CANADIAN COVID-19 VACCINE REGISTRY FOR PREGNANT & LACTATING INDIVIDUALS (COVERED): AN EVALUATION OF SAFETY, EFFECTIVENESS & ACCEPTABILITY

Immune Sub-Study PROTOCOL

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1.0 BACKGROUND

1.1 Epidemiology of SARS-CoV-2

The World Health Organization (WHO) declared the respiratory syndrome, SARS-CoV-2 as an official global pandemic on March 11, 2021. As of Dec 13, 2021, Canada has had 1,827,691 confirmed cases and 29,900 deaths resulting from international travel, close contact exposure, and significant community spread (1). Pregnant individuals are at an increased risk for severe illness resulting in hospitalization and ICU admission compared to non-pregnant individuals (2-4). Based on this susceptibility and consequences of this respiratory syndrome in pregnant individuals, it is imperative to evaluate the COVID-19 vaccination safety, effectiveness, and acceptability in this population.

Building upon the national surveillance program CANCOVID-Preg, the reproductive infectious disease team is suited to conduct vaccine evaluation in pregnant individuals. Based on the data from the CANCOVID-Preg study, the rate of hospitalization and ICU admissions for pregnant individuals are equivalent to those in age grouping of 55-59 in the general population. The National Advisory Committee on Immunizations (NACI), Society of Obstetricians and Gynecologists of Canada (SOGC), American College of Obstetrics & Gynecology (ACOG), and other national and international organizations have declared approval of vaccination availability for pregnant individuals (5-7). The organizations have provided insight on the vaccination and encourage pregnant individuals to make an informed decision on COVID-19 immunization (7-9).

1.2 Study Rationale

Currently, there have been no formal clinical trials regarding COVID-19 vaccinations in pregnancy. As such, is currently inadequate data on the immunogenicity of COVID-19 vaccines in pregnancy and the transmission of antibodies to the fetus trans-placentally or through breastmilk. Data pertaining to the use of COVID-19 vaccines in pregnant individuals is limited to an initial v-safe report (8) resulting in significant knowledge gaps that have affected vaccination rollout federally and globally. These knowledge gaps may be contributing to decreased vaccination confidence in pregnant individuals across the globe (9). The purpose of this study is to therefore better understand the immune response generated to immunization during pregnancy.

2.0 OBJECTIVES

1. To examine the immunogenicity of COVID-19 vaccines within a subset of pregnant individuals and their infants.
2. To measure IgG and IgA antibody levels in breastmilk samples provided by pregnant individuals.

3.0 STUDY DESIGN

3.1 Study Design

This sub-protocol builds on the Canadian COVID-19 Vaccine Registry for Pregnant and Lactating Individuals (COVERED), a prospective cohort study to assess safety, effectiveness, and acceptability of the COVID-19 vaccination in pregnant individuals. If inclusion is met through the completion of the survey or upon other

recruitment strategies, individuals will be asked if they would like to be included in the immune sub study. Initial engagement has been established at BC Women's Hospital with the potential for additional sites throughout BC.

Permission will be sought to pull and test early prenatal samples for SARS-COV-2 antibodies. Prenatal and delivery samples are identified on arrival to BCCDC and are stored. The BCCDC will create a cross walk file with the study team, using first and last name, full date of birth, and PHN. Consent will be obtained to access previously collected antenatal serology and collect parent and infant blood prospectively for serological assessment.

3.2 Inclusion Criteria

1. Currently pregnant and scheduled to or have been vaccinated
2. 19 years of age or older
3. Able to communicate in English
4. Registered to deliver at BC Women's Hospital

4.0 PROTOCOL

4.1 Recruitment

The baseline survey in the COVERED Vaccine Registry study offers participants who meet the outlined eligibility criteria to be contacted regarding the Immune Sub-study. Additionally, an Immune Sub-study information page built with REDCap will be accessible online or through QR code. Other recruitment strategies include social media and poster campaigns, and recruitment through healthcare provider referrals and pre-established health clinics, such as the Penicillin Allergy Clinic, the Urgent Care Clinic, the Oak Tree Clinic, and local maternity and midwifery clinics. Research staff will reach out to potential participants who consented to be contacted, explain the details of the study and seek fully informed consent. The consent will also request access to previously collected antenatal serology and the prospective collecting of parent and infant blood for serological assessment.

