INNOVATIVE TECHNOLOGIES IN PUBLIC HEALTH

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CANADA COMMUNICABLE DISEASE REPORT

INNOVATIVE TECHNOLOGIES IN PUBLIC HEALTH

CONTENTS

Coupling wastewater-based epidemiological surveillance and modelling of SARS-CoV-2/COVID-19: Practical applications at the Public Health Agency of Canada

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Abstract

Wastewater-based surveillance (WBS) of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) offers a complementary tool for clinical surveillance to detect and monitor coronavirus disease 2019 (COVID-19). Since both symptomatic and asymptomatic individuals infected with SARS-CoV-2 can shed the virus through the fecal route, WBS has the potential to measure community prevalence of COVID-19 without restrictions from healthcare-seeking behaviours and clinical testing capacity. During the Omicron wave, the limited capacity of clinical testing to identify COVID-19 cases in many jurisdictions highlighted the utility of WBS to estimate disease prevalence and inform public health strategies; however, there is a plethora of in-sewage, environmental and laboratory factors that can influence WBS outcomes. The implementation of WBS, therefore, requires a comprehensive framework to outline a pipeline that accounts for these complex and nuanced factors. This article reviews the framework of the national WBS conducted at the Public Health Agency of Canada to present WBS methods used in Canada to track and monitor SARS-CoV-2. In particular, we focus on five Canadian cities—Vancouver, Edmonton, Toronto, Montréal and Halifax—whose wastewater signals are analyzed by a mathematical model to provide case forecasts and reproduction number estimates. The goal of this work is to share our insights on approaches to implement WBS. Importantly, the national WBS system has implications beyond COVID-19, as a similar framework can be applied to monitor other infectious disease pathogens or antimicrobial resistance in the community.

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Introduction

Epidemics caused by infectious pathogens are traditionally monitored through clinical surveillance of individuals. Wastewater-based surveillance (WBS) is an alternative epidemiological surveillance approach that consists of assessing the concentration of a pathogen of interest in wastewater to estimate its associated infection prevalence in a community. Wastewater-based surveillance has been integrated as part of poliovirus eradication initiatives since 2010 (1). In Canada, it has been used to monitor drug consumption and viral pathogens for seasonal viral load changes and inactivation by wastewater treatment processes (2–6). During the coronavirus disease 2019 (COVID-19) pandemic, WBS has attracted a lot of attention for surveillance of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) (the virus that causes COVID-19) both in Canada and globally (7). Wastewater-based surveillance provides a complementary tool for clinical surveillance to detect and monitor trends of disease caused by SARS-CoV-2. In contrast to clinical surveillance of COVID-19 (8), WBS is not limited by

underdiagnosis of asymptomatic individuals because most individuals infected with SARS-CoV-2 shed viral particles in their stools (9,10). Wastewater-based surveillance utilizes a pooled community sample from the catchment area of a sampling location to measure the levels of SARS-CoV-2 within the community (11). Multiple studies have shown that SARS-CoV-2 concentration measured in wastewater correlates with the real prevalence affecting the community living in the catchment area (12–15).

Wastewater-based surveillance garnered high interest during the emergence of the variant of concern Omicron in November 2021 (16). Its large number of genetic mutations compared to the previous circulating lineages conferred the variant a higher transmissibility and immune escape that fuelled a rapid growth of cases (17). Hence, during the Omicron wave, testing capacities in many countries, including in major Canadian cities, were overwhelmed, forcing the polymerase chain reaction (PCR)-testing of SARS-CoV-2 in clinical samples to be restricted to certain high-risk or vulnerable populations (18). Previous research demonstrated that SARS-CoV-2 was detected in 29%–100% of fecal samples in infected individuals (19) and that WBS detection of SARS-CoV-2 preceded confirmed clinical cases by 5–63 days (11), confirming WBS as 1) an alternative measure of disease prevalence, especially when clinical surveillance is limited by overwhelming demand or test-seeking behaviours and 2) an early indicator of COVID-19 presence to inform testing and public health strategies at the community level (20). Overall, WBS offers a non-invasive and low-cost method to estimate the community prevalence of COVID-19 that addresses the limitations of traditional clinical surveillance.

However, WBS is not free of biases and uncertainties. Wastewater-based surveillance can be influenced by various pre- and post-analytical factors, including methods of sample collection and storage, laboratory analysis protocol, engineering of the sewer network and wastewater treatment plants (WWTP), changes in weather conditions and data analysis procedures (21–23). Moreover, since WBS for SARS-CoV-2 is still evolving, there is a lack of standardized procedures to address these factors. Considering the potential sensitivity of WBS data to these factors, it is crucial to establish a pipeline that specifies standardized protocols and methodologies from sample collection to analysis to ensure the accuracy of WBS. While it may be impossible to control for some sources of uncertainty, minimizing their effects remains crucial. Importantly, there is a need to implement a framework to combine the results of WBS and clinical surveillance to clearly communicate the epidemiological findings to inform public health strategies (24).

In Canada, WBS is performed by laboratories at federal, provincial and municipal levels as well as by academic groups (7). The National Microbiology Laboratory (NML) at the Public Health Agency of Canada (PHAC) collates and analyses samples from multiple provinces to conduct WBS at the national level.

The objective of this review article is to provide a comprehensive overview of the WBS pipeline at the NML and a framework to incorporate WBS and clinical surveillance to enhance the national surveillance of COVID-19. We describe how mathematical modelling can be utilized to facilitate the interpretation of WBS outputs. To increase the usefulness of the WBS data, we assess key factors that influence WBS signals at each step of the pipeline and methods to address them.

Results

Wastewater-based surveillance pipeline
The Canadian national WBS program involves the collaboration of municipal WWTP and multiple government divisions and agencies, including Statistics Canada, NML and PHAC. The WBS pipeline was developed to streamline the WBS processes from sample collection to reporting in an accurate and timely manner (**Figure 1**).

Figure 1: Wastewater-based surveillance data and analysis pipeline at Public Health Agency of Canada/ National Microbiology Laboratory
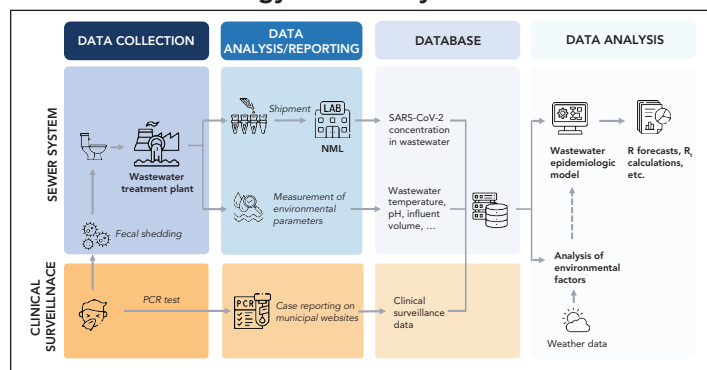
Abbreviations: NML, National Microbiology Laboratory; PCR, polymerase chain reaction; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2

Data collection
The Canadian Wastewater Survey, jointly led by Statistics Canada and PHAC, currently involves 102 WWTPs across Canada. We focus on 15 WWTPs of five cities—Vancouver, Edmonton, Toronto, Montréal and Halifax—where mathematical modelling is applied to analyze the trends of SARS-CoV-2 (**Figure 2**). The wastewater sampling in the five cities began in September 2020. The samples are collected approximately twice a week from the raw influents. Samples are collected before de-gritting in one WWTP in Edmonton, three WWTPs in Montréal and three WWTPs in Vancouver and post-grit removal in four WWTPs in Toronto and two WWTPs in Vancouver. Wastewater can be sampled using composite or "grab sample" methods. Grab sampling constitutes rapid sampling at a specific point in time, which represents the influent at that time; therefore, the results are more subject to changes in the influent flow of the day. Composite sampling involves collecting multiple samples using

an automatic sampler during a set period (typically 24 hours) to represent the wastewater composition for that period. For the Canadian Wastewater Survey, the composite sampling method was used where automatic samplers collected wastewater samples during a 24-hour period. These samples were kept at 4°C and shipped to the NML in Winnipeg, Manitoba.

Figure 2: Wastewater-based surveillance of COVID-19 is actively conducted by federal, provincial and municipal governments and by academic institutions across Canada
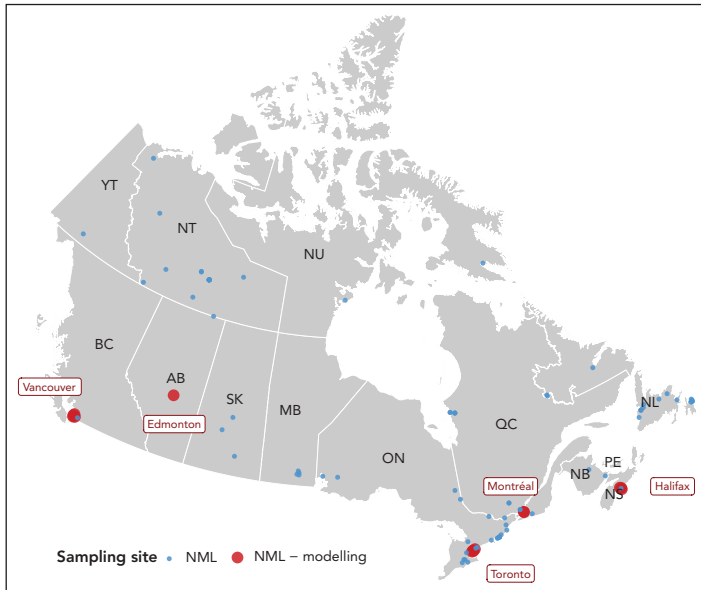
Abbreviations: AB, Alberta; BC, British Columbia; MB, Manitoba; NB, New Brunswick; NL, Newfoundland and Labrador; NML, National Microbiology Laboratory; NS, Nova Scotia; NT, Northwestern Territories; NU, Nunavut; ON, Ontario; PE, Prince Edward Island; QC, Québec; SK, Saskatchewan; WWTP, wastewater treatment plant; YT, Yukon

In addition to sample collection, wastewater quality and environmental parameters of the wastewater, such as influent daily volume, temperature and pH, are measured at the WWTP. The wastewater data from NML are collated together with the environmental parameters from each WWTP by Statistics Canada for data management and shared with PHAC/NML.

Clinical surveillance data are retrieved from the PHAC line list of COVID cases (an anonymized list of COVID cases, at the individual level, communicated by the provinces and territories to PHAC during the COVID-19 response) or publicly available sources on municipal websites for each city in cases where the PHAC line list does not have sufficient spatial resolution. When available (e.g. Toronto, Vancouver), we collect data at the sub-municipal level to map the spatial location of the clinical reported cases with the catchment area of each WWTP. Weather-related environmental data, including amount of precipitation and snow on ground for each city, are obtained from Environment Canada.

Laboratory analysis of SARS-CoV-2 concentration

The SARS-CoV-2 concentration was measured with two methods. The laboratory protocols for the two methods were described in detail by Nourbakhsh et al. (25). Briefly, before February 12, 2021, SARS-CoV-2 ribonucleic acid (RNA) was extracted from the liquid supernatant portion of clarified wastewater samples. However, early studies found that the solid portion of clarified wastewater samples yield a higher viral concentration (26–28). Therefore, after February 12, 2021, RNA extraction was performed on the solid pellet after clarification. The change in protocol improved the efficiency of RNA quantification.

Data quality and sources of uncertainty

The WBS data are influenced by several factors, including environmental conditions, laboratory protocols and engineering of the WWTPs. Below, we summarize how environmental and laboratory factors can impact WBS data. This is still an area of active research, and many knowledge gaps remain.

Environmental factors such as precipitation or snowmelt have been described as critical factors that could influence viral signals in the wastewater (29). However, the impact of environmental factors could vary depending on the type of the sewer system serviced by a WWTP. There are two major types of sewer systems—combined and sanitary. Combined systems collect storm water from surface runoff and wastewater together within the same pipes. While combined systems would only collect wastewater as influent water to the WWTP during dry weather, wet weather or high precipitation events (including snowmelt) would increase the influent flow rate and dilute viral concentration present in the wastewater (29). In contrast, sanitary systems mostly separate storm water and sewage, which means the influent volume do not change significantly based on the weather, avoiding the dilution of the viral signal.

Combined systems are present in older parts of the cities monitored by PHAC. To ensure the quality of the WBS data, we investigated the potential mediating effects of precipitation on the WBS SARS-CoV-2 signal. Our quantitative analyses of environmental factors (manuscript in preparation) revealed that while some fluctuations in influent volume were recorded with changes in precipitation, they do not appear to significantly impact the SARS-CoV-2 concentration in wastewater for the dates and sites analyzed. Snowmelt has also been suggested to influence the SARS-CoV-2 signal in wastewater (21,30). Although some studies showed the influent volume increased during snowmelt season (30–32), there is a paucity of evidence that snowmelt events have a significant impact on the viral signal.

Laboratory factors

Viral concentration measurement from a wastewater sample is a multi-step process, where each step can introduce a potential source of error. The duration and conditions of transport of the

sample from the sampling location to the laboratory may impact the final concentration measurement of SARS-CoV-2. By their nature, wastewater samples are very "active"; i.e., there is a high degree of biological activity that causes the nature of the sample to change rapidly. In addition, the equipment and containers of the sampling system may be contaminated. Therefore, storing, transporting and handling wastewater samples are critical to maintain their integrity and are potential sources of errors. Moreover, the complex and variable nature of wastewater requires the proper use of control samples to account for variations in the composition of wastewater and evaluate overall efficiency of the process. Failure to properly run these controls are other potential sources of error. Molecular detection by real-time quantitative polymerase chain reaction (RT-qPCR) testing may also be prone to errors (e.g. standard curve not updated, new viral mutations affecting the identification by primers). Hence, rigorous protocols to ensure consistency and reliability of SARS-CoV-2 concentration measurements from wastewater samples should be in place at this stage of the WBS pipeline. Guidance regarding such protocols is presented in detail in the **Supplemental material**.

Normalization

As mentioned above, many factors can affect the viral concentration in wastewater. Ideally, those factors would be identified, measured and controlled for before communicating a "final" viral concentration in wastewater. Wastewater is a complex matrix that contains biological, chemical and physical factors that may affect the RNA concentration and/or detection. Wastewater not only contains domestic sewage, but may also have industrial/agricultural discharge and storm water depending on weather conditions (33). From these influents, the composition of wastewater may change in pH, chlorine and dissolved oxygen content, which may reduce the viral concentration (23). Moreover, transportation of wastewater through the sewage network involves fluctuations in wastewater temperature, flow rate, sedimentation/resuspension and travel time. For these reasons, it is unlikely to have a consistently smooth viral signal in wastewater, especially when monitoring small communities. However, several normalization approaches have been employed by different groups to address these uncertainties. Normalization is not yet standardized in WBS; even the word "normalization" may not be appropriate because it attempts to correct for various factors. Viral signal in wastewater should be controlled for 1) human fecal mass to account for population (e.g. using biomarkers like pepper mild mottle virus [PMMoV], crAssphage and ammonia); 2) environmental events (e.g. WWTP influent flow) and 3) transport and dispersion dynamics in the sewer (e.g. using metrics of particle suspension in wastewater). There is likely no global solution for controlling for these (and other) factors, as each sewer has unique specificities. Normalization is still an area of investigation at PHAC/NML, where collection of several normalizing variables (e.g. concentration of PMMoV, pH, mass of total solids in suspension) has been performed since the start of the federal WBS program.

Wastewater epidemiologic model

A mathematical model that describes both SARS-CoV-2 transmission at the population level and SARS-CoV-2 concentration in the wastewater (by explicitly modelling fecal shedding) was developed at PHAC/NML (25) and implemented as a publicly available R package (34). A simple representation of this model, called the wastewater epidemic model (WEM), is shown in **Figure 3**.

Figure 3: The wastewater epidemic model is based on standard mathematical modelling of disease spread in population[a]
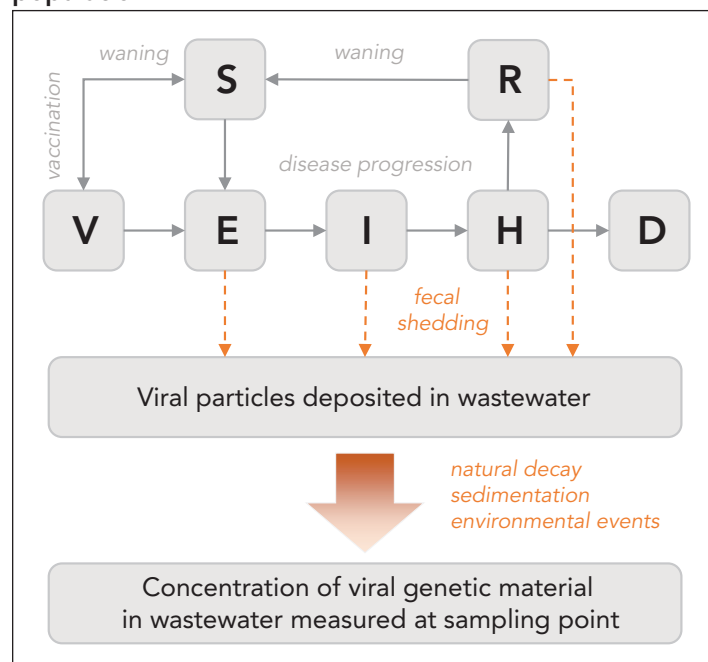
Abbreviations: D, deceased; E, latent exposure; H, hospitalized; I, infectious; R, recovered; S, susceptible; V, immunization
[a] Nourbakhsh *et al.* reference (25)

Like other mathematical models, WEM provides a principled framework to estimate unobserved epidemiological parameters (e.g. prevalence, effective reproduction number R_t) and to forecast cases, hospitalization and deaths. Importantly, WEM incorporates both the wastewater data and the traditional data based on clinical surveillance. These two data types, wastewater and clinical, can be used either in combination when more information is needed to triangulate the state of the pandemic, or as a substitute for one another when one of the two data source is missing or deemed inaccurate. We provide an example of the latter in the section analyzing the Omicron wave.

Because WEM integrates wastewater data, it translates the wastewater signal—that can be hard to interpret epidemiologically—into practical and well-known metrics for public health (e.g. prevalence, effective reproduction number). The lack of data and good understanding of the fate of viral RNA in the sewer prevented us from associating, *a priori*, the viral concentration measurement with the "true" prevalence level of infection in the catchment area of a WWTP. Thus, we

were limited to estimating prevalence as if it was reported by clinical surveillance. This means we considered the historical data points of both the viral concentration in wastewater and the reported clinical prevalence to calculate their average ratio. We used this ratio to convert viral concentration into estimated "reportable" cases in WEM (i.e. reportable cases=ratio x viral concentration) (25). In other words, we did not try to estimate the reporting fraction. Although technically possible with WEM, we did not attempt to forecast hospitalizations or deaths because these data were not available to us at the sewer shed level (i.e. sub-municipal level), thus preventing us from fitting the model parameters associated with hospitalization and mortality. Hence, we limited our forecasts to reportable cases and the Supplemental material **Figure S1** shows our estimations for five Canadian cities.

We did not see significant differences in the forecasts produced by WEM, whether the concentration was normalized by PMMoV or not. Hence, we decided to simply use raw (unnormalized) SARS-CoV-2 concentration in wastewater since normalization is still an area of investigation at NML. The case forecasts are key indicators in planning public health actions because they predict the transmission of disease at the population level. We monitored the four-week forecast accuracy of WEM using log scores (35).

The effective reproduction number (R_t) is another important measure that summarizes the current state of transmission dynamics. We show R_t estimates obtained from WEM in the Supplemental material **Figure S2**. These epidemiological indicators of virus transmission played an important role in the national COVID-19 surveillance, and modelling allows to incorporate information from WBS to enhance the estimation of these indicators.

Wastewater-based surveillance reporting

Wastewater-based surveillance is, by nature, conducted locally—typically at the level of a municipality (sampling at a WWTP), a neighbourhood (sampling in a manhole) or an institution (e.g. hospital, university campus). When data from several sampling sites are available, it may be more relevant to aggregate the data to provide a trend indicator for a broader geographical area. A possible approach to aggregate viral concentrations in wastewater from different sites is to perform a weighted average where the weights represent the population sizes of each catchment area. Of course, the viral concentrations must be standardized beforehand.

