Optimizing the implementation of a participant-collected, mail-based SARS-CoV-2 serological survey in university-affiliated populations: lessons learned and practical guidance

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Abstract
The rapid spread of SARS-CoV-2 is largely driven by pre-symptomatic or mildly symptomatic individuals transmitting the virus. Serological tests to identify antibodies against SARS-CoV-2 are important tools to characterize subclinical infection exposure.

During the summer of 2020, a mail-based serological survey with self-collected dried blood spot (DBS) samples was implemented among university affiliates and their household members in Massachusetts, USA. Described are challenges faced and novel procedures used during the implementation of this study to assess the prevalence of SARS-CoV-2 antibodies amid the pandemic.

Important challenges included user-friendly remote and contact-minimized participant recruitment, limited availability of some commodities and laboratory capacity, a potentially biased sample population, and policy changes impacting the distribution of clinical results to study participants. Methods and lessons learned to surmount these challenges are presented to inform design and implementation of similar sero-studies.

This study design highlights the feasibility and acceptability of self-collected bio-samples and has broad applicability for other serological surveys for a range of pathogens. Key lessons relate to DBS sampling, supply requirements, the logistics of packing and shipping packages, data linkages to enrolled household members, and the utility of having an on-call nurse available for participant concerns during sample collection. Future research might consider additional recruitment techniques such as conducting studies during academic semesters when recruiting in a university setting, partnerships with supply and shipping specialists, and using a stratified sampling approach to minimize potential biases in recruitment.

Keywords: Serosurveys, Serology, SARS-CoV-2, Epidemiologic methods; field epidemiology

Background
Serological surveys are an important tool to estimate population-level prevalence of infectious diseases worldwide and are critical for estimating the distribution of SARS-CoV-2 infections [1–3]. Reported in 2021, nearly one-third of people infected with SARS-CoV-2...
are asymptomatic [4]; serological surveys can expose underlying transmission dynamics that would otherwise remain hidden due to the prioritization of symptomatic testing in most clinical settings. Therefore, population-based seroprevalence studies directly inform public health decisions by providing data on the distribution of infections towards understanding consequent immunity. To monitor the seroprevalence of populations in the United States, the COVID-19 Seroprevalence Studies hub was created (https://covid19serohub.nih.gov/). This hub serves as a repository for studies that determine the seroprevalence of populations including the general public, healthcare-based populations, military populations, and university-affiliated populations [5].

Understanding viral exposure through serological testing is especially useful in university populations where a large percentage of young people may have asymptomatic or subclinical infections. University populations also experience cyclical population mobility due to the academic schedule. Viral transmission among university students may also affect those in close proximity to the university. Though not the case in all large universities and surrounding communities [6], it was found that counties with large university populations offering in-person instruction experienced a 56.2% increase in COVID-19 incidence when comparing the 21 days before and after the first day of classes in fall 2020 [7]. These data suggest that viral transmission networks extend beyond universities and into surrounding communities, and may impact vulnerable community members [8]. This also highlights a need for evidence of transmission for policymakers regarding controlling potential outbreaks [7, 9, 10].

Seroprevalence studies benefit from standardized methods, which might include a comparison of the general population with subpopulations, analysis of potential factors associated with infection, and investigation of variations in immune response. A standardized approach is essential to facilitate the comparison of results across studies [3, 11]. Serosurveys of the general population are strengthened with the inclusion of higher-risk subpopulations to help prioritize interventions [3]. At-home serosurvey sampling using self-collected dried blood spot (DBS) cards from a mailed testing kit has been validated and found to be a reliable alternative to serum samples for detecting SARS-CoV-2 antibodies [12, 13]. Additionally, there is evidence that collecting venous blood either in-person or at home might be less acceptable to participants than an at-home dried blood spot (DBS) collection [3, 10, 11, 14, 15] and while micro-vacutainers exist for at-home finger-prick sampling, the cost (approx. 10x the cost of DBS cards), and complexities in shipment and long-term storage make DBS a robust, sustainable, and more-convenient global standard [16]. While simpler to use a convenience sample for serological measures, this might reduce validity and generalizability of results. Therefore, many studies employ a population-based random sample which may also include household members to increase the sample size [2, 3, 11]. In addition to testing individuals and their household members for serological results, additional insight regarding demographics and potential risk factors for infection is gained through the combination of serological tests and survey items [3, 11, 14].

