37)	34 (31, 37)
Race			
Asian	12.0 (12)	4.5 (3)	9.6 (7)
Black/African American	10.0 (10)	16.4 (11)	1.4 (1)
White	75.0 (75)	73.1 (49)	83.6 (61)
Other	3.0 (3)	6.0 (4)	5.5 (4)
Hispanic or Latino	10.0 (10)	14.9 (10)	9.6 (7)
Vaccine Exposure up to Delivery			
2 doses Pfizer only	98.0 (98)		
2 doses Pfizer+Pfizer boost	2.0 (2)		76.7 (56)
2 doses Pfizer+Moderna boost	0.0 (0)		16.4 (12)
2 doses Moderna only		92.5 (62)	
2 doses Moderna+Moderna boost		6.0 (4)	4.1 (3)
2 doses Moderna+Pfizer boost		1.5 (1)	2.7 (2)
Weeks Between Last Dose and Post-Vaccination Visit	18.8 (14.6, 23.4)	18.3 (12.9, 23.6)	6.0 (3.3, 8.1)

Weeks Between Primary Series Completion and Delivery	21.9 (18.0, 26.4)	22.9 (18.3, 27.3)	46.7 (40.4, 50.9)
Weeks Between Last Dose and Delivery	21.6 (17.1, 25.7)	21.9 (15.4, 24.6)	10.4 (6.9, 14.1)
Gestational Age at First Dose During Pregnancy, weeks	13.1 (9.3, 17.9)	12.7 (7.6, 15.7)	-10.9 (-14.9, -4.6)
Gestational Age at Last Dose During Pregnancy, weeks	16.9 (13.4, 21.3)	17.1 (14.0, 23.7)	28.6 (24.9, 31.7)
Gestational Age at Delivery, weeks	39.3 (37.9, 40.1)	39.1 (38.7, 39.9)	39.1 (38.3, 39.6)
SARS-CoV-2 infection prior to or during study, up to delivery, self-reported OR N protein positive at delivery	15.0 (15)	13.4 (9)	17.8 (13)
Gravidity			
Primigravida	30.0 (30)	44.8 (30)	42.5 (31)
At least one prior pregnancy	70.0 (70)	55.2 (37)	57.5 (42)
At least two prior pregnancies	32.0 (32)	29.9 (20)	28.8 (21)
Maternal obstetric comorbidities			
Gestational diabetes	8.0 (8)	13.4 (9)	6.8 (5)
Hypertensive disorders	10.0 (10)	9.0 (6)	13.7 (10)
Obesity	9.0 (9)	10.4 (7)	13.7 (10)
Pre-eclampsia	0.0 (0)	0.0 (0)	2.7 (2)
Mode of delivery			
C-section	33.0 (33)	43.3 (29)	30.1 (22)
Vaginal	67.0 (67)	56.7 (38)	69.9 (51)

Infant Characteristic	**Pfizer-BioNTech n=102 % (n)**	**Moderna n=68 % (n)**	**Booster n=75 % (n)**
Preterm birth (<37 weeks)	6.9 (7)	7.4 (5)	14.7 (11)
Infant Birthweight (grams)	3370 (3040, 3640)	3340 (3030, 3560)	3280 (2860, 3540)

614

615 **Table 2. Differences in Antibody Levels Between Primary 2-Dose and Booster Groups at Delivery**