To inform its analyses, PHAC aggregates WBS from samples collected at the wastewater treatment plants to municipal and national levels. PHAC analyzes WBS through the lens of modelling; hence, the weighted average aggregation is performed on the epidemiological metrics (e.g. forecasted incidence, R_t) after fitting WEM to the data of each sampling site. In other words, we do not fit WEM to an aggregated wastewater

signal. Wastewater-based surveillance is reported in combination with clinical surveillance and modelling forecasts to show the wastewater concentration and cases to date, and predictions based on WEM.

Application to the analysis of the Omicron wave in Canada

The Omicron variant of SARS-CoV-2 was classified as a variant of concern on November 26, 2021 (16). By January 2022, over 90% of SARS-CoV-2 samples collected in Canada were identified as Omicron (36). Omicron spread rapidly across Canada, which prompted a change in testing policies to restrict PCR testing to high-risk or vulnerable populations in many jurisdictions to meet the overwhelming demand. This change likely led to an underestimation of disease burden by clinical surveillance. Importantly, case forecasts from models using the case data could no longer serve as reliable indicators to inform public health policies. In fact, in all five cities analyzed with WEM, wastewater viral loads increased concordantly with clinical cases, but the trends diverged with the implementation of PCR testing restrictions (Supplemental material, Figure S1). While clinical cases appeared to have peaked around the date of the restriction, wastewater signals remained elevated or continued to increase. The discordance between clinical surveillance and WBS during the Omicron wave emphasized the utility of WBS when clinical testing was restricted (3).

To assess the impact of the data source on case estimates and forecasting with WEM in absence of reliable clinical testing, the model was calibrated alternatively to clinical and WBS data. In addition, the model parameters representing the asymptomatic proportion and vaccine efficacy—assumed constant for all the waves before Omicron—were calibrated on available Omicron-specific data once they became available (e.g. early studies on vaccine effectiveness). After these adjustments were made, data from WBS, clinical surveillance and model forecasts were reported with epidemiological interpretations for internal monitoring of the national SARS-CoV-2 trends (Figure 1). The WEM provided estimates of reportable cases (i.e. clinical cases that would have been reported without PCR testing restrictions) using wastewater data only, in comparison with actual reported clinical cases, to assess the extent of under-reporting and the likelihood of having passed peak incidence of the wave. In **Figure 4**, we illustrate how modelling outputs were used in two different cities during the Omicron wave. In this example, WEM was fitted alternatively to clinical or wastewater data in Toronto (the largest city in Canada) and Edmonton (a medium-size city). The model suggests that under-reporting of cases in the former was more pronounced than in the latter. This modelling analysis of the Omicron wave, together with the estimates for five cities over a longer period presented in the Supplemental material, Figure S1, highlighted the limitations of clinical surveillance, especially after the change in PCR testing guidelines. From WBS, the under-reporting of cases was evident through the

comparison of cases estimated from clinical surveillance and WBS. Moreover, WBS complemented the information from clinical surveillance including the timing of the peak and increasing/decreasing trends. Overall, the Omicron wave in Canada has allowed for an appreciation for the utility of WBS as an alternative approach to monitor SARS-CoV-2 transmission when clinical surveillance became overwhelmed and struggled to provide high quality data on disease prevalence trends.

Figure 4: Example of model output interpretation during the Omicron wave
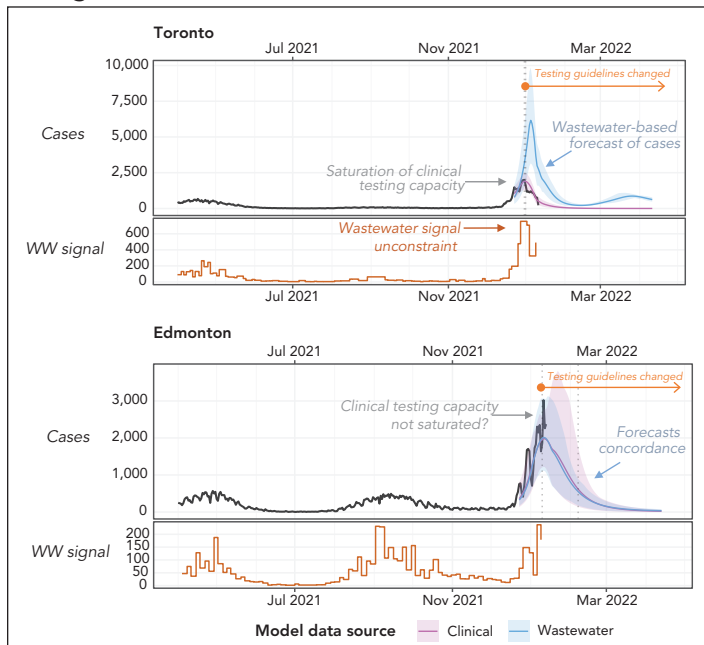
Abbreviation: WW, wastewater

Discussion

Limitations of wastewater-based surveillance in Canada

Currently, the wastewater-based modelling focuses on five major cities in Canada. While the combined catchment area of WBS for these five cities is about 23% of the Canadian population (37), it cannot provide a comprehensive overview of the SARS-CoV-2 trends with the limited scope of surveillance. It is not clear what minimum proportion of the population should be monitored through WBS to provide a reliable estimate of national prevalence. Monitoring large cities may be a good starting point to assess the intensity of transmission nationwide because most of the transmission likely occurs there. Although the expansion of the Canadian Wastewater Survey may increase the coverage of WBS, several challenges are anticipated given the geography and population distributions in Canada.

First, WBS in small or remote communities will require different sampling methods, such as sample collection from a septic tank, manhole or lagoon, due to the absence of WWTPs in such areas. Although previous research has demonstrated that sampling from manholes, did not result in significant RNA decay (38), it poses as a logistical challenge. Moreover, our current modelling framework (WEM) is not adapted to analyze small populations, mainly because WEM is not a stochastic model.

Although still an area of active research, controlling for uncertainty in the viral signal in wastewater, such as fecal shedding dynamics and in-sewer RNA decay, is critical. Since the viral signal is meant to be used to inform public health, normalization may improve its accuracy in estimating the prevalence of infections. The uncertainty of the efficacy of normalization techniques, at PHAC/NML but also for many other groups, is currently a limitation that hampers the interpretation of WBS and an area of active research.

Beyond COVID-19
The implementation of WBS as a routine surveillance tool has broader implications beyond COVID-19. Wastewater-based surveillance can also be used for monitoring respiratory pathogens other than SARS-CoV-2 (including influenza viruses, respiratory syncytial virus), sexually transmitted infections, antibiotic resistance and antibiotic use in the community (39,40). Importantly, the active research of WBS during the COVID-19 pandemic allowed for a better understanding of in-sewer factors, environmental factors and population dynamics that affect WBS and the development of mathematical modelling to estimate population prevalence of the health risk and its future predictions. However, we note that for any pathogen surveyed in wastewater, it is critical to understand its fecal shedding dynamics and in-sewer decay to improve estimates of infection prevalence in the community from viral concentration measured in wastewater. Unfortunately, there is a dearth of such clinical studies, even for SARS-CoV-2. While the expansion of WBS to other pathologies will require the development of novel laboratory assays, the current framework and knowledge of WBS and modelling with WEM will provide a strong foundation to facilitate the surveillance of other infectious pathogens.

Next steps
While the present framework provides a comprehensive WBS pipeline for the current scope of national WBS, changes and improvements can be implemented to respond to the dynamic nature of the COVID-19 pandemic. A crucial step in further developing WBS is to standardize the surveillance data, including its measurement metrics and storage, across many laboratories. The Public Health Environmental Surveillance Open Data Model (41) is an initiative to develop an open data structure, including metadata and vocabulary, to support environmental surveillance such as WBS. PHAC is in the process of incorporating its national WBS into the Public Health Environmental Surveillance Open Data Model to augment its capacity to monitor multiple

pathogens and geographical locations for WBS—facilitating the scalability of data analysis, thanks to its standardized data structure. In addition to incorporating data from concurrent WBS programs, WBS has the potential to expand to more geographical locations with diverse environments, such as remote or small communities. However, remote communities pose unique challenges because they often lack a WWTP and require alternative sampling methods for WBS. Hence, the framework may also expand to incorporate data analysis processes from these varying sources of WBS samples to standardize the analyses. Lastly, WBS can serve as an indicator of emerging variants of concern through SARS-CoV-2's genome sequencing. Although this is currently conducted at NML, the epidemiological interpretations of the results are not yet incorporated in the WBS pipeline described here.

Conclusion

Although WBS has previously been used to inform public health responses for other health risks, the COVID-19 pandemic stimulated an expansion of WBS to an unprecedented scale. As demonstrated during the Omicron wave, COVID-19 WBS has the potential to have high policy implications, especially when traditional epidemiological surveillance methods are curtailed. The present framework outlines the first national WBS of COVID-19 in Canada. In particular, the use of mathematical modelling is a critical tool to interpret WBS because it translates wastewater concentrations into prevalence for easier interpretation in public health settings. While WBS of COVID-19 provides unique information on the community spread of SARS-CoV-2, there remain many uncertainties and inconsistencies to be addressed in WBS data. The establishment of this framework will support further expansion and development of the WBS program, including monitoring other geographical areas and other pathogens.

Authors' statement

MJJ — Writing original draft, data analysis, review and editing, final approval
CSM — Data acquisition, data analysis, review and editing, final approval
EMM — Data acquisition, writing original draft of Supplemental material, data analysis, review and editing, final approval
ANagasawa — Data acquisition, data analysis, review and editing, final approval
ANichani — Data acquisition, review and editing, final approval
CPI — Data acquisition, data analysis, review and editing, final approval
SWP — Data acquisition, writing original draft of Supplemental material, data analysis, review and editing, final approval
DC — Writing original draft, writing original draft of Supplemental material, data analysis, review and editing, final approval

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Competing interests

None.

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Supplemental material

These documents can be accessed on the Supplemental material file.

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Evolution of nucleic acid amplification testing across Canada as observed through the Canadian Laboratory Response Network's SARS-CoV-2 Proficiency Test Program, May 2020 to June 2021

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Abstract

To help accommodate the surge in severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) clinical testing due to the coronavirus disease 2019 pandemic, the decentralization of testing from provincial public health laboratories to regional laboratories and private facilities was necessary. To further support the growing number of test sites in Canada, the National Microbiology Laboratory developed a proficiency test program for the detection of SARS-CoV-2 using nucleic acid amplification tests and administered it under an arm of the Canadian Laboratory Response Network (CLRN). Since its conception in May 2020, CLRN has conducted three proficiency test schemes, from May 2020 to June 2021, and has observed an increase in participation of more than 400%. This article will explore the evolution of CLRN's SARS-CoV-2 Proficiency Test Program and its support of the Canadian pandemic response.

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Introduction

In December 2019, a virus capable of causing acute respiratory disease in humans was reported in the Wuhan area, within the Hubei province of China. Since then, this virus, severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has spread rapidly around the world and has led to the most significant global pandemic of the 21st century. Rapid identification and contact tracing are essential to maintaining and managing critical public health infrastructure. Due to the ever-growing number of coronavirus disease 2019 (COVID-19) cases, it was necessary to establish decentralized testing and equipment laboratories, hospitals and healthcare centres with the ability to conduct SARS-CoV-2 diagnostics independently. Nucleic acid amplification tests (NAAT) are the current standard for diagnosis. In addition to equipping and training these centres for testing, accreditation and licensing to conduct SARS-CoV-2 testing were required.

In April 2020, a request for support from the Canadian Public Health Laboratory Network was made to the Canadian Laboratory Response Network (CLRN) for the rapid provision of a SARS-CoV-2 proficiency test program to aid the provincial and regional public health partners, since commercial proficiency test programs were not available at the time. The first CLRN SARS-CoV-2 proficiency test scheme was distributed through the National Microbiology Laboratory in May 2020, a second one in November 2020 and a final one in June 2021. Since then, a number of national and international organizations have developed open-participation proficiency test programs for SARS-CoV-2, allowing for de-escalation of this national emergency support measure. Concurrent with the CLRN testing program, in March 2021, the Canadian Microbiology Proficiency Testing organization deployed their first SARS-CoV-2 proficiency test scheme, consisting of four test samples, simulating fresh swab specimens with three shipments per year (1). The College of American Pathologists distributed their first SARS-CoV-2 molecular test scheme in November 2021, consisting of three liquid simulated respiratory specimens, with two shipments per year (2). Quality Control for Molecular Diagnostics, another international external quality assurance provider, delivers a five-specimen SARS-CoV-2 panel annually (3). The World Health

Organization also hosts an external quality assurance program for SARS-CoV-2; however, this program is limited to national and subnational laboratories around the world (4).

This article discusses the various trends and insights into SARS-CoV-2 testing observed in Canada from May 2020 to June 2021 through the delivery of the CLRN SARS-CoV-2 Proficiency Test Program.

Results and discussion

Three CLRN proficiency test schemes, which make up the CLRN Proficiency Test Program, were distributed to public health partners between May 2020 and June 2021. Participants were provided with six contrived-clinical test samples containing inactivated virus and were asked to employ their respective laboratory's algorithms for NAAT to detect the presence of SARS-CoV-2. As the pandemic evolved, testing demands and COVID-19 cases across the country increased dramatically (**Figure 1**). As such, it was necessary to further decentralize testing and expand testing centres to include regional hospitals and private laboratories. By June 2021, significant scale up by every province and territory was evident, including the increase in testing capacity in northern, remote and isolated communities, which normally would have depended on large urban facilities (Figure 1) (5,6). This trend was reflected over the course of three CLRN proficiency test schemes. The CLRN engaged with provincial and territorial jurisdictional partners to identify appropriate participants. Fifty-three laboratories participated in the May 2020 test scheme and participants increased to 118 and 214 for the November 2020 and June 2021 test schemes, respectively (**Figure 2**). Increasing participation at all Canadian jurisdictional levels from May 2020 to June 2021 was observed; provincial laboratories increased site participation by 160% (nine new participating centres), regional hospital participation grew by 443% (120 new participating centres) and private laboratories expanded by 550% (18 new participating centres) (**Figure 3**).

Furthermore, we observed a 285% increase, between May 2020 to June 2021, in laboratory participation from partners located in remote and isolated communities in northern Canada. Canadian Federal Surge Laboratories, sites that support the overflow of public health samples from provincial laboratories, participated for the first time and accounted for seven new centres during the June 2021 test scheme (Figure 3). Finally, members of the Global Health Security Action Group, involving five international participants, participated in the June 2021 test scheme.

Increased testing nationwide correlated with an increase in test panel requests and result submissions: from 69 and 73 for the May 2020 test scheme, respectively; to 206 and 194 for the November 2020 test scheme, respectively; and 368 and 394 for the June 2021 test scheme, respectively (Figure 2). Additional breakdown by geographical or population demographics

Figure 1: Population and SARS-CoV-2 demographics across Canada[a]
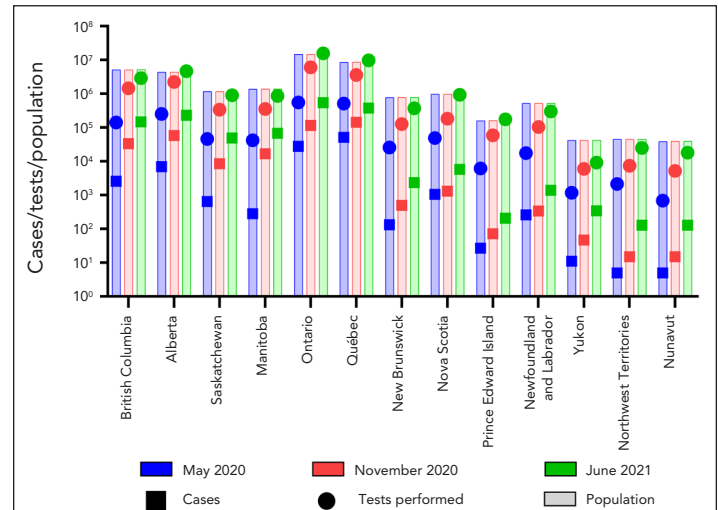
Abbreviation: SARS-CoV-2, severe acute respiratory syndrome coronavirus 2
[a] The number of SARS-CoV-2 cases, tests performed, and case counts across the country were tracked using data collected from the Government of Canada's Coronavirus disease (COVID-19): Outbreak update reference page (5). Population counts at each time point were determined using the Government of Canada's population estimates tool (6). A breakdown of cumulative COVID-19 cases per province is presented (square). A breakdown of cumulative COVID-19 tests performed per province are presented (circle). An estimated of provincial/territorial population is presented (faded bar)

Figure 2: The number of participating sites, requested panels and results submitted over the course of the three CLRN SARS-CoV-2 proficiency test schemes
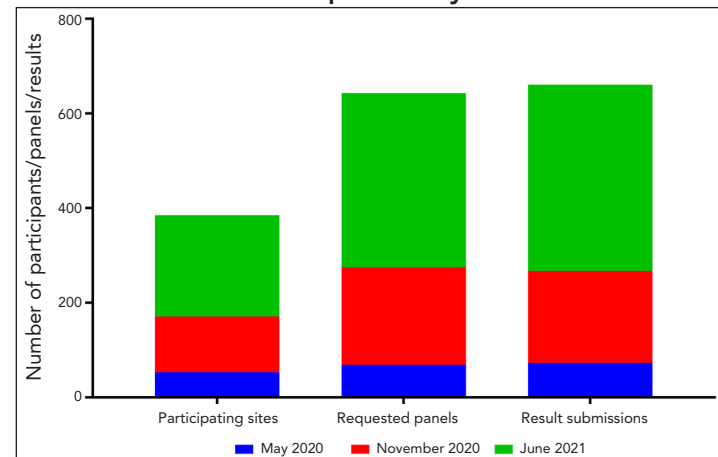
Abbreviations: CLRN, Canadian Laboratory Response Network; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2

was not possible since collection of secondary metadata was not done and variations between jurisdictional participation due to resource limitations at the time would misrepresent any observations that could be made. As participation in this proficiency test program was successfully embraced by all partners, there were logistical challenges surrounding the facilitating of the large-scale test distribution in a short period of time. Future planning needs to be mindful of inter-provincial networks, available resources and rapid deployment of material transfer agreements.

Figure 3: The number of jurisdictional laboratory centres participating in CLRN's SARS-CoV-2 Proficiency Test Program period

Abbreviations: CLRN, Canadian Laboratory Response Network; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2

The variety of nucleic acid extraction and reverse transcription polymerase chain reaction (RT-PCR) platforms expanded and correlated with the surge in countrywide testing; there was a 227% and 383% rise in different extraction and RT-PCR platforms used, respectively (**Figure 4**).