Typically, each participant must participate in the informed consent process and sign and date the informed consent form before any procedures specific to this protocol are performed on that participant.

A modified recruitment method may be utilized if clinical care flow for a patient would be excessively interrupted by research. To ensure clinical procedures occur in a timely manner, a study team member may not always have the opportunity to speak with a potential study participant prior to collection of blood samples at delivery. In these circumstances, for example if a patient comes in to deliver, the nurse or staff may choose to discuss out study with them. The nurse or staff will follow a script to request verbal permission to collect blood samples for the purposes of this study and fill out the provided verbal consent checklist. Ability to verbally consent, in these instances, will better respect both nurse and patient time and reduce participant burden by allowing study sample collection to align with clinical sample collection. If a patient gives their verbal consent, the nurse will then collect their blood sample. Additionally, if there is a research staff member on site when the participant comes to deliver, they may obtain the verbal consent themselves and document this in the Approach Log.

Following the same protocol as with patients who have already formally consented, the nurse will place the sample in the labeled bin provided. They will include the checklist with the study kit. Then, they will call the study's research coordinators to come collect the sample. The research coordinators will bring the study kits with the samples and checklists to their lab to be processed immediately. Samples will be assigned and labeled with a pre-determined study ID and participant identifiers will be removed from samples prior to storage. Verbally consented samples will be stored in a temporary box in the fridge and will be transferred to a separate box once formal consent has been obtained. The research assistants will track the verbal consent process in their tracking logs.
Following sample collection, a study team member will immediately, or as soon as feasible in the setting of labour and delivery, follow up and meet with the study participant to go over the full consent form, and answer any questions or concerns prior to their signing the informed consent document. The study tracking log will have a column denoting verbal consent (y/n), the date of verbal consent (d/m/y), in addition to the already existing column for when formal consent was given (y/n), as well as the date of formal consent (d/m/y) or if they chose to withdraw their samples from the study (y/n).
If the participant who has given verbal consent chooses not to go through with the formal consent process, their samples will be safely disposed of and all identifying information on the participant will be removed from the tracking log, except for the pre-assigned study ID and the date of the destruction of the samples.
Consenting a posteriori will confer benefits to the participating women, as their clinical care flow will not be interrupted by research. The procedures followed while obtaining informed consent will be appropriately documented, as a note to file if necessary. At each subsequent study visit, participants will be asked if they wish to continue in the study and give their ongoing consent to participate. This will be documented in the source documentation.

4.2 Data Collection

An initial set of 100 mother-infant/gestational parent-infant pairs within BC will be consented for biospecimen collection. Gestational parents do not have to agree to every sample. They can therefore consent to the collection of certain samples, and not others for both themselves and their infants. As future funding permits, additional mother-infant/gestational parent-infant pairs can be consented, depending on nature of future waves and duration of the pandemic.

The following outline describes the possible study visits. Some of the visits might overlap with each other; should this occur, only one sample will be collected. If you have already received vaccine doses prior to signing this consent, the schedule of visits may occur in a different order and fewer visits will be required for study participation. Each study visit requires about 10-15 minutes of your time, and they will all occur at BC Women's Hospital.