During the summer of 2020, there were important considerations to inform the safety of campus populations returning to campus at the University of Massachusetts Amherst due to the COVID-19 pandemic. Research has demonstrated that individuals with asymptomatic or subclinical infections may still transmit SARS-CoV-2, potentially contributing to the rapid growth of outbreaks [4, 17]. To better characterize SARS-CoV-2 transmission in Massachusetts, a mail-based serosurvey was conducted to quantify the prior viral exposures among UMass students, faculty, staff, and their household members.

The primary aim of this case study is to describe in detail the logistics and implementation of a mail-based serosurvey for the identification of SARS-CoV-2 antibodies in asymptomatic individuals without prior COVID-19 diagnosis while using participant-collected dried blood spots (DBS). We describe participant recruitment methods, design of the data storage platform, logistics of navigating the shipping supply chain, and statistical analysis. The description of the challenges encountered throughout this study can be used to inform researchers implementing similar mail-based serological studies.

**Main text**

**Study population and recruitment**

The study population included University of Massachusetts Amherst (UMass) affiliates (undergraduate students, graduate students, faculty, staff, and librarians) plus a single member of these affiliates’ households. UMass affiliates were eligible to participate in the study if they were 18 years of age or older, resided in Massachusetts, reported never having a COVID-19 diagnosis from a healthcare professional, remained in Massachusetts for the 8 weeks prior to survey administration, and did not report a fever greater than 100.4°F at the time of survey completion. Each UMass affiliate was also asked to invite a single member of their household to participate in the study. Eligibility criteria for this household member sample were identical, except household members had to be between 23 and 78 years old to ensure age variation in the population. The restriction to Massachusetts residents
who had not left the state was intended to assess the amount of exposure present in The Commonwealth.

UMass affiliates were contacted via email to participate in this study between June 23rd, 2020, and June 26th, 2020. The invitation email described the study goals, methods, and a unique link to a screening and study eligibility survey. The eligibility survey included an embedded video demonstrating the sample collection process to familiarize participants with blood spot collection. If participants were eligible and agreed to participate, they were asked to electronically sign an informed consent document. Upon consenting, the link opened a detailed survey instrument capturing detailed demographics and potential COVID-19 risk factors. The associated household members were also invited to complete these forms. The eligibility, consent, and survey forms were created specifically for this study. All survey questions are made available in Additional File 1.

Survey development and distribution

All survey instruments were created and administered using Research Electronic Data Capture (REDCap) software version 9.9.0, provided through the University of Massachusetts Medical School. REDCap was chosen for its ability to send unique and HIPAA-compliant survey links to a large number of individuals from a pre-defined address list. Storing records within this system provided a secure linkage between participant demographic information and laboratory results. Email addresses for UMass affiliates were obtained from the University’s Office of Academic Planning and Assessment and were uploaded to REDCap. Emails containing unique survey links for each participant were created in REDCap and sent through the system interface to participants. Emails were sent in batches of several hundred to avoid potential server capacity issues. Surveys were pre-tested with diverse user groups and took approximately 5 minutes to complete. Reminder emails were sent to non-respondents on day 3 and day 6 after the initial email.