Figure 4: The number of different extraction and real-time PCR platforms used in each CLRN's SARS-CoV-2 proficiency test scheme

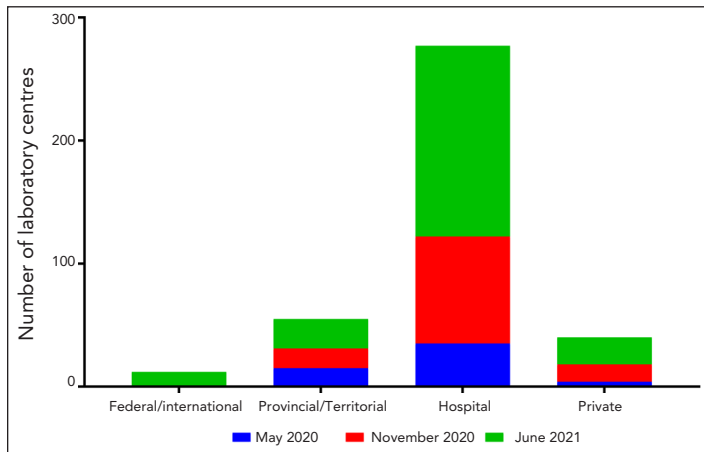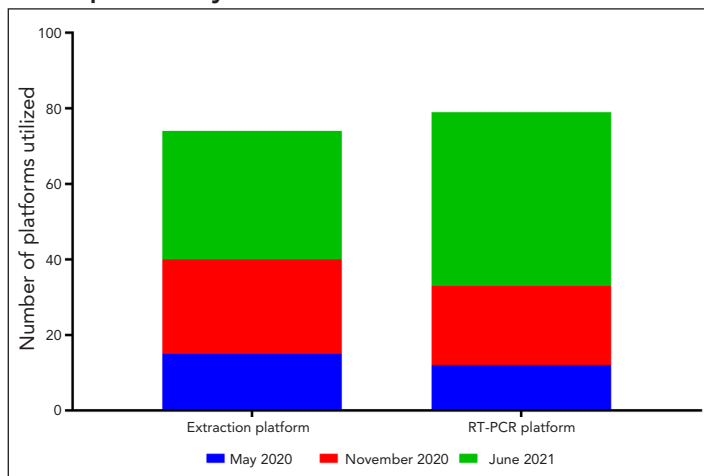Abbreviations: CLRN, Canadian Laboratory Response Network; RT-PCR, reverse transcription polymerase chain reaction; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2

The May 2020 proficiency test scheme had 53 participants submitting 73 sets of panel results (all results were as expected), while the November 2020 test scheme had 118 participants submitting 194 sets of results (94.3% obtaining expected results) and the June 2021 test scheme had 214 participants submitting 394 sets of results (99.5% obtaining expected results). Consistent with the high success rates, results were comparable across provincial, regional and private facilities, with no discernable

pattern associated with discordant results. The only exception was seen during the November 2020 test scheme, where a marginally lower success rate was observed in comparison to the other two test schemes. These results correlated with the inclusion of regional facilities to support an increase in testing capacity. Result discrepancies were identified and corrective actions were proposed through the evaluation portion of the test program. Successful remediation and functional workflows were observed in the subsequent June 2021 test scheme.

As capacity grew across Canada and as the pandemic approached the 2020 "cold and flu" season, the need for laboratories to distinguish between SARS-CoV-2 and other respiratory pathogens of significance grew. Many testing facilities began running multiplexed RT-PCR assays or equivalent assays to test for a multitude of respiratory pathogens, including SARS-CoV-2. To support this, participants had the option to report on other respiratory pathogens that may have been detected during their testing. The November 2020 test scheme was modified and made up of six contrived-clinical samples: three samples containing SARS-CoV-2, one sample containing both SARS-CoV-2 and respiratory syncytial virus (RSV), one sample containing influenza A virus, and one sample with no virus. Twenty-four participants implemented testing parameters to detect influenza A virus and 22 implemented testing for RSV; all participants correctly identified the samples containing these viruses. The June 2021 test scheme extended these parameters to consist of two contrived-clinical samples containing varying amounts of SARS-CoV-2, two contrived clinical samples containing SARS-CoV-2 and rhinovirus or influenza B virus, one contrived clinical sample containing influenza A virus and one containing no virus. Fifty-four sites employed rhinovirus testing, 116 sites implemented testing parameters for influenza A virus and 106 sites conducted influenza B virus testing; in all cases the various viruses were correctly identified in their respective samples. Only one discordant result was observed, an equivocal RSV result was obtained for a sample containing SARS-CoV-2 only.

The emergence of SARS-CoV-2 variants of concern (VOCs) became a reality in the latter part of 2020. The first SARS-CoV-2 VOC (B.1.1.7) is suspected to have emerged in the United Kingdom, with the earliest samples reported in September 2020, and had spread to multiple countries by December 2020 (7,8). Ontario confirmed Canada's first case of the B.1.1.7 variant on December 26, 2020, and by April 26, 2021, all provinces and territories had reported confirmed cases. Since then, VOCs continued to emerge and spread throughout the world (8) and laboratories and reference facilities began developing assays to identify and flag VOCs. To further support these laboratories, the June 2021 CLRN SARS-CoV-2 proficiency test scheme incorporated three SARS-CoV-2 VOCs into the test panel. The June 2021 test scheme had samples containing the SARS-CoV-2 wild-type virus, B.1.1.7, B.1.351 or P.1 variants. Forty-seven participants performed a variety of short nucleotide

polymorphism (SNP) assays and two participants conducted whole genome sequencing. Sixty-eight percent of participants identified the sample containing the B.1.1.7 variant, while 24% reported detection of an unspecified variant and 8% were incorrect or undetermined. Thirty-nine percent of participants identified the sample containing the B.1.351 variant, while 59% identified either B.1.351/P.1 variants, and 2% reported detection of an unspecified variant. Twelve percent of participants correctly identified the sample containing the P.1 variant, while 51% identified either B.1.35/P.1, 22% reported detection of an unspecified variant, and 14% were incorrect or undetermined. Finally, 78% of participants correctly identified the wild-type strain while 22% were undetermined. Overall, the majority of sites were able to identify the presence of a VOC; however, typing the variant utilizing SNP assays was inconsistent due to a limited combination of assays being used and would require additional SNP assays or genomic sequence analysis to get a definitive lineage. For example, the B.1.351 and P.1 variants both share an E484K and N501Y mutation in the spike protein; without a distinguishing target, such as K417N/T, identifying a lineage would not be possible.

Therefore, understanding the objective and subsequent public health outcome is necessary to determine the complexity of the workflows required. While whole genome sequencing provides a large dataset, there are a number of advantages to using SNP assays: higher throughput; increased sensitivity; reduced impact on resources and infrastructure; and better cost effectiveness.

Conclusion

The provision of the CLRN SARS-CoV-2 Test Program from May 2020 to June 2021 demonstrated the scaleability of Canadian public health external quality assurance programs through the CLRN. Having a centralized Canadian proficiency test program enabled the laboratory network to identify performance metrics and considerations, such as the need to expand testing assays for VOC identification, if laboratories prefer to discriminate between circulating VOCs with a PCR screen. The comprehensive program also demonstrated the fluidity of the public health system in Canada to adapt to rapidly changing environments. A hallmark of the Canadian laboratory response to the COVID-19 pandemic was the rapid and successful implementation of testing laboratories across the country to accommodate the surge in testing requirements. Whether it was increasing testing capacity in urban settings through the participation of hospital laboratories, private facilities and federal surge sites, or implementing testing centres in remote and isolated communities in northern Canada, the CLRN SARS-CoV-2 Proficiency Test Program clearly demonstrated successful surge capacity while maintaining testing standards, providing Canadians with rapid identification of SARS-CoV-2 infection.

Authors' statement

CR — Conceptualization, data analysis, writing–original draft, writing–review
KA — Conceptualization, writing–original draft, reviewed final draft
CC — Conceptualization, writing–original draft, reviewed final draft

Competing interests

None.

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Comparison of fifteen SARS-CoV-2 nucleic acid amplification test assays used during the Canadian Laboratory Response Network's National SARS-CoV-2 Proficiency Program, May 2020 to June 2021

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Abstract

Background: On March 11, 2020, the World Health Organization declared a pandemic caused by the recently emerged severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). This led to increased clinical testing and decentralizing of this testing from provincial health laboratories to regional and private facilities. Leveraging the results from the Canadian Laboratory Response Network's National SARS-CoV-2 Proficiency Test (PT) Program, this study compares multiple commercial and laboratory-developed nucleic acid amplification tests, assessing both sensitivity and specificity across multiple users.

Methods: Each panel consisted of six blinded, contrived-clinical samples. Panels were distributed to international, provincial and territorial laboratories and subsequently to partner facilities. Participating laboratories were asked to run these sample through their respective extraction/PCR workflows and submit results to the National Microbiology Laboratory, outlining the nucleic acid extraction platform and nucleic acid amplification test employed, as well as the viral gene target and Ct values or equivalent obtained. Data were compiled for each molecular platform and gene target used.

Results: The PT schemes were deployed in May 2020, November 2020 and June 2021, resulting in 683 data sets using 37 different nucleic acid amplification tests. Over the course of three PT schemes, the average score obtained was 99.3% by participants demonstrating consistent testing between laboratories and testing platforms.

Conclusion: This study confirmed the rapid and successful implementation of a Canadian PT Program and provided comparative analysis of the various emergency use authorized and laboratory developed tests employed for the detection of SARS-CoV-2 and demonstrated an overall 99.3% test concordance nationwide.

Introduction

In late 2019, a novel respiratory virus, severe acute respiratory coronavirus 2 (SARS-CoV-2), emerged in the Hubei province of China and subsequently caused the coronavirus disease 2019 (COVID-19) global pandemic. As the case numbers rapidly grew, it became necessary to decentralize testing to support testing at the federal, provincial/territorial and municipal levels, including private laboratories, hospitals and healthcare facilities.

The Canadian Laboratory Response Network (CLRN) at the National Microbiology Laboratory (NML) in Winnipeg, Canada provides high-consequence proficiency panels for biothreat agents to ensure that public health laboratories are ready to respond with high quality diagnostic testing. During the COVID-19 pandemic, the CLRN was leveraged to develop a Proficiency Test (PT) program to support facilities conducting SARS-CoV-2 clinical testing using molecular methods. Similar to other international efforts, the National SARS-CoV-2 PT Program supports the ability of public health testing facilities to establish competency and obtain or maintain accreditation to conduct SARS-CoV-2 clinical testing against a known reference standard to ensure consistency between testing platforms and laboratories across the country and across the globe (1–3). Nucleic acid amplification tests (NAAT) have been considered the gold standard method for the detection of active SARS-CoV-2 cases. Since the emergence of SARS-CoV-2 in December 2019, there have been a variety of NAATs developed, both laboratory-developed tests and commercial assays. This study provides a comparison of the various NAAT platforms employed within Canada over the course of three PT schemes from May 2020 to June 2021.

Materials and methods

Production, quality control and panel distribution

Irradiated viruses were diluted in a pooled, negative human nasal secretion as the background matrix at varying concentrations and immediately aliquoted into pre-labelled tubes. Each panel consisted of six blinded, contrived-clinical samples. Samples were sorted by site number, packaged appropriately for transport and stored at −80°C until distribution.

Prior to distribution, quality control measures were taken to ensure sample homogeneity and stability. In short, ten aliquots of each sample were removed from storage, nucleic acids were extracted as per manufacturer's instructions (MagMax™ CORE Nucleic Acid Purification Kit, Applied Biosystems™, Ontario) and assayed by quantitative real-time polymerase chain reaction (qRT-PCR) (QuantiNova® Probe RT-PCR Kit, Qiagen®, Ontario) targeting the E gene of SARS-CoV-2 (4). Coefficient of variations were calculated for each set of panel samples using GraphPad® Prism's descriptive statistics. An average Ct value with a coefficient of variation less than 10% was necessary to pass sample homogeneity quality controls. Stability testing began day 1 post-production and continued at specified intervals for the duration of the PT scheme using the same approach outlined above. If quality controls passed for homogeneity and stability testing on day 1 and seven post-production, the panels were released for distribution. Stability testing continued for the duration of the test scheme.

Panels were packed on dry ice and distributed to the international, provincial and territorial laboratories, who subsequently distributed panels to their partner facilities within their jurisdiction. Cold chain was monitored and if not maintained, a new panel was shipped directly from NML.

Participant selection and intended use

Provincial and territorial members of the Canadian Public Health Laboratory Network (CPHLN) approached the NML to assist the pandemic response by producing and administering a SARS-CoV-2 PT Program, as one was not readily available at the time. The CPHLN provincial and territorial partners provided NML with a list of participants and were responsible for distribution of the test panels within their respective jurisdictions. Participants included provincial and territorial laboratories, public health laboratories, hospitals and healthcare facilities in both urban and rural communities. Specific metadata and details on individual site licensing and accreditation for SARS-CoV-2 were not made available to NML.

The PT panel was intended to be used as an internal validation of SARS-CoV-2 molecular processes, which are performed in conjunction with a nucleic acid extraction method. This panel was not intended to be used on platforms requiring fresh swab material, or the detection of viral antigens or virus-specific antibodies.

Test result submission and analysis

Participating laboratories submitted results to NML outlining the nucleic acid extraction platform and NAAT employed, as well as the viral gene target and Ct values or equivalent obtained. Data were compiled for each molecular platform and gene target used. Coefficient of variation for each gene target within a single platform was determined using GraphPad® Prism's descriptive statistics. Probit analysis using a 95% cut-off was used to determine limit of detection based on sample detection (5).

Results and discussion

The PT schemes were deployed in May 2020, November 2020 and June 2021, resulting in 683 data sets using 37 different NAAT (**Table 1**). Each PT scheme assessed assay sensitivity and specificity. The most commonly used platforms were fully automated low-throughput assays such as the DiaSorin Simplexa™ COVID-19 Direct Molecular Assay, Cepheid Xpert® Xpress SARS-CoV-2, Cepheid Xpert® Xpress SARS-CoV-2/Flu/RSV and BioFire® FilmArray RP2.1 Test Panel. These systems were employed mainly in hospital laboratories and in rural communities. Larger diagnostic centres, such as provincial laboratories and reference centres, generally employed high-throughput assays, including the Roche Cobas® SARS-CoV-2 Test (for Cobas 6800/8800), Seegene Allplex™ 2019 nCoV Assay, Thermo Fisher TaqPath™ COVID-19 Combo Kit and LDT targeting the E gene (Table 1).

Table 1: Nucleic acid amplification test platforms utilized for the detection of SARS-CoV-2 during the Canadian Laboratory Response Network's SARS-CoV-2 Proficiency Test Panels, May 2020 to June 2021

Nucleic acid amplification test platform		Proficiency test scheme, Number of sites/platform		
Manufacturer	Product name	May 2020	Nov 2020	June 2021
Abbott™	Alinity™ m SARS-CoV-2 AMP Kit	0	5	16
	SARS-CoV-2 Real Time PCR	1	3	3
Agena Bioscience	MassARRAY® SARS-CoV-2 Panel	0	0	1
Altona	AltoStar® SARS-CoV-2 RT-PCR Kit 1.5	1	1	2
BD	SARS-CoV-2 Reagents for the BD MAX™ System	2	9	4
BGI™	Real Time Fluorescent RT-PCR Kit for detecting SARS-CoV-2	0	2	1
BioFire®	Film Array® Respiratory 2.1 Panel	0	20	49
Biomeme	SARS-CoV-2 Go Strips™	0	1	1
Cepheid	Xpert® Xpress SARS-CoV-2	34	36	52
	Xpert® Xpress SARS-CoV-2/Flu/RSV	0	0	29
DiaSorin	Simplexa™ COVID-19 Direct Molecular Assay	5	42	81
Hologic	Panther Fusion® SARS-CoV-2 Assay	0	2	2
	Aptima® SARS-CoV-2 Assay (Panther System)	0	6	8
Hyris	Virus Finder COVID-19 bKit™	0	0	1
Laboratory-developed test	3' UTR Target	0	0	1
	5' UTR Target	0	2	4
	CDC CoVPlex Real-Time PCR Assay	0	0	1
	E Gene Target	12	27	49
	N Gene Target	1	1	10
	ORF1a/b Gene Target (RdRp)	5	5	8
	S Gene Target	0	1	0
	E and N Gene Pooled Targets	0	1	6
	E and ORF1a/b Gene Pooled Targets	0	0	1
	N, ORF1a/b and S Gene Pooled Targets	0	1	1
Luminex	Aries® SARS-CoV-2 Assay	0	1	1
	NxTAG® Respiratory Pathogen Panel + SARS-CoV-2	0	1	1
LuminUltra	GeneCount® COVID-19 RT-qPCR Assay	0	0	1
Quidel	Lyra® SARS-CoV-2 Assay	0	0	3
	Solana® SARS-CoV-2 Assay	0	0	1
RIDA® Gene	SARS-CoV-2 Test	0	2	1
Roche	Cobas® SARS-CoV-2 Test (for Cobas 6800/8800)	13	6	19
	Cobas® SARS-CoV-2 & Influenza A/B Test (for Cobas 6800/8800)	0	0	1
	Cobas® SARS-CoV-2 (for Liat®)	0	0	1
	Cobas® SARS-CoV-2 & Influenza A/B Assay (for Liat)	0	0	9
Seegene	Allplex™ 2019 nCoV Assay	4	19	19
	Allplex™ SARS-CoV-2/FluA/FluB/RSV Assay	0	0	1
ThermoFisher Scientific	TaqPath™ COVID-19 Combo Kit	1	6	15
Total number of results submitted		79	200	404

Abbreviations: CDC, Centers for Disease Control and Prevention; COVID-19, coronavirus disease 2019; PCR, polymerase chain reaction; RT-PCR, real-time polymerase chain reaction; RSV, respiratory syncytial virus; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2

Panel results obtained using commercially available NAATs that have at least three datasets in any given test scheme are presented in **Figure 1**. Infrequently used platforms were not assessed further. Abbott produces two high-throughput, laboratory-based molecular assays for the detection of SARS-CoV-2: the Alinity m SARS-CoV-2 AMP Kit used with the Alinity m System; and SARS-CoV-2 RealTime PCR employing the m2000 RealTime System. Both systems obtained expected results for all samples across three test schemes. All sites demonstrated consistent results from November 2021 to June 2021 with coefficient of variations less than 10% (Figure 1).

Figure 1: Commercial nucleic acid amplification test performance obtained during the Canadian Laboratory Response Network's SARS-CoV-2 Proficiency Test Program, May 2020 to June 2021[a]
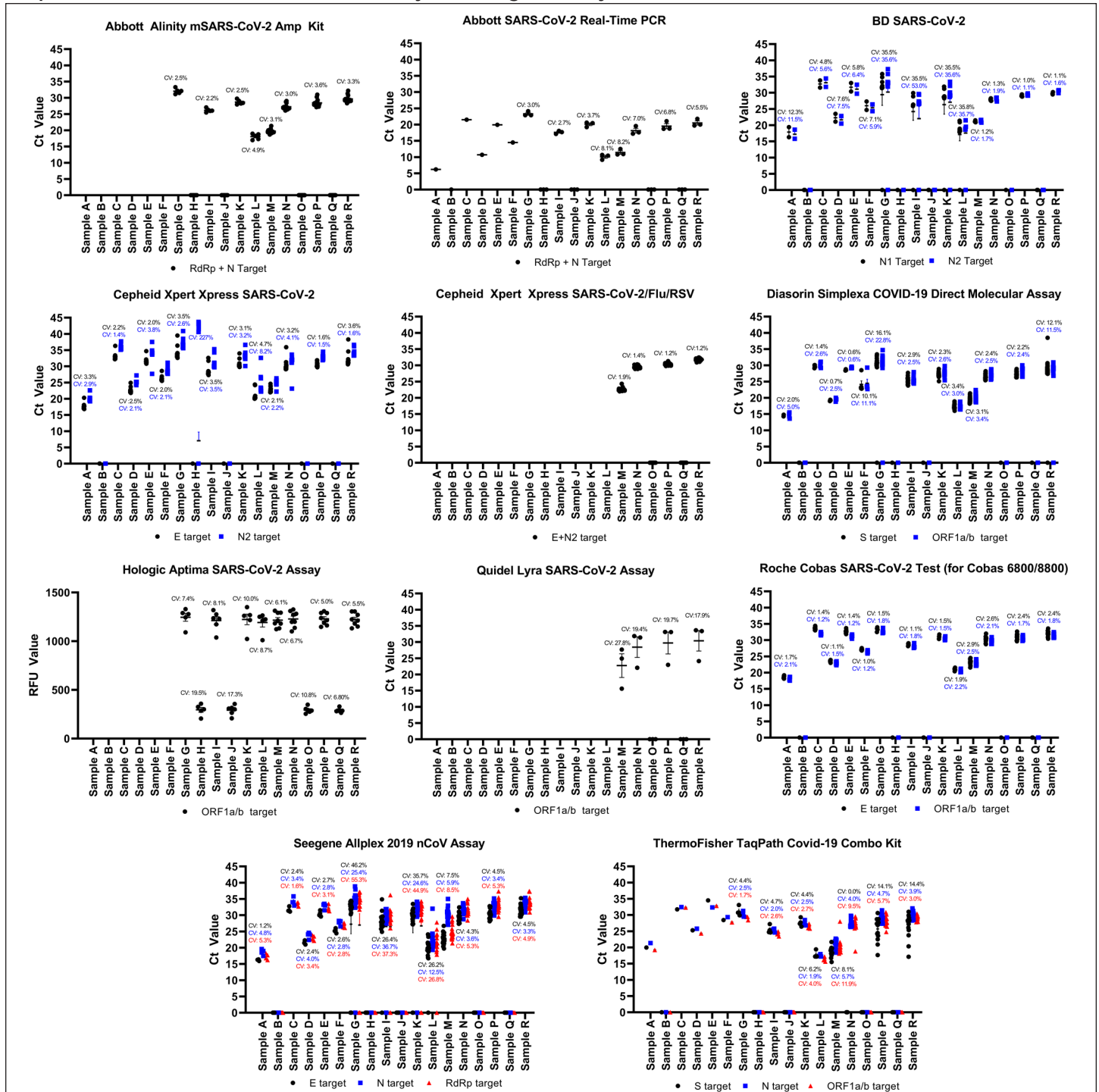
Abbreviations: COVID-19, coronavirus disease 2019; PCR, polymerase chain reaction; RFU, relative fluorescence unit; RSV, respiratory syncicial virus; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2

[a] Ct values are presented for each nucleic acid amplification platform tested. Each data point is presented with the mean and standard error. The coefficient of variation is denoted for each target in its respective colour. Data points at the 0 value on the axis indicate there were no detectable SARS-CoV-2 RNA

The BD SARS-CoV-2 Reagents for the BD MAX™ System targeting the N gene were utilized for the detection of SARS-CoV-2. The BD MAX System is a fully automated system, allowing the user to run up to 24 samples at a time. Over the course of 13 months, the BD SARS-CoV-2 Reagents for the BD MAX System performed with variable accuracy. During the May 2020 test scheme, samples were accurately detected in all cases, but the coefficient of variation ranged from 4.8%–12.3%, indicating increased variation between users. Discordant results were observed during the November 2021 test scheme; 6/7 failures to detect SARS-CoV-2 were attributed to user error (Figure 1, **Table 2**); therefore, the data obtained for Sample G–L were skewed and the accuracy and consistency were negatively affected. Removing these data points would regain an overall 100% target accuracy for the N1 target and 99% accuracy for N2; the latter target failed to identify the presence of Sample I (Figure 1, Table 2). During the June 2021 test scheme, the BD SARS-CoV-2 Reagents for the BD MAX System performed with 100% accuracy. Ct values were consistent among all users denoted by a coefficient of variations of less than 5% (Figure 1).