Lab Assessment	Sample	Primary GMT (95% CI)	Booster GMT (95% CI)	Mean Log_{10} Difference* (95% CI)	p-value
Spike IgG BAU/mL	Maternal Delivery	313.8 (260.7, 377.8)	2200.6 (1764.9, 2743.8)	0.55 (0.42, 0.68)	<0.0001
	Cord Blood	500.5 (424.1, 590.7)	3290.4 (2713.6, 3989.9)	0.57 (0.45, 0.69)	<0.0001
RBD IgG BAU/mL	Maternal Delivery	428.6 (351.9, 522.1)	3382.9 (2703.7, 4232.86)	0.59 (0.45, 0.73)	<0.0001
	Cord Blood	673.1 (564.5, 802.6)	5005.7 (4126.5, 6072.1)	0.62 (0.49, 0.75)	<0.0001
D614G live virus nAb ID50	Maternal Delivery	83.4 (65.4, 106.4)	446.4 (342.1, 582.5)	0.43 (0.26, 0.60)	<0.0001
	Cord Blood	96.4 (80.2, 115.9)	742.5 (588.5, 936.8)	0.65 (0.52, 0.79)	<0.0001
Delta live virus nAb ID50	Maternal Delivery	39.0 (30.9, 49.2)	390.2 (299.2, 508.9)	0.73 (0.57, 0.89)	<0.0001
	Cord Blood	52.1 (42.3, 64.2)	644.5 (507.5, 818.4)	0.84 (0.69, 0.99)	<0.0001
Omicron live virus nAb ID50	Maternal Delivery	13.5 (11.9, 15.3)	60.2 (43.6, 83.1)	0.52 (0.39, 0.64)	<0.0001

	Cord Blood	13.8 (12.2, 15.6)	109.2 (81.6, 146.1)	0.79 (0.67, 0.91)	<0.0001
Pseudotyped WT nAb IC50	Maternal Delivery	93.4 (77.3, 112.9)	512.5 (399.1, 658.3)	0.52 (0.36, 0.67)	<0.0001
	Cord Blood	220.8 (183.5, 265.7)	941.4 (735.7, 1204.7)	0.42 (0.27, 0.57)	<0.0001

*From regression analysis adjusted for days since last vaccination prior to delivery and prior self-reported SARS-CoV-2 infection or N protein positive at delivery.

Table 3. Differences in Response Rate for Neutralizing Antibodies Between Primary 2-Dose and Booster Groups at Delivery

Lab Assessment	Sample	Primary Response Rate*	Booster Response Rate*	Prevalence Ratio** (95% CI)	p-value
D614G live virus nAb	Maternal Delivery	79.0% (132/167)	100% (73/73)	1.10 (1.03, 1.18)	0.0039
	Cord Blood	93.5% (159/170)	100% (75/75)	1.04 (1.00, 1.09)	0.0553
Delta live virus nAb	Maternal Delivery	56.3% (94/167)	100% (73/73)	1.46 (1.28, 1.68)	<0.0001
	Cord Blood	71.8% (122/170)	100% (75/75)	1.22 (1.11, 1.33)	<0.0001
Omicron live virus nAb	Maternal Delivery	14.4% (24/167)	72.6% (53/73)	3.16 (2.13, 4.71)	<0.0001
	Cord Blood	17.1% (29/170)	88.0% (66/75)	3.71 (2.53, 5.44)	<0.0001
Pseudotyped WT nAb	Maternal Delivery	92.8% (155/167)	100% (73/73)	1.05 (1.01, 1.09)	0.0138
	Cord Blood	100% (169/169)	100% (75/75)	1.00 (1.00, 1.00)	n/a

*Response rate defined as number of samples above the lower limit of detection divided by number of samples tested.

**From robust Poisson regression analysis adjusted for days since last vaccination prior to delivery and prior self-reported SARS-CoV-2 infection or N protein positive at delivery.

625 **Table 4. Antibody Transfer Ratios and Cord Blood and Maternal Sera Levels at Delivery for**
626 **Primary 2-Dose and Booster Groups**

Lab Assessment	Group	Transfer Ratio Median (IQR)	Cord Blood Serum GMT (95% CI)	Maternal Serum GMT (95% CI)
Spike IgG BAU/mL	Primary	1.77 (1.38, 2.25)	500.5 (424.1, 590.7)	313.8 (260.7, 377.8)
	Booster	1.55 (1.17, 1.98)	3290.4 (2713.6, 3989.9)	2200.6 (1764.9, 2743.8)
RBD IgG BAU/mL	Primary	1.76 (1.33, 2.15)	673.1 (564.5, 802.6)	428.6 (351.9, 522.1)
	Booster	1.58 (1.05, 1.93)	5005.7 (4126.5, 6072.1)	3382.9 (2703.7, 4232.86)
D614G live virus nAb ID50*	Primary	1.10 (0.76, 1.89)	96.4 (80.2, 115.9)	83.4 (65.4, 106.4)
	Booster	1.78 (1.12, 2.25)	742.5 (588.5, 936.8)	446.4 (342.1, 582.5)
Delta live virus nAb ID50*	Primary	1.10 (1.00, 1.69)	52.1 (42.3, 64.2)	39.0 (30.9, 49.2)
	Booster	1.74 (1.06, 2.38)	644.5 (507.5, 818.4)	390.2 (299.2, 508.9)
	Primary	1.00 (1.00, 1.00)	13.8	13.5