After a three-week response period, the survey was locked, and a randomized subset of participants was selected to receive a bio-sample collection kit. Different sampling schemes were used for employees (graduate students, faculty, staff, and librarians) and undergraduate students. The employee sample was generated through a probability proportional to the population size using the 2019 census data for each of the Massachusetts Emergency Regions [18]. Emergency regions were used instead of counties because sampling at a county-level was not feasible. Specifically, there was insufficient representation to provide stable estimates for all 14 Massachusetts counties, even though many students had returned to their hometowns at the time of survey administration. Hereafter, UMass undergraduate students and their associated family members are referred to as the “primary” sampling group; UMass employees (graduate students, faculty, staff, and librarians) and their family members are the “secondary” sampling group.

Supply chains and shipping logistics

Each household was sent a single box with supplies for the UMass affiliated participant and their enrolled household member. While dried blood spots are generally exempt from special mailing considerations, to comply with biosafety considerations during the pandemic boxes appropriate for the transportation of Category B Biological Substances (UN3373) [19] were used. These were labeled “Biological Substance, Category B, UN3373”, “Exempt Human Specimen.” Bubble wrap was included in the shipping boxes to protect the contents. Bio-box contents contained a bloodspot card with a pre-affixed unique barcode identifier for each participant (stored in bags with silica gel), a sample collection supply bag (gloves, alcohol pads, lancets, bandages, gauze), a biological hazard bag for returning all collection materials, and a detailed pamphlet with sample collection instructions (Fig. 1). This pamphlet contained detailed “how-to” information to properly collect the dried bloodspot sample plus instructions for properly sealing the biological hazard return bag (Supplemental Fig. 1). Boxes containing all information were assembled in a laboratory at UMass-Amherst by a team of three researchers prior to shipment (Fig. 2A). After the sample was collected and air-dried, participants were instructed to place

![Fig. 1](https://via.placeholder.com/150)

Fig. 1 Contents of shipped bio-boxes. Example of box mailed to study households, which contained: dried blood spot collection cards with barcodes, two silica packets per DBS card, collection supplies (gloves, lancets, gauze, alcohol pads, and adhesive bandages), biospecimen bags, return labels, tape to seal the box for return, and instruction sheets for how to collect the sample and how to close the biospecimen bag. Image was captured by the authors of this paper and all company logos have been hidden for copyright purposes.
the bloodspot card into the bag with a silica desiccant packet. This bag was then placed into the biological hazard return bag, and all box contents were placed into the shipping box for return.

Two different shipping modes were used. Boxes were shipped to participants using United Parcel Service (UPS) as it was the most economical and simplest for bulk mailings and allowed for tracking to ensure arrival at each participant’s home. United States Postal Service (USPS) was used to deliver to PO boxes, as UPS was unable to deliver to these locations. Generic return labels and shipping tape were included in all kits so the same boxes could be returned via USPS. USPS was used for all returns to make the shipment as simple as possible for participants, as boxes could be returned at a post office, into blue mailboxes, or via pickup as part of routine home mail delivery. UPS was a less desirable option for return shipments, as participants would have needed to locate a UPS location for drop-off, many of which were closed due to the pandemic itself. The vast majority of boxes were returned without any issues, however some boxes were damaged during delivery or during the return process. An example of an issue was water damage (Fig. 2B).

Sample processing
Returned test kits were processed in a biosafety level-2 laboratory. External box surfaces were sanitized with 70% ethanol before being opened in a HEPA filtered class II biosafety cabinet. All excess supplies were either autoclaved or disposed of in sharps containers as per standard laboratory practice and institutional biosafety guidance. Bloodspot cards were removed from return bags and matched to individuals in REDCap using the name provided on the outgoing shipping label. Barcodes attached to the DBS cards were verified with the participant-entered barcodes in REDCap and entered into the second online survey form (Additional File 1). Outgoing shipping labels were consulted to confirm and triangulate the barcodes and to link the few participants who did not complete the final online survey signifying that they had received the boxes. Most of the DBS cards had usable bloodspots, however, some cards, most often those performed with the small lancets (see Conclusions), had spots with insufficient sample for analysis (Fig. 2C).
DBS cards were heat-treated at 56 °C for 30 minutes before bloodspots were manually punched using a 0.25-in. diameter stationery hole puncher. A single bloodspot per participant was extracted and tested using an in-house ELISA assay (IgM and IgG for SARS-CoV-2 receptor-binding domain) as described in published protocols [20]. Each 96-well ELISA plate contained one positive control and seven negative controls. Optical densities were read at 405 nm and normalized daily to the mean optical density of all negative controls.