The BioFire Film Array RP2.1 test kit uses a fully automated system to test for the presence of 22 different pathogens, including SARS-CoV-2. This assay has a nucleic acid extraction step followed by reverse transcription/nested PCR step coupled with deoxyribonucleic acid melt curve technology to identify the presence of target pathogens qualitatively. Out of 414 samples tested, it missed identifying the presence of SARS-CoV-2 in only one sample; demonstrating a 99.8 concordance rate (**Table 3**). One site was unable to detect SARS-CoV-2 in Sample P; however, it was determined that insufficient mixing of the test sample was likely responsible for the discrepant results. Furthermore, this site correctly identified the presence of other target pathogens, which were present in the samples such as rhinovirus (Sample M), respiratory syncytial virus (Sample K), influenza A virus (Sample H and O) and influenza B virus (Sample R) (Table 3).

Table 2: Nucleic acid amplification test platform discordant target results for SARS-CoV-2 obtained with the Canadian Laboratory Response Network's SARS-CoV-2 Proficiency Test Panels, May 2020 to June 2021

PCR platform	Assay target	PCR platform SARS-CoV-2 discordant results (%)	Sensitivity: 95% detection[a,b] (copies/ml)	Specificity	
				Positive agreement (%)[b]	Negative agreement (%)[b]
BD SARS-CoV-2 Reagents for the BD MAX™ System	N1	6/96 (6.25%)	1,100 or fewer	100	100
	N2	7/96 (7.29%)	1,100 or fewer	100	100
BioFire® Film Array® RP2.1	M/S	1/414 (0.24%)	1,100 or fewer	100	100
Cepheid Xpert® Xpress SARS-CoV-2	E	0/730 (0.00%)	1,100 or fewer	100	100
	N	6/730 (0.82%)	1,100 or fewer	100	97
DiaSorin Simplexa™ COVID-19 Direct Molecular Assay	ORF1a/b	3/768 (0.39%)	1,100 or fewer	99.7	100
	S	4/768 (0.52%)	1,100 or fewer	99.6	100
NxTAG® Respiratory Pathogen Panel + SARS-CoV-2	ORF1a/b	0/12 (0.00%)	1,100 or fewer	100	100
	M	1/12 (8.33%)	1,100 or fewer	100	100
RIDA® gene SARS-CoV-2 Test	E	2/18 (11.11%)	1,100 or fewer	100	100
Seegene Allplex™ 2019 nCoV Assay	E	11/252 (4.37%)	1,100 or fewer	100	100
	RdRp	14/252 (5.56%)	1,358	99.1	100
	N	9/252 (3.57%)	1,100 or fewer	100	100

Abbreviations: PCR, polymerase chain reaction; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2
[a] Assessment using Sample C, G and P
[b] Off-label procedural methods and user errors were removed from the assessment

Table 3: Qualitative performance of the BioFire Film Array Respiratory 2.1 Panel and Roche Cobas SARS-CoV-2 and Influenza A/B Assay (for Liat) during the Canadian Laboratory Network's SARS-CoV-2 proficiency test schemes, May 2020 to June 2021

Platform	Sample ID	Sample G	Sample H	Sample I	Sample J	Sample K	Sample L
BioFire® Film Array® Repiratory Panel 2.1	Expected results	Detected SARS-CoV-2	Detected Influenza A	Detected SARS-CoV-2	No agent detected	Detected SARS-CoV-2 RSV	Detected SARS-CoV-2
	Sample concordance	100% (20/20)	100% (20/20)	100% (20/20)	100% (20/20)	100% (20/20)	100% (20/20)
	Sample ID	Sample M	Sample N	Sample O	Sample P	Sample Q	Sample R
	Expected results	Detected SARS-CoV-2 rhinovirus	Detected SARS-CoV-2	Detected Influenza A	Detected SARS-CoV-2	No agent detected	Detected SARS-CoV-2 Influenza B
	Sample concordance	100% (n=49/49)	100% (n=49/49)	100% (n=49/49)	98.6% (n=48/49)	100% (n=49/49)	100% (n=49/49)
	Overall concordance						99.8% (413/414)
Roche Cobas® SARS-CoV-2 & Influenza A/B Assay (for Liat®)	Sample ID	Sample M	Sample N	Sample O	Sample P	Sample Q	Sample R
	Expected results	Detected SARS-CoV-2	Detected SARS-CoV-2	Detected Influenza A	Detected SARS-CoV-2	No agent detected	Detected SARS-CoV-2 Influenza B
	Sample concordance	100% (n=9/9)	100% (n=9/9)	100% (n=9/9)	100% (n=9/9)	100% (n=9/9)	100% (n=9/9)
	Overall concordance						100% (n=54/54)

Abbreviations: RSV, respiratory syncicial virus; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2

The Cepheid GeneXpert platform is readily used across Canada for the detection of SARS-CoV-2 employing the Xpert Xpress SARS-CoV-2 and Xpert Xpress SARS-CoV-2/Flu/RSV assays. The Xpert Xpress SARS-CoV-2 E assay performed with accuracy (100% detection rate) and consistency (coefficient of variation less than 5%) for all samples; however, discordant results were observed using the N target, specifically for Sample H. Sample H did not contain SARS-CoV-2 but did contain a moderate amount of influenza A virions (Ct 27); there were six instances where the SARS-CoV-2 N2 target produced a Ct greater than 40, which was deemed positive for SARS-CoV-2 by the GeneXpert software (Figure 1, Table 2). Apart from Sample H, the Ct values for the N target were consistent and had a coefficient of variation less than 10%, Figure 1. The recently developed Cepheid Xpert Xpress SARS-CoV-2/Flu/RSV assay was employed during the June 2021 test scheme and the result output for SARS-CoV-2 was combined for both E and N2 targets. The platform had a 100% accuracy and produced very consistent results with a coefficient of variation less than 2% among all users (Figure 1). The Xpert Xpress SARS-CoV-2/Flu/RSV assay also correctly identified the presence of influenza A and B in Samples O and R, respectively (data not shown).

The Diasorin Simplexa COVID-19 Direct Molecular Assay is a low throughput, automated system that can run up to eight samples at once. Its main distinction from other similar systems, such as the BioFire Film Array and Cepheid GeneXpert platforms, is that it eliminates the nucleic acid extraction/purification step. Discordant results were observed for Sample G and Sample R, the ORF1a/b target missed detecting SARS-CoV-2 n=2/768 times (0.26%), while the S target did not detect SARS-CoV-2 n=3/768 times (0.39%) (Figure 1, Table 2). According to the manufacturer, the S assay has a 95.8% detection rate of 500 copies/ml (2,000 copies/ml for 100% detection) and the ORF1a/b is detected 93.8% of the time at 1,000 copies/ml (2,000 copies/ml for 100% detection (6). Similar observations were observed here: the S assay performed better than the ORF1a/b assay (Table 2). Sample G and R are approximately 1,100 and 3,500 copies/ml respectively, which is the range of the assay's limit of detection (LOD) for both targets, and is the likely cause for the discrepant results (**Table 4**). Furthermore, there was an additional discordant result for each target due to a software error that reported "no result" when Ct values were obtained for both targets (Table 2). For samples where all targets were correctly identified (Samples A–F and H–Q), coefficient of variations were 5% or less, except for Sample F which had coefficients of variations of 11.1% and 10.1% for the ORF1a/b and S targets, respectively (Figure 1).

Table 4: Sample identity and approximate viral loads for test samples provided during the Canadian Laboratory Network's SARS-CoV-2 proficiency test schemes, May 2020 to June 2021

Sample	Identity	SARS-CoV-2 E approximate copies/ml	Approximate Ct value (SARS-CoV-2 E target)[a]
CLRN's SARS-CoV-2 proficiency test scheme – May 2020			
A	SARS-CoV-2 wild type	120,000,000	20
B	Blank	0	0
C	SARS-CoV-2 wild type	1,600	36
D	SARS-CoV-2 wild type	2,700,000	25
E	SARS-CoV-2 wild type	3,900	35
F	SARS-CoV-2 wild type	216,000	29
CLRN's SARS-CoV-2 proficiency test scheme – November 2020			
G	SARS-CoV-2 wild type	1,100	36
H	Influenza A virus	0	0
I	SARS-CoV-2 wild type	54,000	31
J	Blank	0	0
K	SARS-CoV-2 wild type	10,800	33
	Respiratory syncytial virus	0	
L	SARS-CoV-2 wild type	13,000,000	22
CLRN's SARS-CoV-2 proficiency test scheme – June 2021			
M	SARS-CoV-2 B.1.351	280,000	28
	Rhinovirus	0	
N	SARS-CoV-2 B.1.1.7	2,100	35
O	Influenza A virus	0	0
P	SARS-CoV-2 P.1	1,600	36
Q	Blank	0	0
R	SARS-CoV-2 wild type	3,500	35
	Influenza B virus	0	

Abbreviations: CLRN, Canadian Laboratory Response Network; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2
[a] Corman *et al.* reference (4)

Hologic produces two SARS-CoV-2 assays that were employed during the scope of the CLRN SARS-CoV-2 PT schemes: Panther Fusion SARS-CoV-2 assay and Aptima SARS-CoV-2 assay. The Panther Fusion SARS-CoV-2 assay was not presented here as only two sites employing this platform, while the Aptima SARS-CoV-2 assay was employed during the November 2020 and June 2021 test schemes with six and eight users respectively (Table 1). This platform demonstrated 100% concordance (n=90/90 samples); however, the Ct values obtained were quite variable, with coefficients of variation ranging from 5% to 19.5% across samples (Figure 1).

During the June 2021 CLRN PT scheme, the Quidel Lyra SARS-CoV-2 Assay targeting the ORF1a/b was employed for the first time by three participants (Table 1). This assay was able to correctly identify all test samples (n=18); however, the variability between Ct values was large, with a coefficient of variations ranging from 17.9 to 27.8 (Figure 1). This variation in Ct values is largely attributed to one set of test panel results, which provided substantially lower Ct values than the other participants, indicating differences in threshold settings between participants.

The Seegene Allplex 2019 nCoV Assay is a multiplex RT-PCR assay that detects the E, N and RdRp targets and can be automated for high volume testing. This test performed well during the May 2020 and June 2021 PT schemes demonstrating a 100% concordance and consistent results conveyed by a coefficient of variation less than 10% (Figure 1); however, a number of discordant results were observed during the November 2020 PT scheme, causing subsequent decreases in reproducibility and elevated coefficients of variation. Sample G was associated with n=3/19 E target failures, n=4/19 RdRp target failures and n=1/19 N target failures. While n=2/19 RdRp target failures were associated with the use of a nucleic extraction platform, the remaining failures were associated with a divergence from manufacturer's recommendations and did not employ a nucleic acid extraction step. Furthermore, the reported LOD for the Seegene Allplex 2019 nCoV Assay is approximately 4,000 copies/ml, which is higher than the Sample G titer and is likely responsible for the failure to detect SARS-CoV-2 in this sample (7) (Table 4) Conversely, Sample I was associated with n=1/19 E target failures and n=2/19 RdRp and N target failures; while Sample K had n=2/19 E target failures, n=3/19 RdRp target failures and n=1/19 N target failures. Sample L, H and J were also associated with one discordant result for each target due to the inability to acquire a valid result. These remaining failures to detect SARS-CoV-2 were all associated with off-label use of not employing a nucleic acid extraction procedure, and are likely the cause of the discordant result since sample titers were all above 4,000 copies/ml. The practice of not implementing an extraction protocol was not observed in the subsequent test scheme. Overall, the E, RdRp and N targets produced discordances of 4.37%, 5.56% and 3.52%, respectively (Figure 1, Table 2).

Two different Roche assays were utilized during the CLRN SARS-CoV-2 PT schemes, Roche Cobas SARS-CoV-2 Test, a fully automated, high throughput assay intended for use with the Roche Cobas 5800/6800/8800, and Roche Cobas SARS-CoV-2 & Influenza A/B Assay for Liat, a fully automated qualitative point of care test to be used on the Cobas Liat. The Roche Cobas SARS-CoV-2 Test for Cobas 5800/6800/8800 was employed during all three test schemes, producing accurate and consistent results with the coefficient of variations less than 3% (Figure 1). The Roche Cobas SARS-CoV-2 Test for Liat accurately detected all test samples from nine users (Table 1 and Table 3). Overall, the Roche Cobas SARS-CoV-2 Test for use on the Cobas 5800/6800/8800 performed the best when comparing commercial platforms across the CLRN SARS-CoV-2 PT schemes; it demonstrated 100% accuracy and produced the most reproducible results across users.

The LDT were also employed during the CLRN SARS-CoV-2 PT Scheme from May 2020 to June 2021. Data sets obtained using LDTs that have at least three sets of submitted results in any given test scheme are presented (**Figure 2**). In all cases, all tests were able to detect SARS-CoV-2 effectively and accurately from the test samples provided (Figure 2). The E and RdRp targets were used in all test schemes (Table 1). The reproducibility of the E target and RdRp target ranged from coefficients of variation between 3.9% and 8.4% and between 3.2% and 10.2%, respectively (Figure 2). The use of the 5' UTR target emerged during the November 2020 test scheme and results were consistently detected with coefficients of variation less than 7% (Figure 2). Laboratories began employing the N target test during the June 2021 test scheme with coefficients of variation ranging between 6.9% and 9.6% (Figure 2). It should be noted that, apart from the targeted gene, we do not have the specific details regarding the primer/probe sequences implemented by each user and it is possible that the sequences utilized are different. In general, Ct values were similar between all the target tests indicating similar detection affinities; however, a more detailed direct comparative analysis was not conducted, since the assays were not identical. Furthermore, shifts between gene targets are expected, as individual gene expression may differ during viral replication; but this finding could also be attributed to technical variations in the threshold/detection settings by different laboratories. Overall, the 5' UTR target on average demonstrated the most consistent results with an average coefficient of variation of 4.3%, followed by the RdRp (4.7%), E (5.2%) and N (7.9%) targets. All targets performed within designated specifications of coefficients of variation of less than 10%.

Figure 2: Laboratory-developed nucleic acid amplification test performance obtained during the Canadian Laboratory Response Network SARS-CoV-2 Proficiency Test Program, May 2020 to June 2021[a]
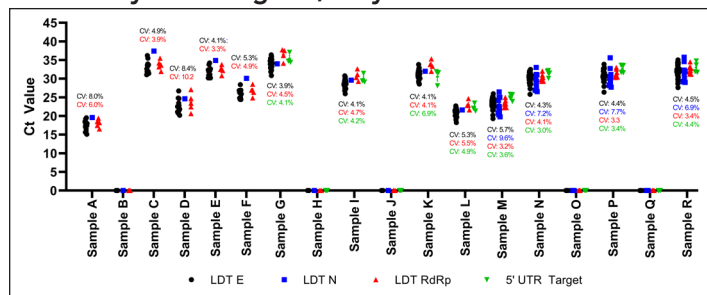
Abbreviations: LTD, laboratory-developed tests; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2
[a] Ct values are presented for each nucleic acid amplification platform tested. Each data point is presented with the mean and standard error. The coefficient of variation is denoted for each target in its respective colour. Data points at the 0 value on the axis indicate there was no detectable SARS-CoV-2 RNA

Overall, these results provide insights into test sensitivity; each test scheme involved testing a sample, which contained low concentrations of virus particles, ranging from 1,100 to 1,600 copies/ml (Sample C, 1,600 copies/ml, Sample G, 1,100 copies/ml or Sample P, 1,600 copies/ml). Effective test sensitivity was observed across all presented commercial and LDT assays employed across the country. A 100% concordance rate for these low concentration samples was observed for all SARS-CoV-2 targets, with a few exceptions. The BioFire Film Array RP2.1 test kit missed detecting Sample G 1/414 times (Table 3); however, this error occurred due to a procedural mishandling of the sample, and upon repetition for remediation purposes, it was detected. Therefore, this error was not included in the general assessment of sensitivity (Table 3).

The Diasorin Simplexa COVID-19 Direct Molecular Assay missed detecting two low concentration samples, both targets were unable to detect Sample G on two occurrences and the S target failed to detect Sample R (3,500 copies/ml) in one instant (Table 2); however, these discordant results did not cause the 95% limit of detection rate to be affected. The Seegene Allplex 2019 nCoV Assay was associated with a number of failures to detect Sample G. The majority of these failures were attributed to off-label use, where a required nucleic acid extraction process was omitted; for this reason, these results were removed from the subsequent analysis of sensitivity. However, there were two instances associated with proper use, where the RdRp target failed to identify SARS-CoV-2 and were included in the analysis. These discordant results elicited a minor effect on test sensitivity; a 95% detection limit was determined to be 1,358 copies/ml (Table 2). With the exception of the Seegene Allplex 2019 nCoV assay, all other assays had 95% detection limits below 1,100 copies/ml. These observed results are in line with the manufacturers reported limits of detection for their respective assays (6–16). While outside of the scope of the intended use of this PT scheme, this study was not able to calculate the limit of detection for all the assays due to lack of samples below detectable levels and therefore further comparison of assay sensitivity was not possible.

In addition to test sensitivity, specificity of the assays was also assessed during the PT schemes. More specifically, the May 2020 PT scheme focused on positive and negative agreement, while the November 2020 test scheme added a component for the detection of other respiratory pathogens of significance, and finally the June 2021 test scheme built upon the last by including relevant SARS-CoV-2 variants of concern (Table 4). Negative agreement for Sample B was 100% across all platforms. The November 2020 test scheme consisted of two samples, neither of which contained SARS-CoV-2: instead, Sample H contained a moderate dose of influenza A virus (Ct 27) and Sample J contained the negative nasal secretion/UTM matrix only. Sample J had 100% negative agreement across all platforms; however, Sample H demonstrated some inconsistencies when the Cepheid Xpert Xpress SARS-CoV-2 platform was employed. In six instances, according to the manufacturer's instructions for reporting, the N target incorrectly identified the presence of SARS-CoV-2 in a sample that only contained influenza A virus (Table 2). In each circumstance, the Ct values were >40 and suggested that there was some degree of cross reactivity

with influenza A virus, as this was never observed with any of the negative samples. Since, all discordant results were above the 40 Ct value, recommendations were made to investigate modifying the Ct cut-off to 40 instead of 45, as recommended by the manufacturer to avoid reporting false positives (17). Over the course of the three PT schemes, the Cepheid Xpert Xpress SARS-CoV-2 platform had a 100% negative agreement for the E target and a 97% negative agreement for the N target. Negative agreement for Sample O and Q were 100% across all platforms.