Omicron live virus nAb ID50*			(12.2, 15.6)	(11.9, 15.3)
	Booster	1.69 (1.00, 2.75)	109.2 (81.6, 146.1)	60.2 (43.6, 83.1)
Pseudotyped WT nAb IC50*	Primary	2.36 (1.96, 2.84)	220.8 (183.5, 265.7)	93.4 (77.3, 112.9)
	Booster	1.79 (1.46, 2.20)	941.4 (735.7, 1204.7)	512.5 (399.1, 658.3)

*All data are included in the calculation of the transfer ratio including those below the LLOQ of the assay.

629 **Figure Legends**

630

631 **Figure 1. SARS-CoV-2 binding IgG and pseudovirus nAb activity in maternal and cord blood sera**
632 **by study group and study visit.**

633 Pregnant participants received a 2-dose series of an mRNA vaccine (top – Pfizer, middle – Moderna) or a
634 booster mRNA vaccine (bottom panel). Sera derived from maternal blood collected pre- and post-
635 vaccination and at delivery, and cord blood, were evaluated for binding IgG to full-length Spike (left
636 panels) and RBD (middle panels), or pseudovirus nAb titers (IC50) (right panels). Binding IgG titers were
637 bridged to international standards and reported as Binding Antibody Units (BAU/mL). Box plots
638 represent median (horizontal line within the box) and interquartile range; GMT is displayed at the top of
639 each panel and the dashed line is the cutoff for positivity (17 BAU/mL).