Statistical analysis
Seropositivity cutoff values were determined using finite mixture models [21, 22]. These models identified the optimal breakpoint between seronegative and seropositive subpopulations. For this survey, samples with an optical density $\geq 2.49$ above daily control values were considered positive for RBD antibodies. Logistic regression models were used to determine propensity scores for each individual to calculate non-response weights that were applied via inverse weighting. Undergraduate students were self-weighted due to the probability proportional to population size sampling scheme; employees did not require weighting as simple random sampling was used. Poisson models with robust errors to account for household clustering were utilized for separate multivariable regressions within each sampling scheme. Final models were identified using Akaike & Bayesian information criterion (AIC/BIC) and were adjusted for age, race, and self-reported gender. Analyses were performed with R version 4.0.3 [23] and SAS version 9.4 (SAS Institute, Cary, NC, USA).

Study power and sample size
This study was designed to provide sufficiently precise estimates of seropositivity to inform policy. With 750 persons, and an assumed 5% positivity, the 95% CI for this estimate is 3.6 to 6.9%. Within the five emergency response subregions, at 5% seropositivity, the survey is powered for a precision of 2.3 to 10.2%. The secondary sampling group ($n = 250$) sample size was based on logistic limitations but was powered to a precision of 2.8 to 8.8%. All confidence intervals are binomial exact, without adjustments for study design effects or non-response [24].

Survey response rates
Initial emails were sent to 27,339 UMass affiliates; of the 4531 individuals who completed the required documents; 407 persons were ineligible. Of the 4124 eligible individuals, 1001 were randomized to receive a test kit (752 undergraduate students and 249 employees). Among the undergraduate students, 330 enrolled a household member; 548 students and 231 household members returned samples and were included in the analysis. Among employees, 101 enrolled a household member; 214 employees and 78 household members returned samples and were included in the analysis. Overall, 76% of randomized participants returned blood samples for analysis (Fig. 3).

Forty-six samples out of 1071 total samples were positive for IgG SARS-CoV-2 antibodies; full seroprevalence results have been published previously [24].

Conclusions
Inclusion / exclusion criteria
By restricting participation to individuals currently residing in Massachusetts and not traveling out of state, we were able to categorize seroprevalence among Massachusetts residents and the transmission of SARS-CoV-2 within the state at the earliest stages of the pandemic. However, this prevented the analysis of the impact of mobility on the prevalence rates of asymptomatic COVID-19 exposure. By studying only those who did not exhibit fevers, we were able to capture the asymptomatic exposure levels, however the true amount of SARS-CoV-2 exposure in the state at the time was likely an underestimate. Future studies may consider fewer exclusion criteria to allow for more generalizability. We would recommend including individuals who have traveled in future studies so that the impact of mobility can be analyzed.

Acquisition of Supplies and Staffing Considerations
A test batch of approximately 100 boxes was sent out, to pilot test the data collection. Feedback indicated that many participants found the lancet needle was too small to obtain strong blood flow, making it difficult or impossible to collect sufficient sample for full bloodspot samples. Additionally, these safety lancets self-retracted after a single use, and some participants used all that were sent as they were unfamiliar with the design. This situation allowed packing all subsequent boxes with larger bladed lancets plus shipping new lancets to some from the pilot participants. Ensuring that the types of lancets chosen have large enough blades to obtain the desired sample is essential (e.g., 1.5 mm bladed, with 1.6 mm penetration depth). The cost of lancets is minimal relative to overall survey implementation and so multiple extras should be included for all mailings.