All commercial and laboratory developed tests were successfully able to detect the variants of concern. Of note, the ThermoFisher TaqPath COVID-19 Combo Kit had a drop off in one of its three target genes; the S gene was not able to detect the B.1.1.7 variant, while the other two target genes were successfully identified. According to the manufacturer's recommendations for reporting, a positive result requires n=2/3 targets to have Ct values less than 37; therefore, the loss of the S gene did not impair the assays ability to detect the presence of SARS-CoV-2 in Sample N (14). Failure of the BioFire Film Array RP2.1 to detect SARS-CoV-2 P.1 was attributed to a technical error and not an assay failure; therefore, this test was not included in the analysis. The BioFire Film Array RP2.1 successfully detected the P.1 variant in all other attempts (n=48).

Overall, test specificity was comparable across all three PT schemes and platforms; a 99.5% negative agreement was observed.

Conclusion

Over the course of three PT schemes conducted across Canada between May 2020 and June 2021, the average score obtained by participants was 99.3%, demonstrating consistent testing between laboratories and testing platforms. Similarly high levels of agreement have been observed internationally. The American Proficiency Institute conducted a study across the United States and reported an overall score greater than 97% (3). Similarly, the Royal College of Pathologists of Australasia conducted three PT schemes within Australia and New Zealand between March 2020 and November 2020, with an initial score of 75% concordance early in the pandemic but then dramatically increasing to 95% concordance in the two latter test schemes (2). Finally, a third program from South Korea demonstrated 93% agreement (1). While each program varied in its sample composition and intended uses, it is encouraging to see that rapid deployment of SARS-CoV-2 testing resulted in consistently high degrees of agreement across the globe.

The ability to support quality assurance of testing measures through the provision of an external PT Program is essential during a novel or emerging public health threat. CLRN provides a framework to support the quality assurance required for the decentralization and increase in testing capacity within Canada. All Canadian public health laboratories follow a

quality management program required by their respective jurisdictions, and on-site verification and validation schemes are essential to achieve these processes. Furthermore, the comparison of PT panel results allows for the assessment of various NAAT platforms at different locations across multiple users providing an overall assessment of platform performance. The cumulative performance of the NAAT employed during the three CLRN SARS-CoV-2 PT schemes was 99.3% concordant. A future consideration would be to collect additional data from participants to gain a greater scope of demographics, population statistics and accreditation status. This study demonstrates the rapid and successful implementation of a Canadian PT Program and provided comparative analysis of the various emergency use authorized and laboratory developed tests employed for the detection of SARS-CoV-2.

Authors' statement

CR — Conceptualization, data analysis, writing–original draft, writing–review
KA — Conceptualization, writing–review
CC — Conceptualization, writing–review

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Innovative technology and established partnerships—a recipe for rapid adaptability under emerging pandemic conditions

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Abstract

Background: Aided by a collaborative partnership dating back to 2011, the Canadian Network for Public Health Intelligence (CNPHI) and the Canadian Paediatric Surveillance Program (CPSP) quickly undertook substantial enhancements to the CPSP's data collection instruments on the CNPHI platform to characterize the impacts of the coronavirus disease 2019 (COVID-19) on children and youth in Canada. Faced with an emerging public health threat with impacts yet unknown, the objective of the intervention was to rapidly complete enhancements to existing data collection and analytical tools to enable the CPSP's ability to characterize the impacts of COVID-19 in Canadian children and youth.

Intervention: Reporting frequency from CPSP's network of paediatric practitioners was increased from monthly to weekly, and the flexibility of detailed case data collection was substantially enhanced using complex survey instruments, interactively designed using CNPHI's Web Data technology. To ensure their data collection proceeded along all required lines of surveillance, CPSP's data collection tools were enhanced to collect demographic, epidemiological, microbiological and clinical data including comorbidities of cases identified.

Outcomes: Less than a month after the World Health Organization declared the COVID-19 pandemic, CPSP was able to start collecting detailed weekly case data on emerging cases of COVID-19 among Canadian children and youth. By May 2020, CPSP was able to launch a detailed study, supporting research into potential risk factors for severe COVID-19-related illness in children and youth.

Conclusion: In response to a novel public health threat, CNPHI and CPSP were able to implement rapid adaptations and enhancements to existing data collection instruments while fortifying their preparedness to do the same in the future, when needed. With innovative and agile technologies at the ready, this experience helps to emphasize the importance of established collaborative partnerships across public health disciplines as a factor contributing to preparedness and agility to respond to the unforeseen. Canadian Network for Public Health Intelligence's Web Data technology showed agile adaptability and a capacity for complex and detailed data collection, supporting timely surveillance and response.

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Introduction

Emerging and re-emerging infectious disease threats continue to test our preparedness to respond in a timely and effective manner to protect public health. Researchers and public health professionals require adaptable tools that yield intelligence to advance their understanding of known threats while providing the agility to pivot in response to the unforeseen. Experiences

over the past two decades, such as the emergence of severe acute respiratory syndrome (SARS) in 2003 or pandemic H1N1 influenza in 2009, have prompted an increased focus on defining the elements that contribute to enhanced preparedness. One key element of preparedness is the establishment and fostering of ongoing collaborative partnerships across public health disciplines (1).

One such partnership exists between the Canadian Network for Public Health Intelligence (CNPHI) and the Canadian Paediatric Surveillance Program (CPSP). Since 2011, CNPHI and CPSP have worked together to establish and enhance data collection and analysis through a secure and easy-to-use environment that started with the design, development and launch of the e-CPSP system on CNPHI. Data collection enhancements made at that time resulted in the modernization and improved timeliness of CPSP's data collection activities by enabling the transition from a paper-based system to an electronic system for the collection of data on rare conditions and diseases from paediatric practitioners across Canada (2). Although a small proportion of practitioners preferred to keep using the paper-based means of reporting, these enhancements resulted in increased agility and adaptability to support the rapid collection of surveillance information on emerging issues such as the Zika virus, for example (3).

This article discusses agile adaptations made to CPSP's data collection tools on the CNPHI platform in early 2020 in response to the arrival of the coronavirus disease 2019 (COVID-19), enabling enhanced surveillance on the impacts of COVID-19 on children and youth across Canada. As a novel public health threat, very little was known about the epidemiology of COVID-19, particularly its impacts on children and youth. Within one month of COVID-19 being declared a pandemic by the World Health Organization (4), the level of detail of CPSP's data collection was substantially increased and data collection frequency was increased from monthly to weekly. Subsequently, when healthcare providers were starting to see patients with a new condition called paediatric inflammatory multisystem syndrome/multisystem inflammatory syndrome in children (PIMS/MIS-C), the flexibility of CPSP's data collection tools allowed the rapid adaptability needed to collect detailed information on this novel syndrome. Both the flexibility and volume of data collection were substantially increased using CNPHI's innovative Web Data technologies to support the creation of lengthy and complex survey instruments, in English and French, distributed via the e-CPSP system on CNPHI. Web Data enabled the data collection and extraction and the analysis of information collected on a weekly cycle. Flexibility also allowed for participants to report on cases seen in prior weeks that had not yet been reported. These rapid adaptations enabled CPSP to launch three studies in one to characterize risk factors for severe illness within hospitalized cases of acute COVID-19 in children and youth, non-hospitalized cases with acute COVID-19 and chronic comorbid conditions, as well as PIMS/MIS-C, temporally

associated with COVID-19. Such adaptations have enhanced the preparedness and ability of CPSP to respond quickly to emerging health concerns both presently and in the future, including subsequent waves of COVID-19 (5).

The Canadian Paediatric Surveillance Program

Established in 1996 as a joint program between the Public Health Agency of Canada and the Canadian Paediatric Society, CPSP actively collects data from approximately 2,800 paediatricians and paediatric subspecialists across Canada, representing a paediatric population of over seven million Canadian children and youth. This allows CPSP to play an important role in supporting and coordinating national public health surveillance, research as well as raising awareness of childhood disorders that highly impact disability, morbidity and economic costs to society, despite their low frequency.

Importantly, this also allows CPSP to participate as an International Paediatric Surveillance Unit, engaging in international knowledge exchange across four continents through the International Network of Paediatric Surveillance Units. International collaboration is essential in the study of rare and ultra-rare conditions, and is key to understanding and tracking novel public health threats, as was demonstrated when congenital Zika syndrome emerged, and most recently during the COVID-19 pandemic.

Historically, some important examples of CPSP's contributions include the following:

- Capturing serious adverse events related to recreational cannabis use in children and youth, following the legalization of cannabis in Canada in late 2018. The study revealed that significant harm is caused by unintentional exposures in young children through the ingestion of edibles. These findings highlight the urgent need to keep these products out of the hands of our youngest citizens and informed the Canadian Paediatric Society's response to the legislative review of the Cannabis Act (6).
- Collecting data to demonstrate that although seat belts are proven to save lives, if worn incorrectly, they can lead to seat belt syndrome and cause significant injuries, including permanent paralysis (7).
- Capturing data on Vitamin D rickets—a condition that, although entirely preventable, continues to be a global health problem among children, even in developed countries such as Canada (8).
- Capturing data that led to the creation of national clinical guidelines for paediatricians and other child health providers on the management of severe hyperbilirubinemia, stimulating practice change that ultimately improved outcomes for children and youth across Canada (9).

The Canadian Network for Public Health Intelligence

Established in 2004 following lessons learned after the SARS pandemic, CNPHI is a secure, web-based scientific public health informatics and biosurveillance platform currently serving a large number of users from a diverse array of public health disciplines within federal, provincial and territorial agencies across Canada. A wide array of specialized applications and tools support surveillance and analytical requirements, data exchange and research, alerting and intelligence generation. In addition, CNPHI Collaboration Centre technologies support coordination, collaboration and knowledge exchange among various national groups in support of decision-making, research and program implementation. Canadian Network for Public Health Intelligence's Web Data technology is available within the suite of Collaboration Centre tools on the CNPHI platform and has proven to be an agile and flexible technology supporting rapid data collection needs. It offers an intuitive interface that allows public health users to create data fields of various types to support data collection through surveys, questionnaires or the creation of *ad hoc* databases to meet unique public health program needs. Web Data can also be applied to address more complex data collection requirements, with the support of the CNPHI team.

As a core tenet of its organizational philosophy, CNPHI, within the National Microbiology Laboratory Branch at the Public Health Agency of Canada, recognizes the fundamental importance of the collaborative partnerships formed with public health professionals across a wide array of programs and disciplines. All applications and related enhancements are developed in close collaboration with public health program experts, ensuring that the public health informatics solutions developed meet their evolving needs. Partnerships are sustained and nurtured over time, allowing for an ongoing understanding of a program's vision. In turn, this detailed familiarity directly supports preparedness and the readiness to adapt to changing needs related to emerging or re-emerging public health threats.

Adaptability of Web Data technologies in various settings

Canadian Network for Public Health Intelligence's Web Data has continued to evolve, benefitting from the advancement of technologies and experience gained in supporting public health stakeholders across various disciplines for a number of years and under various conditions. Web Data is a technology available on the CNPHI platform, designed to alleviate challenges related to agile data collection during outbreaks. It provides a mechanism for non-technical users to rapidly deploy a secure, web-based system for managing data and undertaking subsequent analyses and reporting. Developed in 2008 by CNPHI, Web Data was first put to the test, and its effectiveness evaluated, in partnership with public health authorities in 2009 in response to the

H1N1 influenza pandemic, during which significant benefits and capabilities were realized (10).

Also, in the context of the 2009 H1N1 pandemic, Web Data showed rapid adaptability for data collection and analysis in response to the detection of the H1N1 influenza virus in swine. In this instance, CNPHI worked in partnership with the Canadian Animal Health Surveillance Network (CAHSN) of the Canadian Food Inspection Agency. Canadian Network for Public Health Intelligence and CAHSN undertook rapid adaptations and quickly commenced data collection from federal, provincial and university animal health laboratories across Canada, at a time when the impacts of H1N1 influenza (swine flu), as an animal health threat, and its interspecies transmissibility were not yet known (11).

Working with researchers and child welfare institutions, CNPHI also applied Web Data to assist in exploring the feasibility and benefits of abstracting added surveillance value from child welfare administrative data by coding categories of child maltreatment. Web Data questionnaires were used to analyze and assess the reliability and level of agreement with which individuals coded specific categories of child maltreatment. Results showed that coding of information from child welfare files had good potential for adding value to broader surveillance of child maltreatment to support research, policy development and decision-making (12).

As a means of leveraging enhanced public health surveillance and response capabilities from mobile devices and field sensors, CNPHI worked in partnership with Health Canada, First Nations and Inuit Health Branch (Alberta Region) and Sunnybrook Research Institute on a project called "CNPHI on the Go". This initiative successfully applied Web Data technologies to enable data collection and analysis as well as two-way communication between the mobile environment and the CNPHI platform (13), a capability with numerous applications of benefit.

With the arrival of the COVID-19 pandemic in Canada, there was an urgent requirement to rapidly adapt the existing data collection tools used by CPSP in order to implement surveillance of emerging cases of COVID-19 among children and youth seen by paediatric practitioners.

Intervention

The existing e-CPSP system on CNPHI, in place prior to the emergence of the COVID-19 pandemic, offered two types of data collection instruments. These included Type 1 Surveys, designed for singular monthly responses for data collection related to established studies undertaken by CPSP researchers, and Type 2 Surveys, designed for singular responses to *ad hoc* requests for information not linked to any specific studies. With the emerging COVID-19 pandemic in early 2020, CPSP had a pressing need to modify its data collection tools in order to increase both the frequency and flexibility of data collection.

Building on their established working relationship, CNPHI and CPSP worked together to initiate enhancements to the CPSP's data collection system on CNPHI, leveraging CNPHI's agile Web Data technology.

At the outset of the intervention, very little was known about the potential impacts of COVID-19-related illness among children and youth. Given the unknown nature of the emerging public health threat, CPSP needed to focus on collecting broad information of surveillance value at a much greater frequency. Recognizing the need to commence data collection without delay, CNPHI and CPSP worked together to establish an innovative process to initiate rapid data collection in full anticipation of the need for flexibility to accommodate unknown challenges and adaptations.

Using the built-in interactive survey designer within Web Data technology, a Type 3 Survey was developed within a few days, allowing multiple responses from participants to capture weekly reporting of emerging COVID-19 cases. Type 3 Surveys were significantly longer and more complex—designed to enable the concurrent collection of more detailed information, such as demographic, epidemiological, microbiological and clinical data including comorbidities of cases identified. Despite their length and complexity, Type 3 Surveys needed to retain the flexibility to accommodate survey content enhancements to adapt to details yet unforeseen at the outset of the pandemic, such as PIMS/MIS-C.

With data collection underway, CNPHI undertook rapid enhancements to the Web Data application to accommodate the length of the Type 3 Surveys (hundreds of questions), non-responses, as well as multiple responses from individual participants. The dynamic and interactive form builder supported versatile data collection by offering a wide variety of response fields ranging from pick lists, date fields, free text and drop-down menus, for example. This versatility was fundamentally important to the collection of required information of surveillance value, yielding data fields to support queries and analysis of survey responses.

Web Data also supported ongoing adaptability to accommodate adjustments arising due to changing case definitions or feedback received. With the capability to produce the surveys in English and French, adaptability also accommodated the need to adjust translated terms to maintain accuracy, consistency and clarity throughout the process.

Importantly, Web Data is intuitive and user-friendly. With the shared experience and lessons learned from the rapid implementation of Type 3 Surveys, CNPHI and CPSP worked towards the creation of Type 4 Surveys. Building on the expertise and confidence gained by CPSP researchers and staff, Type 4 Surveys yielded an in-house capability to design, deploy and manage data collection specific to a targeted study or cases

reported. This has further increased CPSP's preparedness and agility to adapt to emerging issues and implement rapid studies while providing a detailed data collection instrument to advance research and intelligence generation. The Type 4 Survey is currently being used by CPSP to capture data on an unintended consequence of the pandemic response: a worrisome and rapid rise in first-time hospitalizations of patients with anorexia nervosa since the arrival of COVID-19 (14). Below, **Table 1** provides a summary of the attributes and objectives of the four types of surveys and **Figure 1** depicts the enhancements made to the e-CPSP system on CNPHI in response to COVID-19. Type 4 Surveys now support every new study undertaken by CPSP, with all data collection taking place online.

Table 1: Summary of enhancements to Canadian Paediatric Surveillance Program data collection instruments in response to COVID-19

Survey type	Participant responses	Frequency	Objectives
CPSP data collection instruments prior to COVID-19			
Type 1	Singular	Monthly	Data collection for established research studies
Type 2	Singular	*Ad hoc*	Data collection not linked to any established research studies
CPSP data collection instrument enhancements in response to COVID-19			
Type 3	Multiple	Weekly	Lengthy, detailed data collection on COVID-19 cases, adaptable to changing case definitions, feedback received and changing terminology in English and French
Type 4	Per case	Per case	To establish an in-house capability and preparedness to design, deploy and manage detailed data collection per case reported within a given study

Abbreviations: COVID-19, coronavirus disease 2019; CPSP, Canadian Paediatric Surveillance Program

Figure 1: A depiction of the enhancements to the Canadian Paediatric Surveillance Program's web-based system on CNPHI in response to COVID-19
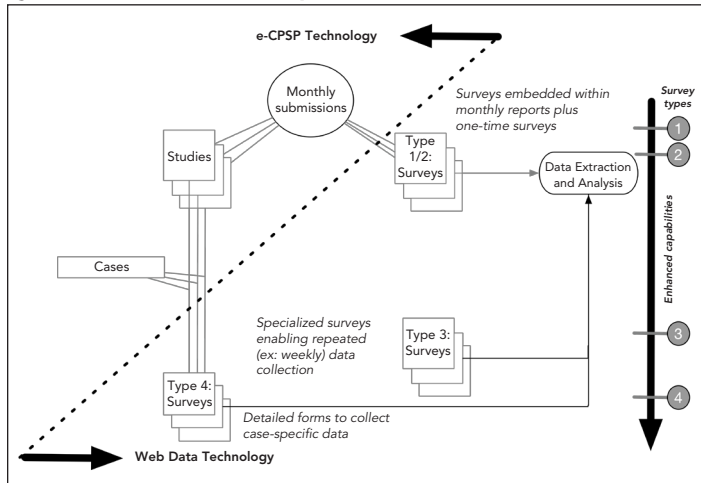
Abbreviation: CPSP, Canadian Paediatric Surveillance Program

Outcomes

The creation and deployment of the Type 3 Surveys occurred in rapid fashion, enabling CPSP to initiate detailed COVID-19 case surveillance by April 8, 2020, less than one month after the World Health Organization declared the COVID-19 pandemic. As the pandemic unfolded, numerous adaptations and modifications to the Type 3 Surveys were successfully completed to keep pace with changing needs. By May 2020, with the arrival of PIMS/MIS-C, which was a new condition temporally associated with COVID-19, CPSP researchers were able to quickly pivot and adapt the survey tool to capture critical information on this new condition.

Importantly, the adaptations made to CPSP's system on CNPHI have also established an ongoing capability for CPSP to quickly respond to changing surveillance needs. Study investigators now have access to the online data, which is far more comprehensive than data collected via the hard copy forms. Also, in response to cases reported, CPSP can now respond with an online questionnaire to the reporting physician either the same day or the following day, a vast improvement in timeliness compared to the mailed questionnaire that could take a week to reach the reporting physician. This overall effort became a staging ground for Type 4 Surveys, yielding the preparedness and confidence for CPSP to advance towards autonomous management of the data collection and analytical tools offered by Web Data technologies on the CNPHI platform and the readiness to apply them in the future when the need arises.