List of all possible study visits:
Vaccine Dose

1. Four weeks up to vaccine
2. Four weeks after vaccine
3. 12 weeks after vaccine

Delivery

4. Between two days before and up to two days after delivery
5. Four to six weeks after delivery

Table 1. Specimens Collected at Different Visit Types

	Vaccine Dose Visits (collected at BC Women's Hospital Research Lab)	Delivery +/- 2 Days (collected at BC Women's Hospital during your delivery)	4-6 weeks after Delivery (collected at BC Women's Hospital Research Lab)
Gestational Parent Blood (8 mL/1.4 tsp)	X	X	X
Breast milk (5-10mL/1-2 tsp)			X
Cord blood (12 mL/2.4 tsp)		X	
Infant blood (2-4 mL/0.4-0.8 tsp)			X

Participants will be assigned a unique identification number that will be used to link Vaccine Registry survey data to their samples. Specimen collection will take place at BC Women's Hospital in the Clinical Research Evaluation Unit or at BC Women's Research Lab. Maternal serum and cord blood will be collected by trained research and laboratory staff and placed into 2 x 4mL goldtop tube. Depending on the nature of the delivery and labour, delivery nurses may collect samples for the delivery timepoint. Infant serum samples will be collected into a 0.5mL gold top sst microtainer through capillary sampling completed by trained research and laboratory staff. After centrifugation, all samples will be sent to British Columbia Centre for Disease Control for testing. Breast milk will be self-expressed into a sterile specimen container at 4-6 weeks following delivery. Each sample will be transferred into a cryovial to be frozen and stored with the purpose of testing for antibody levels.

SARS-CoV-2 serology will be performed for all maternal, cord blood, and infant samples. Infant serum and cord blood will be tested for antibodies and compared to maternal serum antibody levels. The Ortho assay will be used to test for SARS-CoV-2 anti spike protein (RBD) recombinant human monoclonal antibodies. The Roche immunoassay will be used to test for the anti-nucleocapsid antigen which detects antibodies against SARS-CoV-2. As different variants of concern and variants under investigation emerge, a thorough understanding of the strain-specific humoral responses induced by vaccination are necessary to assess for possible immune correlates of protection. Relevant viral strains will be selected for neutralization studies; a combination of live virus microneutralization testing and commercial surrogate tests (e.g., GeneScript, MesoScale Diagnostics) will be used as applicable. Additionally, serology for IgG and IgA antibodies will be performed on breast milk samples.

4.3 Data Management / Stewardship

Completed DCF's will be sent to the coordinating centre for data entry or directly entered by laboratory staff via REDCap database access accounts, which allow for siloed entry. Collected data will be entered into a REDCap database designed to mirror the DCF. Branching will be programmed into the database to allow for more efficient data entry. Each case will be assigned a unique identification number (ID#). No direct personal identifiers will be included in the national database. Support for data management will be provided by the Women's Health Research Institute (WHRI) at BC Women's Hospital.

4.4 Statistical Analysis and Metrics

The immunogenicity sub-study would require 89 participants, assuming 10% loss to follow up to estimate a 70% seroconversion rate with±10% precision and 95% confidence. A seroconversion rate closer to 80% would require 68 participants. For comparisons of antibody levels, using estimates for the 18-55y age group (13) we would need approximately 48 participants to demonstrate a 10% difference in geometric mean titer with 95% power and assuming an attrition rate of 10% per visit.

Seroconversion will be estimated and compared to licensure data using binomial tests. Antibody levels in pregnancy will be compared to licensure data for the relevant vaccine and dose using one-sample t-tests. We will stratify by gestational age, participant age, and other co-factors that might influence immune response. Maternal antibody levels will be compared to infant levels at birth, controlling for infant gestational age at delivery, birth weight, and infant feeding. Exploratory analysis of antibodies in breast milk will be compared to maternal serum antibody levels.

4.5 Knowledge Translation

Analysis and reporting from this project will be ongoing, as data become available. We will produce regular monthly national reports for distribution to our national and provincial public health partners and through the additional networks of the CANCOVID-Preg members. These reports will also be publicly shared, including with study participants, via our website, social media channels, and traditional media outlets. A knowledge broker will be engaged as part of the team to optimize translation of our findings to public health officials and a diverse range of the Canadian population, and to enhance translation of these findings into clinical guidance and public knowledge. In addition, we will leverage our partnerships with pan-Canadian university communications departments to expand the reach of our recruitment and knowledge translation outputs. Finally, our team includes clinician and policy knowledge users who will be active in the interpretation and dissemination of results, including amongst professional organizations that influence vaccine recommendations (e.g., SOGC).

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