Coordination with mail services
Close coordination with the university campus mail services was essential in managing the challenges inherent in shipping approximately individual 1000 boxes. Outgoing boxes were delivered to the university loading dock...
over several days; special pickups were coordinated with UPS due to the size of the shipments. Mail Services added extra staff and additional deliveries to accommodate the large volume of returned boxes. Due to shipping delays and backlogs with both UPS and USPS, several boxes were delivered to participants much later than anticipated. Participants were granted extended deadlines and could return boxes to the laboratory in person to maximize sample collection. Future studies that use at-home test kits should prioritize collaboration with shipping specialists and should anticipate possible delays at all stages of the shipping process.

Response rates
When conducting this survey, there were concerns that many university students might not access their institutional emails on a regular basis over the summer months. Overall, the non-response rate for the study was 83.4% among UMass affiliates. Although the survey completion rate appears low, it is slightly higher than other studies related to COVID-19 conducted on large university college campuses during 2020 [25–27]. Therefore, it appears that UMass affiliates continued to monitor their academic emails over the summer vacation period.

In this survey, attempts to improve participation rates were made by minimizing the length of the surveys (~5 minutes), keeping the enrollment period open for 3 weeks, and sending reminder emails to non-respondents 3 days and 6 days after the initial email. Additionally, attempts to increase participation included informing participants about the length of the survey and giving them the option to receive their serological results upon completion of the study. To achieve higher response rates, future studies may consider sending out the survey from a more familiar office on campus, such as the office of the dean [28]. This could help ensure that emails receive proper attention and aren’t sent to “Junk” folders.

One reason for concern regarding low response rates is the potential of selection bias due to the types of individuals most likely to respond to volunteer-based surveys [29]. Although it is difficult to quantify the extent to which selection bias may have impacted findings, among the primary sampling group, a larger percentage of women responded (62%) compared to the overall proportion of undergraduate women at UMass (51%). There was also slightly less representation of minority populations (African American, Chicano/Latino, and Native American/Alaska Native) in the study population than in the broader UMass undergraduate population. In parallel, among the secondary sampling group, there was a higher percentage of White and Asian individuals, but there was not a difference in the percentage of females
relative to the overall demographics [30]. Men may be more susceptible to SARS-CoV-2 infection and fatal outcomes [31, 32] so this overrepresentation of women may have underestimated the SARS-CoV-2 exposures present in the population. Similarly, during the summer of 2020, minority groups were more likely to contract SARS-CoV-2 [33], therefore, the overrepresentation of White and Asian race categories may have underestimated the overall exposures in this sub-population. In survey studies, it is common that women and individuals with lower risk factors are more likely to participate [29], so future studies should consider oversampling men and minority groups to better capture population-level prevalence.

Randomization
After the three-week enrollment period, randomization of participants to receive an at-home test kit occurred for all eligible participants to avoid choosing the first group of individuals to enroll in the study. These individuals could have been more motivated to participate due to known exposure or other factors. In this study, two sampling approaches were used. For the primary study population, a stratified sampling approach was used to capture a sample representative of individuals from across the state. For the secondary sample, a simple random sample was used. Future studies may consider a more stratified sampling approach to better capture the diversity in race and gender and thus collect a more representative sample of the broader population.

Survey design
A potential source of drop-out from household members who initiated the survey but did not complete it was confusion over a survey eligibility question regarding UMass affiliation. In the eligibility screen, the question was, “Is someone in your household a current UMass student or employee?” Some people were confused by this wording thus answered incorrectly and were deemed ineligible to complete the survey and be included in testing. Given the timing of these surveys, it was not possible to do a large-scale pilot test. Future studies should include as large a pilot as feasible.

Additionally, after sending out test kits, individuals were sent a brief form asking them to enter their barcode information into the web portal from the barcoded blood spot card (as it was not feasible to allocate the >1000 boxed code DBS to individual addresses). Unfortunately, a number of participants returned their kits without completing this form, thus making it difficult to match samples to individuals. A potential solution would be to record the barcode of each test strip mailed to each individual before mailing.