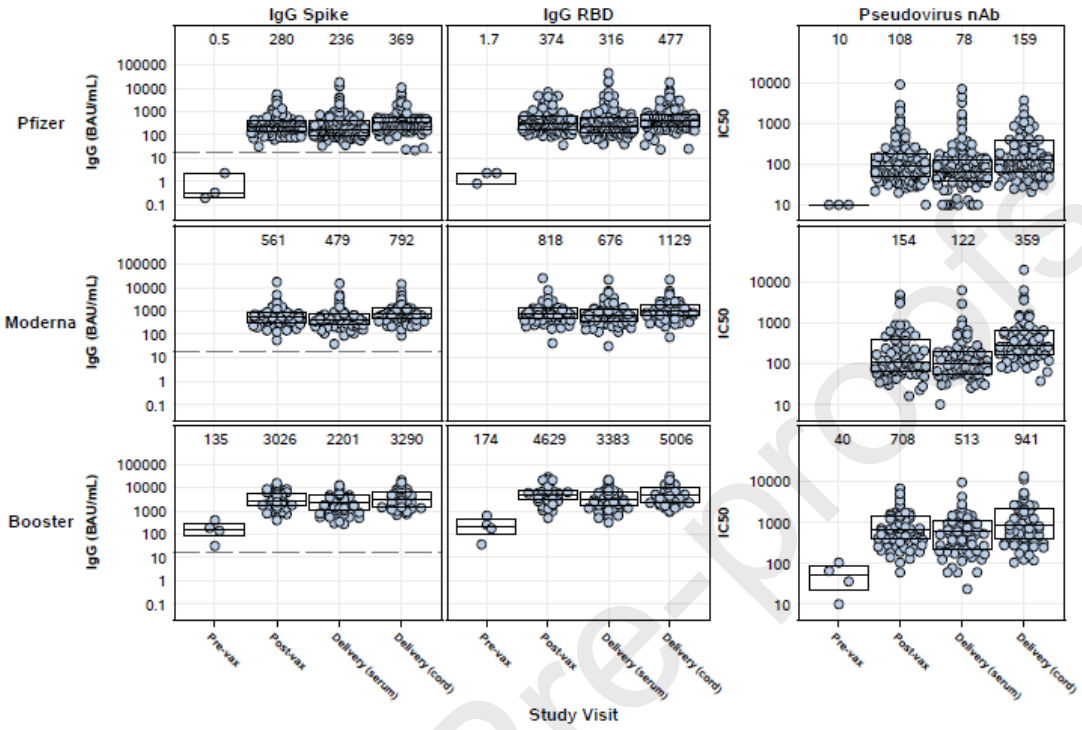
640

641 **Figure 2. SARS-CoV-2 live virus nAb activity of maternal and cord blood sera by study group and**
642 **study visit.**

643 Pregnant participants received a 2-dose series of an mRNA vaccine (top – Pfizer, middle – Moderna) or a
644 booster mRNA vaccine (bottom panel). Sera derived from maternal blood collected pre- and post-
645 vaccination and at delivery, and cord blood, were evaluated for neutralization of D614G, Delta, and
646 Omicron (BA.1) variants. Each point represents the GMT ID50 from two duplicates per specimen
647 (within the same assay run). A value equivalent to half the lower limit of detection (LLOD = 20) was
648 assigned to observations with no detectable response. A specimen was considered as having a positive
649 response if at least one of the duplicates was above the LLOD. Box plots represent median (horizontal
650 line within the box) and interquartile range. Response rate (% with responses > 20 ID50), GMT, and
651 GMT fold reduction compared to D614G are displayed at the top of each panel.

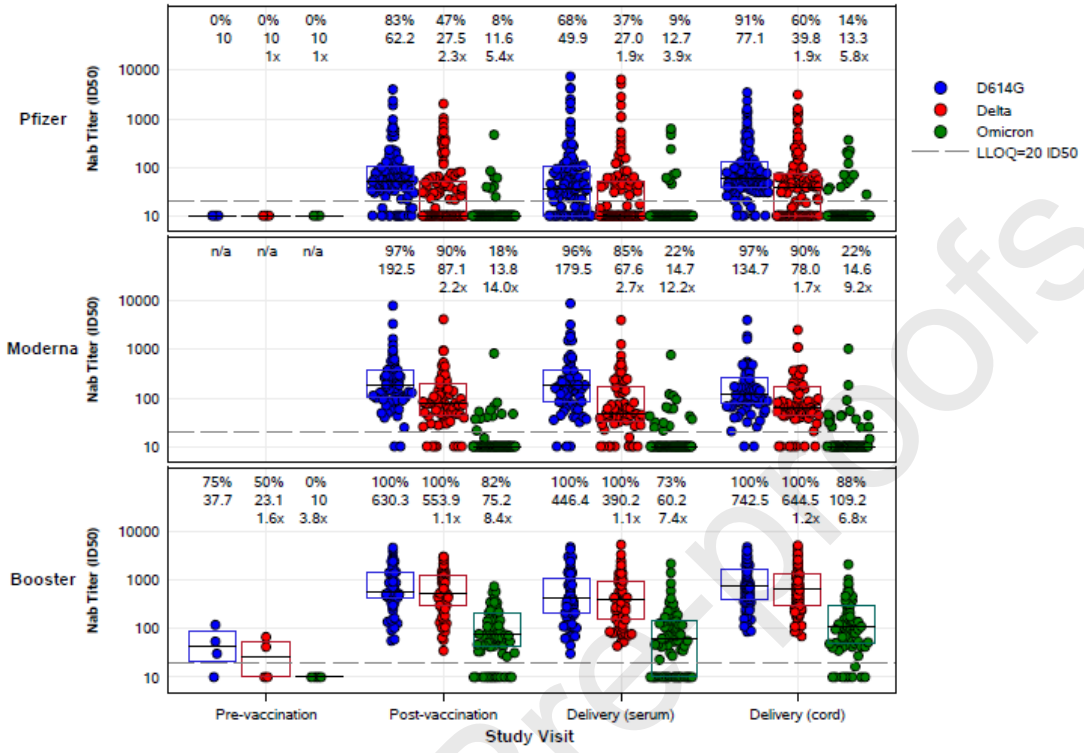
652

Figure 1. SARS-CoV-2 Maternal Serum and Cord Blood IgG (BAU/mL) and Pseudovirus Neutralizing Antibodies (IC50) by Study Group and Study Visit