Web Data provided an agile environment to support rapid survey development as a means of detailed data collection. The interactive form builder offered sufficient data field flexibility to support the complexity and length of the surveys while

maintaining an ongoing ability to accommodate changes and enhancements over time, in English and French. Data extraction and analytical capabilities within Web Data enabled optimal surveillance value to be gleaned from the data collected.

Discussion

The long-standing CNPHI-CPSP partnership has provided an environment of familiarity that comes with a history of working together. A shared knowledge of CPSP's existing surveillance tools and strategies allowed the partners to immediately focus on the adaptations needed to implement a timely surveillance response in the face of a novel public health threat. This, together with the ready availability of the agile Web Data technology, allowed for the successful achievement of the objective of enabling CPSP to adapt and respond in a timely fashion to characterize the impacts of COVID-19 on Canadian children and youth.

Important non-technical aspects contributed to the agility of this response. Logic may dictate that when one is faced with an emerging public health threat, it is not the right time to start getting to know the role and function of your key partners. This sentiment is clearly reflected in discussions that seek to define the elements of preparedness and resilience. As a program dedicated to excellence in providing scientific public health informatics solutions across a wide array of public health disciplines, CNPHI places a fundamental importance on collaborative partnerships by placing them at the core of its philosophy. With Type 3 Surveys deployed and data collection underway, the pandemic was still in its first month. To prepare for the unknown road ahead, the partners built on their experience to establish an in-house capacity for CPSP to design, deploy and manage Type 4 Surveys using Web Data, enhancing their ongoing preparedness and agility to respond to the unforeseen.

Limitations

A diminishing proportion of CPSP's community of participants has not yet made the transition to electronic reporting through CPSP's system on CNPHI. As a result, a paper-based system is still maintained for some participants, which requires extra time and effort for CPSP in terms of data collection, extraction and analysis.

However, as an indirect benefit, with mailing methods also impacted by pandemic-related restrictions and office closures, many remaining paper-based participants elected to make the transition to electronic reporting. There are now fewer than 100, out of approximately 2,800 participants still using the paper-based reporting system—a substantial decline compared to before the pandemic.

With electronic data collection through Web Data, quality assurance steps were required to identify and address duplicate responses in some instances. Furthermore, although CPSP has reached a comfort level for managing and deploying Type 4 Surveys using Web Data, researchers and members of the medical community will still need to achieve consensus on survey and study design with respect to emerging issues of concern. Finally, the creation of detailed surveys in more than one language introduces a need for vigilance to ensure that the accuracy and consistency of translated terms are maintained, particularly in the context of an emerging infectious disease with impacts yet unknown, as terminology and case definitions may vary.

Conclusion

With the arrival of a novel public health threat with impacts yet unknown, CNPHI and CPSP successfully leveraged their long-standing partnership to complete rapid and complex enhancements to existing data collection instruments, enabling CPSP to successfully adapt and begin characterizing the impacts of COVID-19 on Canadian children and youth, less than a month after the World Health Organization declared the pandemic. The strategic importance of this established, long-standing partnership cannot be overstated, as a key element contributing the timeliness and agility of this response.

The Canadian Network for Public Health Intelligence's innovative Web Data technology proved to be agile, adaptable and robust, enabling the rapid creation of lengthy and complex surveys for data collection and the subsequent extraction and analysis of data collected.

Innovative technologies and established partnerships were shown to be important components of preparedness in the face of a novel public health threat.

Authors' statement

SM, ML, and TB collaborated on the conceptualization, drafting and revision of this paper.

Competing interests

None.

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Relative pandemic severity in Canada and four peer nations during the SARS-CoV-2 pandemic

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Abstract

Background: National responses to the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) pandemic have been highly variable. We sought to explore the effectiveness of the Canadian pandemic response up to May 2022 relative to responses in four peer countries with similar political, economic and health systems, and with close historical and cultural ties to Canada.

Methods: We used reported age-specific mortality data to generate estimates of pandemic mortality standardized to the Canadian population. Age-specific case fatality, hospitalization, and intensive care admission probabilities for the Canadian province of Ontario were applied to estimated deaths, to calculate hospitalizations and intensive care admissions averted by the Canadian response. Health impacts were valued in both monetary terms, and in terms of lost quality-adjusted life years.

Results: We estimated that the Canadian pandemic response averted 94,492, 64,306 and 13,641 deaths relative to the responses of the United States, United Kingdom and France, respectively, and more than 480,000 hospitalizations relative to the United States. The United States pandemic response, if applied to Canada, would have resulted in more than $40 billion in economic losses due to healthcare expenditures and lost quality-adjusted life years. In contrast, an Australian pandemic response applied to Canada would have averted over 28,000 additional deaths and averted nearly $9 billion in costs.

Conclusion: Canada outperformed several peer countries that aimed for mitigation rather than elimination of SARS-CoV-2 in the first two years of the pandemic, with substantial numbers of lives saved and economic costs averted. However, a comparison with Australia demonstrated that an elimination focus would have saved Canada tens of thousands of lives as well as substantial economic costs.

Introduction

The global severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) pandemic has taken a fearsome toll on mortality, life expectancy and population health globally, but not all countries have been impacted equally. The reasons for this heterogeneity are only partly understood. Population age structure is a key contributor to SARS-CoV-2 severity (1,2); however, countries with older age distributions (such as Japan) have been less severely affected than its high-income peers (3). Japan's early focus on the airborne nature of SARS-CoV-2, and

the widespread acceptance of masking, may also have been important mitigators (3,4). Marked heterogeneity in severity was seen across countries that have similar age structures but were slow to recognize airborne transmission of SARS-CoV-2.

A case in point is the differential severity of the pandemic in Canada and the United States (US); both are wealthy, federal democracies with advanced medical care systems. In both countries, the coronavirus disease 2019 (COVID-19) pandemic

has had a major impact on population health and the economy. The similarities and differences between the two countries' healthcare systems have made cross-national comparisons an important source of insight into the strengths and weaknesses of their respective health systems (5). During the COVID-19 pandemic, both COVID-19 cases and deaths per capita have been substantially higher in the US than in Canada (6). Australia represents another reasonable peer for Canada for comparison purposes. Australia is similar to Canada in terms of income, culture and governance, but employed more stringent pandemic control measure and consequently had much lower per capita SARS-CoV-2 pandemic mortality as of May 2022 (7). The United Kingdom (UK) and France share ties of economy, culture and history with Canada (as hubs of the British Commonwealth and La Francophonie, both of which include Canada), and may also represent appropriate comparators.

Debate in the Canadian public sphere around pandemic policy has often focussed on whether Canada's approach to disease control should have been more or less stringent. Assuming that differences in outcomes were at least partly driven by policy rather than the independent actions and choices of individuals, we sought to explore the differences in outcomes that Canada would have experienced over the first two years of the SARS-CoV-2 pandemic had it followed the path of the US, the UK, France or Australia. We had previously performed such an analysis in March 2021, with comparison restricted to Canada and the US (6). While our objective was not to perform a formal cost-utility analysis of the Canadian pandemic response relative to responses in these peer nations, the question of costs averted, or excess costs accrued, both through hospitalizations and premature loss of life, is an important one, and we incorporated simple valuations of these quantities into our analysis. These may help inform future cost-utility analyses on this question.

Methods

We obtained national COVID-19-attributed death estimates from the Public Health Agency of Canada, and national health authorities for the US, the UK, France and Australia until late April or early May of 2022, as available (7–11). We chose these countries as comparator peers because all are high income countries with advanced health systems, and all have strong cultural, political, and historical links to, and similarities with Canada. Of these five countries, all but Australia (12,13) sought to mitigate rather than eliminate SARS-CoV-2 during the first two years of the pandemic. Some Canadian provinces and territories, notably Atlantic provinces and Northern Territories, (14) did pursue elimination at times. Population estimates were obtained from national census agencies for all countries (15–19). We calculated the number of excess or deficit deaths that would have been expected in Canada under approaches employed in peer countries using direct standardization (20). Because country death data were reported using slightly different age groupings,

we reallocated Canadian deaths to mirror the distribution of SARS-CoV-2 deaths, by two-year age increments, due to data availability in the province of Ontario (available to January 18, 2022). Deaths were assumed to be equally distributed between years in each two-year category. Standardized mortality ratios (SMR) for Canada, relative to other countries, were estimated by dividing observed by expected deaths (i.e. the deaths that would have occurred with a US, UK, France or Australia-equivalent response). The 95% confidence limits for SMR were calculated by estimating standard errors as $(1/A+1/B)^{1/2}$, where A and B are death counts in each of the two peer countries, as described previously (20).

Observed deaths were subtracted from expected deaths to calculate deaths averted. We divided averted deaths by age-specific case-fatality estimates from Ontario to estimate averted cases. We applied age-specific risks of hospital admission and intensive care admission, derived from Ontario case data, to calculate hospital and intensive care admissions averted. We placed a monetary value on hospitalizations and intensive care unit (ICU) admissions averted based on Canadian cost estimates generated by the Canadian Institute for Health Information (21). The approach of Briggs *et al.*, modified for the Canadian context by Kirwin *et al.*, was used to estimate quality-adjusted life years (QALY) lost for deaths occurring in each age group (22,23). We monetized QALY losses averted by applying a net expected benefit approach, with QALY valued at $30,000 as per Kirwin *et al.* (23). We compared the stringency of pandemic responses using the Oxford Government Coronavirus Response Tracker's Pandemic Stringency Index (24). The stringency was plotted against time and differences in the stringency between Canada and other countries were evaluated with the Wilcoxon rank-sum test. All input data are publicly available.

Results

Fewer SARS-CoV-2-related deaths per capita had occurred in Canada than in the US in all age groups as of May 2022, with SMR significantly less than one for all age groups in Canada. A similar pattern was seen when Canada was compared to the UK, except in children aged 0–14 years, where there was no significant difference between the two countries (SMR 1.02, 95% CI: 0.67–1.55). In comparison with France, Canada experienced significantly fewer deaths per capita in adults aged 40–89 years, more deaths than France in those aged 20–29 years and 90 years and older, and no difference in those younger than 20 years. In comparison with Australia, Canada had significantly higher SARS-CoV-2-related deaths per capita in all age groups except those aged 10–19 years, where differences were not significant (SMR 2.24, 95% CI: 0.81–6.16) (**Table 1**).

Table 1: Standardized mortality ratios for the first two years of the SARS-CoV-2 pandemic in peer countries compared to Canada

Age group (years)	Deaths	Population	Cumulative mortality per 1,000	Expected deaths, Canadian population	Observed Canadian deaths[a]	Standardized mortality ratio	95% CI
United States							
0–17	1,045	73,284,400	0.01	103.42	37	0.35	0.25–0.49
18–29	6,257	52,870,600	0.12	700.11	136	0.19	0.16–0.23
30–39	18,148	43,375,000	0.42	2,244.47	315	0.14	0.13–0.16
40–49	42,961	39,929,000	1.08	5,265.77	660	0.13	0.12–0.14
50–64	187,272	62,110,000	3.02	23,329.55	3,772	0.16	0.16–0.17
65–74	229,682	31,487,000	7.29	29,816.49	6,422	0.22	0.21–0.22
75–84	257,553	15,407,000	16.72	35,486.56	10,899	0.31	0.30–0.31
85 and over	255,780	5,893,000	43.40	37,823.67	18,038	0.48	0.47–0.48
Total	991,396	324,356,000	-	134,770	40,278	-	-
United Kingdom							
0–14	64	11,974,857	0.005	32	33	1.02	0.67–1.55
15–44	2,748	25,311,086	0.109	1,631	685	0.42	0.39–0.46
45–64	21,139	17,286,653	1.223	12,378	4,466	0.36	0.35–0.37
65–74	30,745	6,719,287	4.576	18,703	6,491	0.35	0.34–0.36
75–84	59,945	4,129,982	14.515	30,812	21,317	0.69	0.68–0.70
85 and over	78,125	1,659,369	47.081	41,028	7,286	0.18	0.17–0.18
Total	192,766	67,081,234	-	104,584	40,278	-	-
France							
0–9	37	7,706,041	0.005	19	29	1.54	0.95–2.50
10–19	31	8,421,914	0.004	15	15	0.98	0.53–1.82
20–29	147	7,525,983	0.020	99	128	1.29	1.02–1.63
30–39	465	8,279,577	0.056	301	315	1.05	0.91–1.21
40–49	1,337	8,572,713	0.156	763	660	0.87	0.79–0.95
50–59	4,576	8,813,899	0.519	2,664	1,862	0.70	0.66–0.74
60–69	13,344	8,000,803	1.668	8,074	4,349	0.54	0.52–0.56
70–79	26,358	5,959,261	4.423	13,862	8,633	0.62	0.61–0.64
80–89	43,387	3,214,055	13.499	18,460	13,844	0.75	0.74–0.76
90 and over	25,895	927,995	27.904	9,662	10,443	1.08	1.06–1.11
Total	115,577	67,422,241	-	53,919	40,278	-	-
Australia							
0–9	8	3,156,780	0.003	10	29	2.91	1.33–6.37
10–19	5	3,097,360	0.002	7	15	2.24	0.81–6.16
20–29	22	3,476,779	0.006	32	128	3.97	2.53–6.24
30–39	65	3,780,122	0.017	92	315	3.41	2.61–4.46
40–49	124	3,294,734	0.038	184	660	3.58	2.96–4.34
50–59	322	3,143,647	0.102	526	1,862	3.54	3.15–3.99
60–69	726	2,737,883	0.265	1,284	4,349	3.39	3.13–3.66
70–79	1,579	1,952,572	0.809	2,534	8,633	3.41	3.23–3.59
80–89	2,695	876,320	3.075	4,205	13,844	3.29	3.16–3.43
90 and over	1,925	221,945	8.673	3,003	10,443	3.48	3.31–3.65
Total	7,471	25,738,142	-	11,878	40,278	-	-

Abbreviations: CI, confidence intervals; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; -, not applicable
[a] Due to redistribution of deaths to comparator peer country age categories, fractional deaths were calculated; all deaths rounded to the nearest whole number

When compared to the US, UK and France's SARS-CoV-2 responses, we estimated that Canada's response prevented 94,492 (95% CI: 93,593–95,360), 64,306 (95% CI: 63,394–65,189) and 13,641 (95% CI: 12,489–14,735) deaths, respectively. In contrast, an Australian response applied to Canada would have saved 28,400 (95% CI: 26,097–30,939) lives of the total number of Canadians (n=40,278) that had been lost to SARS-CoV-2 as of May 2022 (**Table 2**).

Distributions of deaths by age differed markedly between the US and the other countries analyzed. For example, half of deaths in the US occurred in individuals under the age of 55 years; in other countries, half of the fatalities occurred in those under approximately 75 years of age with the remainder occurring in those 75 years of age and over (**Figure 1**). A similar divergence between the US response and those in other countries was seen when we applied age-specific QALY losses to death data (**Figure 2**).

We estimated that Canada's response saved over one million QALYs, nearly 500,000 hospitalizations and over 100,000 ICU admissions relative to what would have occurred with a response equivalent to that seen in the US (Table 2). The value of QALY losses and hospitalizations averted is estimated to be approximately $43 billion, with $32 billion due to aversion of lost QALY and the remainder due to averted hospitalizations. The Canadian response also saved QALY and averted hospitalizations and ICU admissions relative to UK and French responses. When compared to the Australian response, Canada's response was estimated to have resulted in approximately 230,000 additional QALY lost, over 80,000 excess hospital admissions and over 15,000 excess ICU admissions as of May 2022, representing a loss of $8.78 ($7.21 to $10.77) billion (Table 2). Age-specific estimates of deaths, healthcare utilization and costs averted for each of the four peer comparator countries are presented in Table 2.

The stringency of the Canadian pandemic response from March 1, 2020, to May 1, 2022, was significantly higher than the stringency in the US, the UK and France, and was also higher than the Australian stringency ($p<0.001$ for all comparisons) (**Appendix**, **Table A1** and **Figure A1**).

Table 2: Health outcomes and costs[a] averted in peer countries compared to Canada

| Outcome	Comparator peer country							
	United States	95% CI	United Kingdom	95% CI	France	95% CI	Australia[b]	95% CI
Deaths averted	94,492	93,593–95,360	64,306	63,394–65,189	13,641	12,489–14,735	−28,400	−30,939–−26,097
Hospitalizations averted	483,009	465,046–516,497	196,611	184,256–209,756	39,367	26,213–50,528	−83,281	−110,498–−67,197
ICU admissions averted	108,157	99,635–117,714	40,131	37,002–43,514	8,984	6,873–10,683	−15,335	−20,059–−12,380
QALY gained	1,060,180	943,164–1,172,874	569,981	514,483–635,306	133,517	107,018–158,498	−231,100	−277,758–−191,373
Hospitalization costs averted	10.73	10.32–11.47	4.37	4.09–4.66	0.87	0.59–1.13	−1.85	−2.42–−1.49
ICU costs averted	5.18	4.78–5.65	1.92	1.77–2.08	0.43	0.33–0.51	−0.73	−0.95–−0.59
Hospitalization costs averted (non-ICU)	5.55	5.55–5.81	2.45	2.31–2.58	0.44	0.25–0.62	−1.12	−1.46–−0.90
Net benefit of QALY gained	31.81	28.29–35.19	17.10	15.43–19.06	4.01	3.26–4.74	−6.93	−8.00–−5.50
Total costs averted	42.54	38.62–46.65	21.47	19.52–23.71	4.88	3.83–5.88	−8.78	−10.77–−7.21

Abbreviations: CI, credible intervals derived via simulation; ICU, intensive care unit; QALY, quality-adjusted life year
[a] All costs are in billions of $CDN
[b] Negative values denote excess health consequences and costs in Canada relative to Australia

Figure 1: Cumulative proportion of COVID-19-attributable death by age[a], March 2020 to May 2022
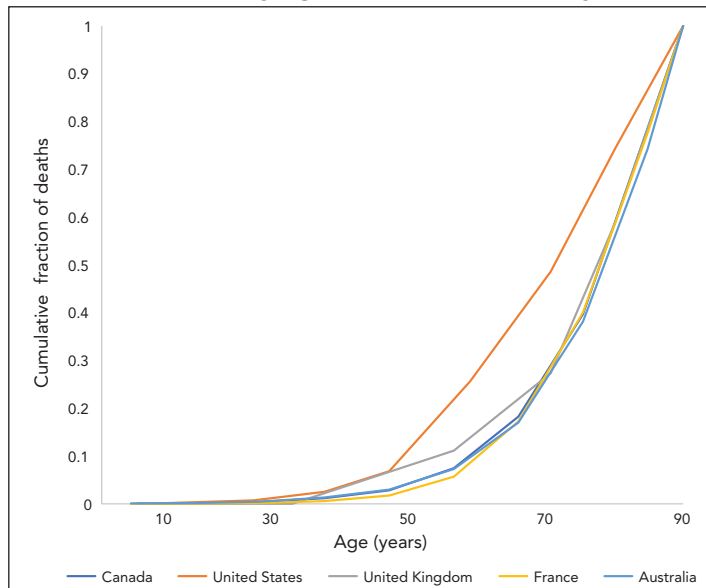
Abbreviation: COVID-19, coronavirus disease 2019
[a] Ages represent the midpoints of age categories. For the oldest age categories in Canada (80 years of age and over) and the United States (85 years of age and over) we assigned an age of 90 years

Figure 2: Cumulative proportion of COVID-19-attributable quality-adjusted life years loss, by age[a], March 2020 to May 2022
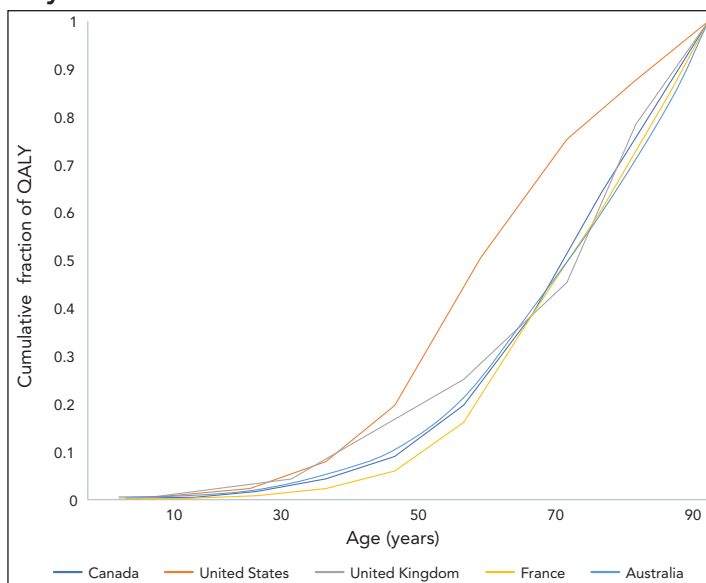
Abbreviations: COVID-19, coronavirus disease 2019; QALY, quality-adjusted life years
[a] Ages represent the midpoints of age categories. For the oldest age categories in Canada (80 years of age and over) and the United States and United Kingdom (85 years of age and over) we assigned an age of 90 years; for Australia and France, the highest age category (90 years of age and over) was assigned a value of 90 years

Discussion

The cultural similarities and integrated economies of Canada and the US, which also have very different health systems, has long encouraged comparative research between these two countries

(5,25–27). During the current SARS-CoV-2 pandemic, this type of research has continued, spurred, in part, by the remarkable difference in the pandemic's impact on the two countries (28). Here, we demonstrate that application of age-specific US data to Canada resulted in a far deadlier pandemic in the US, with a more than three-fold higher total deaths relative to those that had occurred in Canada as of May 2022. A challenge with this type of comparison is that the US's pandemic response has emerged as a global outlier, with SARS-CoV-2 taking a far greater toll in terms of loss of life than in any other high-income peer country. The outlier status of the US (28) has the effect of making Canada-US comparisons predictable in result, perhaps unfairly elevating the effectiveness of the Canadian pandemic response. As such, we also evaluated Canada's response relative to the UK, France and Australia, which given cultural, political, economic and historical similarities to Canada, are also fair comparators.