Dissemination of results
A major unanticipated obstacle occurred when the guidance surrounding the distribution of COVID-19 related laboratory results to participants was clarified. With approval from the IRB, participants were offered the option to receive their individual test results when initially signing up for the study. A few months after this determination, CDC guidance was expanded to cover reporting of all SARS-CoV-2 test results, which meant that any analyses performed in a laboratory setting that was not clinical laboratory improvement amendments (CLIA) certified were deemed “research”, and results could not be disseminated to participants. Due to these changes, only aggregate results could be shared with study participants instead of individual results.

Final summary
Despite the challenges encountered throughout implementing this study, the methods developed for sample collection were shown to be efficient and effective for obtaining contact-free seroprevalence data by mail. This study used novel strategies for its study design by selecting a subset of university affiliates, enrolling their household members, and utilizing participant-collected blood samples using at-home DBS kits. Administering online questionnaires to UMass affiliates and their family members was a rapid method to obtain a representative sample of Massachusetts residents’ potential SARS-CoV-2 antibody statuses. This mail-based, minimal contact study was a safe, cost-effective, and pragmatic way to assess prior infection with SARS-CoV-2 during the earliest waves of the pandemic.

Table 1 Lessons learned from the implementation of a mail-based SARS-CoV-2 serosurvey

- A small test batch of boxes should be sent out before sending to all participants to identify problems with the sample collection kit.
- The logistics of navigating the supply chain, package assembly, and shipping can be challenging and should be carefully planned.
- Professional shipping support for 2-way discrete packages is essential.
- Study participation may be increased by sending out survey invitations from a familiar on-campus office.
- A stratified sampling approach for participant selection may better characterize disease transmission in the broader population.
- Pilot testing the initial survey may prevent confusion with study enrollment.
- Ensuring sample-to-record linkages in remote settings requires careful consideration.
- Processes for return shipment need to be as user-friendly as possible.
- Possible changes to policies regarding test result distribution should be considered.
When conducting future studies relying on the at-home collection of samples, consideration should be given to the availability of shipping supplies, potential complexities with shipping processes, methods to increase response rates among eligible participants, and determining how regulatory changes may impact roll-out and reporting (Table 1).

This report highlights the feasibility and acceptability of self-collected biosamples and has broad applicability for other serological surveys for a wide range of other infectious diseases.

Abbreviation
DBS: Dried blood spots; UMass: University of Massachusetts Amherst.

Supplementary Information
The online version contains supplementary material available at https://doi.org/10.1186/s12889-022-14234-1.

Additional file 1. Eligibility, Consent, and Demographic survey administered to UMass affiliates and their household members. REDCap surveys were administered to participants through campus email accounts.

Supplemental Fig. 1. Instructional guides for collecting and repackaging samples. PDF versions of files were included with every box giving users instructions on how to collect blood spots and properly mail back the packages.

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Authors’ contributions
AAL led study conceptualization and design. EYC, TS, and JR implemented survey design, fieldwork and associated data collection. EYC, JR, TS, and AAL performed data cleaning and analyses and wrote the first draft. All authors contributed to revisions of the final manuscript. All authors read and approved the final manuscript.

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Availability of data and materials
The datasets used and/or analyzed during the current study available from the corresponding author on reasonable request.

Declarations
Ethics approval and consent to participate
I. This study has been reviewed and approved by the University of Massachusetts Amherst IRB, (Approval #2062; 27 Apr 2020). II. All methods were carried out in accordance with relevant guidelines and regulations, and the Declaration of Helsinki (2013). III. Informed signed consent was obtained from all study participants.

Consent for publication
Not applicable; this study reports only aggregate deidentified data.

Competing interests
The authors have no competing interests as defined by BMC, or other interests that might be perceived to influence the results and/or discussion reported in this paper.

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