SOURCE: SCHARP cwkelly /trials/idcrcp002/analysis/manuscripts/brief_report_2022_05/code/f_1_br_v6.sas OUTPUT: f_1_br_v6_out.pdf SAS Version 9.4 (21OCT22 11:00)

Figure 2. SARS-CoV-2 Maternal Serum and Cord Blood Live Virus Neutralizing Antibodies by Study Group and Study Visit



SOURCE: SCHARP cwkelly /trials/idcrc/p002/analysis/manuscripts/brief_report_2022_05/code/f_2_br_v4.sas OUTPUT: f_2_br_v4_out.pdf SAS Version 9.4 (24MAY22 12:48)

656

657

Highlights

- Data are needed to understand responses to primary and booster vaccinations during pregnancy.
- COVID-19 mRNA vaccines during pregnancy elicited robust binding and neutralizing antibody responses in mothers and newborns.
- Booster vaccination during pregnancy elicited significantly higher antibody levels in mothers at delivery and cord blood than 2-dose primary vaccination, including against the Delta and Omicron BA.1variants.
- COVID-19 vaccines, including booster doses, should continue to be strongly recommended during pregnancy.

Conflicts of Interest

F.M.M. is an investigator of pediatric studies of COVID-19 vaccines for Pfizer and for a pediatric remdesivir study conducted by Gilead Sciences, Inc; serves as investigator on projects supported by an NIH contract for a Vaccine Treatment and Evaluation Unit (VTEU), serves as member of the Data Safety monitoring Board (DSMB) for clinical trials conducted by Pfizer, Moderna, Meissa Vaccines, Virometix, and the NIH; and is a member of the American Academy of Pediatrics Section of Infectious Diseases (SOID), the Immunization Expert Group of the American College of Obstetrics and Gynecology (ACOG), and was co-Chair of the COVAX-CEPI Maternal Immunization Working Group.

K.M.N. is a member of the World Health Organization (WHO) Strategic Advisory Group of Experts on Immunization, serves as co-investigator on an NIH contract for a Vaccine Treatment and Evaluation Unit (VTEU), serves as Co-Chair of the NIH COVID Prevention Network (CoVPN), and served as an investigator for Phase I/II Pfizer COVID-19 vaccine grant, with a grant to the institution, but no salary support.

M.J.M. conducts laboratory research and clinical trials with contract funding for vaccines or MABs vs SARS-CoV-2 with Lilly, Pfizer, and Sanofi and receives personal fees for Scientific Advisory Board service from Merck, Meissa Vaccines, Inc. and Pfizer.

M.S.S. served as an advisor for Moderna (ended December 2021) and is currently serving as an advisor for Ocugen, Inc.

B.A.R. currently holds a position on a DSMB for clinical trials at Gilead Sciences, Inc.

R.C.B. at Cincinnati Children's Hospital receives research grant support for clinical trials from PATH, Astra Zeneca and Pfizer on which she serves as co-investigator.

B.B. owns shares in HDT Bio Corp.

J.S.G. receives research funds from NIH for Moderna KidCOVE study.

R.M.N. is a paid advisor to Gilead and an investigator on NIH-funded trials of Moderna, Pfizer and Janssen vaccines.

J.R-K is a medical speaker for Abbott Nutrition with the UIC team.

A.R.F. holds research grants from Pfizer, Janssen, Merck, Cyanvac, Biofire Diagnostics and serves on the DSMB for Novavax.

N.R. receives funds to conduct industry trials from Pfizer, Merck, and Sanofi-Pasteur and serves as a safety consultant for EMMES and ICON.

R.W.F. Jr., MD has received funds to conduct industry trials from Pfizer, Moderna and Astra Zeneca, serves on advisory boards for Merck, Sanofi-Pasteur, Johnson and Johnson and Seqirus and serves on an ICON-sponsored DSMB for a C difficile study.

All authors have completed relevant conflicts of interest in the Disclosure of Potential Conflicts of Interest section of the Authorship Form.