We find that, as with the US, application of the UK's pandemic response to Canada would have resulted in tens of thousands of additional deaths, as well as billions of dollars in excess economic losses. While Canada appears to have outperformed France as well, differences in pandemic repercussions between these two countries were more modest. In contrast, Australia emerges as a model of what Canada might have achieved by taking a more aggressive stance on disease control during the first two years of the SARS-CoV-2 pandemic. Indeed, we estimate that over 75% of Canadian pandemic deaths to date could have been averted through an Australian response, with cost savings of approximately $10 billion.

Our work complements that of Razak et al., who also found that Canada had outperformed most of its G10 peers (except for Japan) with respect to pandemic-attributable mortality (29). However, the use of standardization, as applied here, allows us to see that the Canadian approach was far more effective than the US and UK approaches in preventing deaths in younger adults, with consequently greater gains in quality-adjusted survival. As public health and government officials in these five countries likely had access to similar information for decision-making, differences in outcomes likely reflected active policy choices. The complexity of the pandemic, and societal responses to it, make identification of causal factors challenging. Galvani et al. noted that a key difference between Canada and the US may relate to universal public healthcare in the former (28); however, universal public healthcare is also available in the UK, France and Australia. Razak et al. noted that Canada outperformed many high-income peer countries on vaccination (29). We have also suggested that cultural differences between countries, including differences in social capital and trust in government, may be important (30).

While Canada's pandemic response, as reflected in the Oxford Stringency Index, was more stringent on average than the responses in the US, the UK and France, it was also more stringent than Australia's, suggesting that stringency alone cannot explain differences in outcomes. Data from Aknin et al.

suggest that it may not have been stringency, but the decision to aim for elimination rather than mitigation, which resulted in the low stringency and low deaths seen in countries like Australia (31). Although more aggressive pandemic control strategies have been criticized over perceived negative mental health impacts, Aknin *et al.* also demonstrated that the impact of excess pandemic deaths far outweighed the impact of public health interventions as a driver of negative mental health effects during the pandemic (31). This suggests that Canada's approach, in addition to saving more lives and reducing more costs than US and UK responses, may have been more protective of population mental health. More stringent control strategies have also been criticized as resulting in greater negative economic impacts, and indeed Canada's GDP declined by 1.6% in the first two years of the pandemic (29); however, the $43 billion Canada effectively gained by avoiding a US-style pandemic response represents over 2% of Canadian GDP (valued at around $2.1 trillion $CDN).

Limitations

Our analysis has three key limitations. We have not attempted to capture consequences or costs of the pandemic on mental health. It should be noted that Aknin *et al.* (31) found that a pandemic elimination rather than mitigation stance decreased overall stringency and mental health impacts. Other important costs and impacts that we did not include, and which would likely further widen the gap in health and economic consequences between these peer countries, include disutility and lost earnings associated with hospitalization, long-term costs of chronic disease, including cardiac, respiratory and neurological disease, in those who survive SARS-CoV-2 infection, and the health, economic and societal impacts of parental loss due to the pandemic (32–35). As we have included only QALY gains and losses associated with death, and not incorporated those associated with short-term illness and hospitalization, or with the post-acute COVID syndrome (commonly referred to as "long COVID"), our estimates for QALY lost represent lower bounds for all countries (36). A second limitation of our analysis is our use of Ontario-specific case fatalities and hospitalization and intensive care admission risks to estimate outcomes averted at a national level. We use these data for pragmatic reasons: they were the most complete and granular Canadian death data to which we had access. Furthermore, Ontario's epidemiology is likely similar to that of Canada overall, both because of similarities in demographics and health systems across the country, and also because the population of Ontario represents approximately 40% of the Canadian population and 35% of Canada's COVID-19 case load, such that the province's epidemiology strongly influences that of Canada as a whole. Lastly, we assumed that attribution of COVID-19 deaths in Canada and comparator peer countries occurred in a comparable manner. The best available data (based on ratios of reported COVID-19 mortality to all-cause excess mortality during the pandemic) suggest that this is likely to have been the case for Canada, the US and France; reporting of COVID-19 mortality may have been more accurate in the UK than in Canada, which would tend to exaggerate the

differences in outcomes between these two countries. More accurate reporting of COVID-19 deaths in Australia would lead us to underestimate the degree to which this country outperformed comparator peer countries (37).

Conclusion

Canada's relatively strong pandemic response during the first two years of the SARS-CoV-2 pandemic resulted in large numbers of deaths, hospitalizations and ICU admissions averted relative to responses in the US and UK, and more modest gains relative to France. A disease control stance focussed on elimination rather than mitigation, as was pursued in Australia during the same time period, would have resulted in further health and economic benefits.

Authors' statement

AP — Data acquisition, cleaning and analysis, drafting of manuscript
AA — Conceptualization, manuscript editing and revision
AS — Conceptualization, manuscript editing and revision
AT — Conceptualization, manuscript editing and revision
DF — Project lead, conceptualization, drafting, editing and revision of manuscript

All authors approved the final version for publication.

Competing interests

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Appendix

Table A1: Mean and standard deviation for Oxford Pandemic Stringency Index in Canada and comparator peer countries, March 1, 2020 to May 1, 2022

Country	Mean	SD	p-value[a]
Canada	58.60	21.71	N/A
Australia	54.88	18.76	<0.001
France	48.84	21.08	<0.001
United Kingdom	51.14	24.05	<0.001
United States	53.12	17.98	<0.001

Abbreviations: N/A, not applicable; SD, standard deviation
[a] p-value for Wilcoxon rank-sum test for comparison with Canada

Figure A1: Oxford Pandemic Stringency Index by date, Canada and comparator peer countries[a]
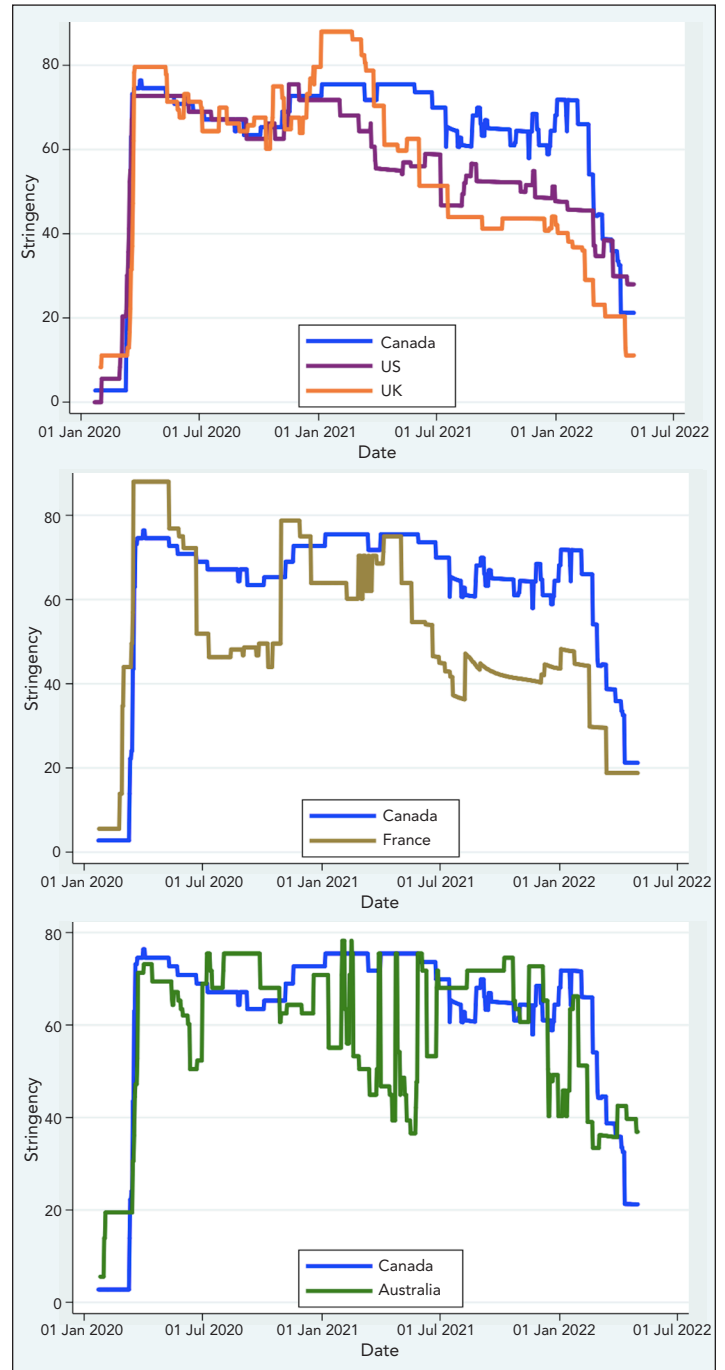
Abbreviations: UK, United Kingdom; US, United States
[a] Stringency values plotted to May 1, 2022. Higher values indicate more stringent control measures

COVID-19 farm outbreaks in Ontario, January–December 2020

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Abstract

Background: Farm workers are critical to Ontario's food supply chain as they grow and harvest the food that Ontario relies on; however, they are subject to several occupation-related coronavirus disease 2019 (COVID-19) transmission risk factors. We describe the epidemiology of farm outbreaks in Ontario over the first calendar year of the pandemic and explore trends in outbreaks by season and type of farm.

Methods: Data pertaining to farm outbreaks in Ontario from January 1 to December 31, 2020, and their associated laboratory-confirmed cases were extracted from the provincial database. Outbreaks were characterized by size, season, farm type and duration. Cases were characterized by age, gender, medical risk factors, clinical presentation and outcomes.

Results: There were 64 farm outbreaks associated with 2,202 confirmed cases of COVID-19 in Ontario during 2020. The majority of outbreaks occurred in spring (n=25, 39.1%) and fall (n=25, 39.1%). The fewest outbreaks occurred in the summer (n=6, 9.4%), corresponding with low community rates during that time, and the majority of these were in greenhouse farms (n=5, 83.3%). The median outbreak size was 14.5 cases (range: 1–240), and the median duration was 23 days (range: 0–128). Among cases, most were male (83.2%), the median age was 35 years, 10.0% had one or more comorbidities, 31.2% were asymptomatic, 16 required hospitalization and three died.

Conclusion: Farm outbreaks were a source of COVID-19 transmission and illness in 2020, particularly in the spring and fall. Outbreaks continued in greenhouse farms despite lower summer community transmission.

Introduction

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) can be spread when infectious respiratory particles are inhaled by individuals or deposited on their mucosal surfaces (1). The risk of transmission is higher as the source-to-receptor distance decreases, which is common when working and/or living with others that are infected with coronavirus disease 2019 (COVID-19) (1). During the COVID-19 pandemic, agricultural workers were deemed an essential workforce and farms remained open and in-person due to their necessary role in growing and harvesting food (2). Farm workers faced unique challenges, increasing their risk of COVID-19 infection compared to other essential workers. Farm work is often in close proximity without physical barriers. This includes work in indoor greenhouses, which account for 32% of farms in Ontario (3). Greenhouse farms differ in humidity, temperature and ventilation compared with outdoor fields, and these conditions can make greenhouses a favourable environment for viral transmission (4). In addition, temporary foreign farm workers, who make up 31% of employees on farms in Ontario, may also share transportation and living quarters, and face language barriers, lower income and decreased access to healthcare services, which make them more susceptible to occupational risks such as COVID-19 (3,5,6).

In response to the unique concerns faced by farm workers during the COVID-19 pandemic, the Ontario Ministry of Agriculture, Food and Rural Affairs first introduced the "Enhanced Agri-Food Workplace Protection Program" in May 2020 to help farms improve the health and safety of agri-food workers in Ontario during the COVID-19 pandemic (7). The Ontario Ministry of Health first developed the "COVID-19 Guidance: On-Farm Outbreak Management" in September 2020 that provides recommendations for safe practices on worksites, transportation and shared accommodations (8). Vaccine distribution for COVID-19 in Ontario began in December 2020; but at the time, vaccines were only eligible for certain populations. Farm workers were not eligible for the vaccine until phase 3 of the vaccine rollout in Ontario, around August 2021 (9).

The objective of this analysis was to describe the epidemiology of COVID-19 outbreaks in farms in Ontario in the pre-vaccine year of the pandemic, for outbreaks with a start date between January 1 and December 31, 2020, all cases associated with these outbreaks up to January 31, 2021, and trends in outbreaks by season and type of farm (i.e. indoor greenhouse vs. outdoor field).

Methods

Data source
We obtained data on COVID-19 outbreaks on farms and laboratory-confirmed COVID-19 cases linked to those outbreaks from the Public Health Case and Contact Management Solution (CCM); a dynamic disease reporting system for COVID-19 case and contact management in Ontario. We also obtained data on laboratory-confirmed COVID-19 cases in the general Ontario population. Data were entered by staff at the 34 local public health units (PHU) and digitally extracted by Public Health Ontario in February 9, 2021.

Outbreak definitions and analysis
Prior to the development of a provincial definition for farm outbreaks, outbreak declaration was at the determination of the local PHU investigating cases associated with a farm. As of September 2020, the Ontario Ministry of Health issued guidance defining a COVID-19 on-farm outbreak. A COVID-19 on-farm outbreak is defined as "one case (of COVID-19) in a congregate living area or two cases of COVID-19 (in the workplace), either asymptomatic or symptomatic, and where there is evidence of COVID-19 transmission in either the congregate living area or the workplace" (8). Outbreaks with no outbreak-associated confirmed cases were removed from the analysis (n=2). Outbreaks were included in the study if their start date was between January 1, 2020, and December 31, 2020.

Outbreak start date was determined by the episode date of the first outbreak case; if this date was unknown or missing, the outbreak reported date was used, followed by the outbreak created date. Episode date for cases is based on an estimate of the best date of disease onset and is calculated using a hierarchy based on the date of symptom onset, specimen collection/test date or the date reported to the PHU.

Outbreaks were characterized by PHU, size (i.e. number of confirmed cases linked to the outbreak by the PHU) and duration (i.e. the time from the episode date of the first case to the episode date of the last case linked to the outbreak, up to January 2021). A manual review of farm outbreak locations was conducted to classify farms with greenhouses, given their additional risk for COVID-19 as crowded, indoor environments.

Outbreaks were also further categorized by season based on the outbreak start date. Spring outbreaks were those starting between March 20 and June 19, 2020; summer outbreaks were those starting between June 20 and September 21, 2020; and fall outbreaks were those starting between September 22 and December 20, 2020. Winter was removed from the analysis as there were limited data for this season in 2020 (10).

Outbreak-associated cases
Laboratory-confirmed COVID-19 cases linked to farm outbreaks were included if their episode date was between January 1, 2020, and January 31, 2021, to include cases associated with outbreaks that were still open after December 31, 2020. Outbreaks were considered closed if they had a "declared over date" in CCM or if it had been five months since the outbreak start date. As of data extraction time, five included outbreaks remained open. Cases were characterized by age, gender, medical risk factors (including presence of one or more comorbidities and high-risk status), symptoms, outcomes and PHU where the outbreak occurred. Comorbidities included anemia, asthma, chronic obstructive pulmonary disease (COPD), cancer, cardiovascular disease, underlying medical condition, liver disease, diabetes, immunocompromised, neurological disorder, obesity, "other", pregnancy, renal disease and tuberculosis. High-risk status was defined as individuals aged 60 years and older, immunocompromised, having cardiovascular conditions or COPD. Clinical symptoms were classified as asymptomatic, symptomatic or missing. Clinical outcomes were classified as ever hospitalized, ever in the intensive care unit (ICU) or death. Clinical outcomes were listed in hierarchical order (i.e. each case is counted with the highest-level outcome only, specifically: death, ICU, then hospitalizations).

We included all cases that were linked to a farm outbreak as a "farm worker" and this may include farm owners, family members, employees on the farm and individuals who visited the farm if they were deemed to be related to the farm outbreak based on the PHU investigation.

We used chi-square tests of proportions to compare medical risk factors outcomes of farm outbreak cases to overall laboratory-confirmed COVID-19 cases in Ontario aged 20–59 years

(corresponding to approximately 95.0% of the cohort population) excluding farm-outbreak associated cases and long-term care home resident cases dated January 1 to December 31, 2020. Long-term care home cases were excluded given their differential risk and the nature of public health measures applied.

Epidemiologic analysis

Descriptive statistics were used to describe COVID-19 farm outbreaks in Ontario. Proportions were calculated for categories of outbreak-associated cases by gender, age, medical risk factors, clinical presentation, outcomes and PHU. Outbreaks and outbreak-associated cases were further subdivided by season and the mean, median and range of the number, duration, and size of outbreaks was calculated for each season. Finally, the percentage of total farm outbreaks and outbreak-associated cases in greenhouses was calculated for each season. An epidemiologic curve was used to display outbreaks among the three PHUs with the most outbreaks, along with the number of outbreak-associated cases over the period included. Descriptive statistics were also used to describe non-farm outbreak-associated cases. All analyses were conducted using SAS Enterprise Guide (version 9.4) and Microsoft Excel.

Results

There were a total of 64 farm outbreaks with 2,202 outbreak-linked cases (Table 1). Outbreaks ranged in size from one to 240 cases (median 15 cases), with 63 outbreaks (98.4%) having two or more cases and six outbreaks (9.4%) having 100 or more cases. Outbreak duration ranged from zero days (i.e. all cases as part of the outbreak had the same episode date) to 128 days (median 23 days).

Table 1: COVID-19 farm outbreaks in Ontario, January 1−December 31, 2020

Overall outbreak descriptions	Frequency	Mean	Median	Range
Total number of outbreaks	64	N/A	N/A	N/A
Total number of outbreak-associated cases	2,202	34.4	14.5	1–240
Duration of all outbreaks (days)	N/A	31.3	23	0–128

Abbreviations: COVID-19, coronavirus disease 2019; N/A, not applicable

A total of 37 (57.8%) farm outbreaks occurred on farms classified as greenhouses. The majority of farm outbreaks occurred in three PHUs (Windsor-Essex County Health Unit, Haldimand-Norfolk Health Unit and Chatham-Kent Public Health) that accounted for 68.8% (n=44/64) of all farm outbreaks in Ontario. Farm outbreaks peaked in May 2020 and again in December 2020. Farm outbreaks were infrequent from the end of June to early September 2020 (Figure 1).

Figure 1: Epidemiologic curve of COVID-19 farm outbreaks in Ontario, January 1−December 31, 2020[a]
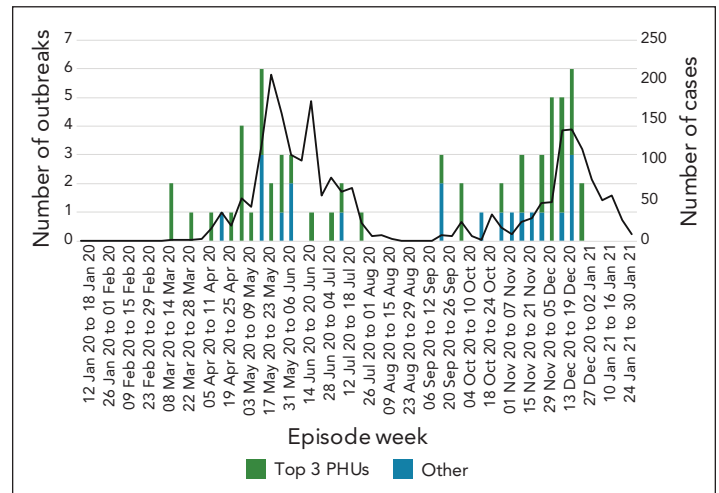
Abbreviations: COVID-19, coronavirus disease 2019; PHU, public health unit
[a] Bars correspond to the number of outbreaks reported by the top three PHUs with the most COVID-19 on-farm outbreaks reported in Ontario and the total number of COVID-19 on-farm outbreaks reported by all other PHUs combined. The black line corresponds to total number of COVID-19 on-farm cases reported across all PHUs in Ontario. Top three PHUs correspond to PHUs with most outbreaks reported in the province: Windsor-Essex county Health Unit (N=35), Haldimand-Norfolk Health Unit (N=6) and Chatham-Kent Public Health (N=4)

When comparing COVID-19 farm outbreaks by season (Table 2), the total number of outbreaks was highest in the spring and fall (25 outbreaks each); however, the total number of outbreak-associated cases was highest in the spring (n=1,292 cases, 58.7%), followed by fall (n=772 cases, 35.1%) and summer (n=69 cases, 3.1%). Duration of outbreaks was also longest for outbreaks starting in the spring, with a mean duration of 43.2 days (range: 1–128 days), followed by fall (mean duration 29.1 days [range: 0–76 days]) and summer 14.2 days (range: 4–29 days). There was a higher proportion of farm outbreaks occurring on farms classified as "greenhouses" in the summer (83.3%) and spring (68.0%), compared to fall (52.0%). The majority of outbreaks occurred in Windsor-Essex, where there is a high density of agricultural farms, regardless of the season.

Outbreak-linked cases were predominantly male (83.2%) with a median age of 35 years. There were 221 (10.0%) cases that had one or more comorbidities and 121 (5.5%) that met criteria for high-risk status. The majority were symptomatic (n=1,375; 62.4%), while 688 (31.2%) were asymptomatic and symptoms were missing in 139 (6.3%) cases. In total, there were 16 (0.7%) outbreak-associated cases that were hospitalized, eight (0.4%) cases admitted to the ICU and three (0.1%) deaths. The majority of outbreak-associated cases were associated with three PHUs with 1,498 (68.0%) from Windsor-Essex, 260 (11.8%) from Haldimand-Norfolk and 143 (6.5%) from Chatham-Kent (Table 3).

Compared with farm outbreak-associated cases, cases in the general population (n=177,092) had more comorbidities (n=29,620, 16.7%, $p<0.05$) and a higher proportion were hospitalized (n=2,733, 1.5%, $p<0.05$). The proportions of cases that were admitted to the ICU (n=651, 0.4%, $p=0.49$) or died (n=237, 0.1%, $p=0.45$) were similar.

Table 2: COVID-19 farm outbreaks in Ontario by season[a], January 1−December 31, 2020

Description of outbreaks and outbreak-associated cases		Spring (March 20–June 19)	Summer (June 20–Sept 21)	Fall (Sept 20–22–Dec 20)	Winter (Dec 20–31)[b]	Total
Total outbreaks		25	6	25	8	64
Total outbreak-associated cases	Total (N)	1,292	69	772	69	2,202
	Percent of total (%)	58.7%	3.1%	35.1%	3.1%	100%
	Mean cases per outbreak (N)	51.7	11.5	30.9	8.6	34.4
	Median cases per outbreak (N)	21	6.5	25	4	14.5
	Range of cases per outbreak (N)	2–240	3–30	3–77	1–27	1–240
Duration of all outbreaks (days)	Mean	43.2	14.2	29.1	13.8	31.3
	Median	38	14.5	24	11	23
	Range[c]	1–128	4–29	0–76	0–34	0–128
Greenhouses	Total outbreaks (N)	17	5	13	2	37
	Percent of total outbreaks (%) by season	68.0%	83.3%	52.0%	25.0%	57.8%
	Outbreak-associated cases (N)	822	63	409	46	1,340
	Percent of total cases (%) by season	63.6%	91.3%	53.0%	66.7%	60.9%

Abbreviation: COVID-19, coronavirus disease 2019
[a] There were no farm outbreaks between January 1 and March 19, 2020
[b] Data for winter is limited due to the study period ending December 31, 2020; data is included here for overall comparison, but not included in the analysis and discussion
[c] Zero days indicates that all cases as part of the outbreak had the same episode date

Table 3: Characteristics of farm outbreak-associated cases for outbreaks dated January 1−December 31, 2020

Outbreak-associated cases	Frequency	Proportion
Total	2,202	N/A
Gender		
Male	1,831	83.2
Female	332	15.1
Unknown or missing	39	1.8
Age (years)		
Younger than 10	1	0.0
10–19	23	1.0
20–29	672	30.5
30–39	740	33.6
40–49	467	21.2
50–59	204	9.3
60–69	81	3.7
70–79	11	0.5
80 and older	1	0.0
Unknown	2	0.1
Medical risk factors		
One or more comorbidities[a]	221	10.0
High-risk status[b]	121	5.5
Clinical presentation		
Asymptomatic	688	31.2

Table 3: Characteristics of farm outbreak-associated cases for outbreaks dated January 1−December 31, 2020 (continued)

Outbreak-associated cases	Frequency	Proportion
Clinical presentation (cont.)		
Symptomatic	1,375	62.4
Missing symptoms	139	6.3
Outcomes[c]		
Death	3	0.1
ICU	8	0.4
Hospitalized	16	0.7
Public health unit where outbreak occurred[d]		
Chatham-Kent Public Health	143	6.5
Haldimand-Norfolk Health Unit	260	11.8
Halton Region Public Health	82	3.7
Middlesex-London Health Unit	31	1.4
Niagara Region Public Health	83	3.8
Region of Waterloo Public Health and Emergency Services	18	0.8
Simcoe Muskoka District Health Unit	24	1.1
Southwestern Public Health	44	2.0
Windsor-Essex County Health Unit	1,498	68.0

Abbreviations: COPD, chronic obstructive pulmonary disease; ICU, intensive care unit; N/A, not applicable
[a] Includes: anemia, asthma, COPD, cancer, cardiovascular disease, underlying medical condition, liver disease, diabetes, immunocompromised, neurological disorder, obesity, "other", pregnant, renal disease, tuberculosis
[b] Includes: Age 60 years and older, immunocompromised, cardiovascular condition, COPD
[c] Listed in hierarchical order (i.e. each case is counted with the highest-level outcome only)
[d] Three public health units with fewer than 15 outbreak-associated cases were not presented in the table

Discussion

Farm outbreaks of COVID-19 in Ontario occurred throughout most of 2020 with increased activity in the spring and fall, and were associated with 2,202 cases, 16 hospitalizations and three deaths. Farm outbreaks peaked in May 2020 and December 2020 corresponding to the increased rates of COVID-19 cases in the province overall (11). The spring peak occurred after the March 2020 implementation of travel restrictions and stay-at-home orders (12), and prior to the implementation of provincial farm outbreak guidance and other public health measures in Ontario issued in September 2020. During the summer months, when COVID-19 transmission was low in the province, there were fewer outbreaks overall and the majority of outbreaks occurred in greenhouses. The indoor and crowded nature of greenhouse work, at a time when indoor masking was not routinely recommended or used, may have promoted the transmission of COVID-19 and could have contributed to outbreaks on farms even when there were lower levels of community transmission. The relative role of indoor, crowded daytime working conditions of greenhouses compared to other risks of transmission on farms, such as congregate living among workers, warrants further investigation.

In a previous study of workplace outbreaks and outbreak-associated cases in Ontario, the agricultural sector had among the highest incidence rates of COVID-19 per hours worked compared to other labour force sectors (13). Additionally, for the time period of April 1 to August 31, 2020, the agricultural sector had the second-highest proportion of outbreak-associated cases and cases that were hospitalized compared to other industry sectors (13). This analysis specifically focuses in on characteristics of farm-related outbreaks and cases to describe their unique characteristics and explores factors that may have contributed to the over-representation of the agricultural sector for outbreaks, specifically greenhouse farms, and their potential role in contributing to farm outbreaks. A previous study of workplace outbreaks in Ontario has also shown that individuals associated with workplace outbreaks are younger, healthier and have lower rates of severe outcomes compared to the general population (14). In comparison to previously published workplace (all industries) outbreak-associated cases, farm outbreak-associated cases were younger, had fewer comorbidities, and had a lower proportion of hospitalizations and deaths. However, in this analysis, compared to the general population of cases of the same age, farm outbreak cases had similar proportions of ICU and death outcomes, despite a lower proportion with comorbidities. This suggests that there were differential risks for the most severe outcomes for farm outbreaks compared with other workplace outbreaks.

A number of previous studies cite challenges for farm workers which may contribute to higher rates of COVID-19 outbreaks on farms. In New York state, it was noted that farm workers did not have adequate access to personal protective equipment until COVID-19 infections were at an "alarmingly high rate" (15). Fear of job loss and deportation, lack of income replacement programs while isolating or sick, language and cultural barriers and having long and irregular hours are believed to contribute to farm workers avoiding testing or treatment (5,16). This is of particular importance in Ontario, as 31% of farm workers are also temporary foreign workers, with limited access to resources (3). It has also been noted that there are deficiencies in housing standards in many jurisdictions in Ontario, including windows that cannot open (limiting ventilation), inadequate laundry facilities (for cleaning work clothing) and high occupancy (limiting physical distancing in sleeping quarters and other shared facilities) which may contribute to spreading of COVID-19 among farmworkers (17).

Limitations

The epidemiologic analysis of this study is subject to limitations. Firstly, only data entered into CCM were available for analysis. The number of cases of COVID-19 in CCM was subject to varying degrees of underreporting as not all individuals with COVID-19 developed symptoms, sought medical attention or testing and, therefore, the disease may have been unreported. Therefore, the number of outbreak-associated cases for each outbreak was likely an underestimate. As well, four outbreaks were classified as "open" as of data extraction and the data for these outbreaks is potentially subject to change. Misclassification of greenhouse status is possible as it was manually coded. Additionally, data in CCM does not specify where on the farm the outbreak occurred and cases may be unrelated to the greenhouse setting. This can make it difficult to draw definite conclusions about greenhouse farms. Other potential factors associated with outbreaks, such as local quarantine procedures, number of foreign workers, number of people living in shared housing, were not available for analysis.

Conclusion

With the introduction of COVID-19 vaccines and workplace infection prevention and control measures over the course of the pandemic, the risk of large and long farm outbreaks has significantly reduced. However, given the relaxation of public health measures, including indoor masking, the return of international travel and the ongoing risk of the emergence of a new and more transmissible variant of concern, farms may continue to be settings vulnerable to COVID-19 outbreaks. Future studies are needed to understand the role of greenhouse work and other factors that may contribute to farm outbreaks of COVID-19.

OUTBREAK REPORT

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Real-time quantitative reverse transcription polymerase chain reaction detection of SARS-CoV-2 Delta variant in Canadian wastewater

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Abstract

Background: Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) variants of concern are associated with increased infectivity, severity, and mortality of coronavirus disease 2019 (COVID-19) and have been increasingly detected in clinical and wastewater surveillance in Canada and internationally. In this study, we present a real-time quantitative reverse transcription polymerase chain reaction (RT-qPCR) assay for detection of the N gene D377Y mutation associated with the SARS-CoV-2 Delta variant in wastewater.

Methods: Wastewater samples (n=980) were collected from six cities and 17 rural communities across Canada from July to November 2021 and screened for the D377Y mutation.

Results: The Delta variant was detected in all major Canadian cities and northern remote regions, and half of the southern rural communities. The sensitivity and specificity of this assay were sufficient for detection and quantitation of the Delta variant in wastewater to aid in rapid population-level screening and surveillance.

Conclusion: This study demonstrates a novel cost-effective RT-qPCR assay for tracking the spread of the SARS-CoV-2 Delta variant. This rapid assay can be easily integrated into current wastewater surveillance programs to aid in population-level variant tracking.

Introduction

The severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) pandemic began in Wuhan, China in late 2019 before becoming a worldwide pandemic in 2020. Beginning in September 2020, variants of concern (VOC) began to emerge which had mutations leading to increased viral transmission rates, increased virulence, or the ability to escape existing vaccines (1–4). On May 11, 2021, the World Health Organization declared the Delta variant (B.1.617.2) to be a VOC (5). The Delta variant has been shown to be both more transmissible and more virulent than the wild-type (WT) (Wuhan) strain (6–8).

Wastewater-based epidemiology (WBE) has proven to be a powerful tool for tracking the spread of SARS-CoV-2 on a population level, and recently has become instrumental in monitoring the dissemination of VOC across Canada and throughout the world (9–12). The real-time quantitative reverse transcription polymerase chain reaction (RT-qPCR)-based assays have previously been developed to identify mutations associated with emerging VOC including Alpha (Sdel69-70, ND3L), Beta (Sdel241, N501Y) and Gamma (N501Y) (13–15). Early detection of VOC can potentially lead to improved public health responses, such as increased sequencing of clinical isolates, enhanced surveillance and enhanced public health measures. Monitoring relative amounts of SARS-CoV-2 variants over time can be useful for monitoring trends in viral transmission and potentially assessing the effectiveness of public health interventions (16).

In this report, we describe a novel RT-qPCR assay for detection of the N gene D377Y allele—associated with the Delta variant—in wastewater. We applied this assay to wastewater samples collected from 36 sampling locations drawn from 23 Canadian cities and towns, both remote and urban, to monitor the dissemination of the Delta variant throughout the country. This population-level surveillance approach could be instrumental for monitoring changes in VOC prevalence and effects of public health interventions for reduction of viral spread within health regions.

Methods

Sample collection and nucleic acid extraction
Wastewater was collected between June 30 and December 1, 2021, from 16 urban wastewater treatment plants (WWTP) from six cities along with 20 WWTP and lift stations from 17 towns and rural locations in Canada. Fifteen of the WWTP from five cities were sampled as part of Statistics Canada's Canadian Wastewater Survey (17). A 24-hour composite sample was collected three times per week at each treatment facility and shipped to the National Microbiology Laboratory at 4°C. The samples were stored at 4°C for up to 24 hours until processed.

A 300 ml sample of primary post-grit influent or raw wastewater was mixed by inversion, then a 30 mL aliquot was drawn and processed as previously described (14). RNA was extracted using the MagNA Pure 96 DNA and Viral NA Large Volume Kit (Roche Diagnostics, Laval, Québec) using the Plasma External Lysis 4.0 protocol as per manufacturer instructions.

Delta variant of concern assay design
An assay was developed to detect the D377Y mutation consisting of a G->T in the N gene (G29406T) due to relative rarity in the general Canadian population of SARS-CoV-2 genomes, and relative exclusivity within the Delta variant genome (*personal communication, G. Van Domselaar*). This assay was designed to detect both WT and variant (V) sequences for each allele, allowing for discrimination between V and WT SARS-CoV-2 RNA.

The WT SARS-CoV-2 (NC_045512.1) sequence, along with Delta variant sequences (EPI_ISL_1372093, EPI_ISL_2134533, EPI_ISL_2134644, EPI_ISL_2134933, EPI_ISL_2135087), were obtained from Global Initiative on Sharing Avian Influenza Data (18) and used for primer and probe design. Oligonucleotide primers and probes were chosen for each target region using Primer Express Software v3.0 (Thermo Fisher Scientific, Waltham, Massachusetts) and Primer3 v4.1.0 (19). Linear dsDNA oligonucleotide gene fragments (Integrated DNA, Coralville, Iowa) consisting of the gene region flanking the variant region for either the WT or variant sequence (**Table A1**) were employed as standards and quantified using a One-Step RT-ddPCR Advanced Kit for Probes (Bio-Rad, Mississauga, Ontario) on a QX200 Droplet Digital PCR System (Bio-Rad).

Real-time quantitative reverse transcription polymerase chain reaction assay conditions
RT-qPCR was performed for D377Y WT and V assays, along with the United States Centers for Disease Control and Prevention N1 and N2 assays and interpreted as previously described (14,20) with concentrations of 500 nM of each primer (D377Y_F: CATTCCCACCAACAGAGCCT, D377Y_R: TGTCTCTGCGGTAAGGCTTG) and 500 nM of each probe (D377Y_WT: AGAAGGCTGATGAAA, D377Y_V: AGAAGGCTTATGAAAC). Each real-time PCR was performed in duplicate or triplicate as indicated with the appropriate non-template controls and positive controls.

Determination of limit of detection
The assay limit of detection (LOD) was assessed as the lowest concentration at which there was >95% test positivity in 15 replicates of a 1.5-fold serial dilution series from 45 copies/reaction (cp/rxn) to 1.8 cp/rxn of dsDNA oligonucleotide standards.

Data analyses
Amplification efficiencies (E) were calculated using $E = -1 + 10^{(-1/slope)} \times 100$. Data analyses were performed using R version 4.1.1 on Rstudio using the tidyverse packages (21).

Results

The assay limits of detection were 4 cp/rxn (WT) and 3 cp/rxn (V) when measured as a pure specimen without interfering alleles. These LODs were near the theoretical limit of RT-qPCR and sufficient for sensitive detection in wastewater, where SARS-CoV-2 RNA concentrations can be very low. Standard curves were as follows: WT (slope=−3.45, intercept=38.52, R^2=0.999); and V (slope=−3.29, intercept=38.25, R^2=0.999). The amplification efficiencies of the WT and V reactions were 101% and 95%, respectively.

As relative amounts of WT and V template within wastewater samples may vary considerably, standard curves were also created for each assay in the presence of 100, 500 and 1,000 cp/µL of the alternate allele to assess their stability (**Figure 1**). Presence of the WT template had a limited effect on V template detection with the variant assay, with a loss of signal at 1 cp/µL V only in the presence of 1,000 cp/µL WT; a concentration much higher than is likely to be detected in wastewater. Presence of V template led to decreased sensitivity and increased range of error of the WT assay, with detection at 10 cp/µL WT, but not 1 cp/µL WT in the presence of any concentration of V template. Standard deviations of both assays were determined using the alternate alleles in this experiment to assess variance over a range of concentrations. Standard deviations were averaged across three concentrations (100 cp/µL, 500 cp/µL and 1,000 cp/µL) and were 0.38 Ct (WT) and 0.31 (V) compared with 0.09–0.18 for previously published assays (14).

Figure 1: Standard curves for D377Y assays in the presence of the alternate genotype for each allele[a]
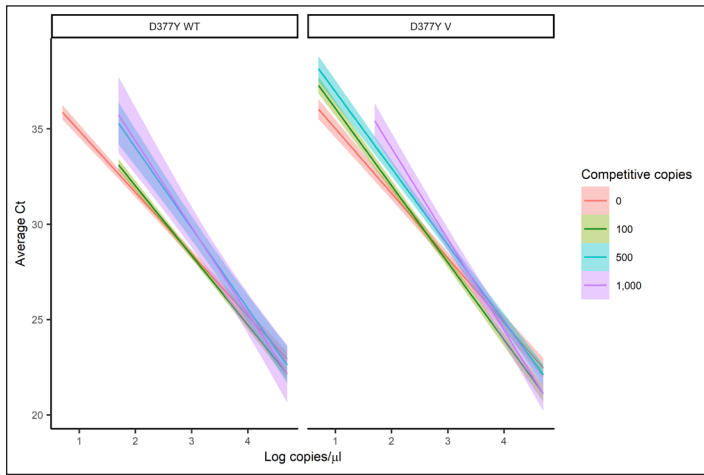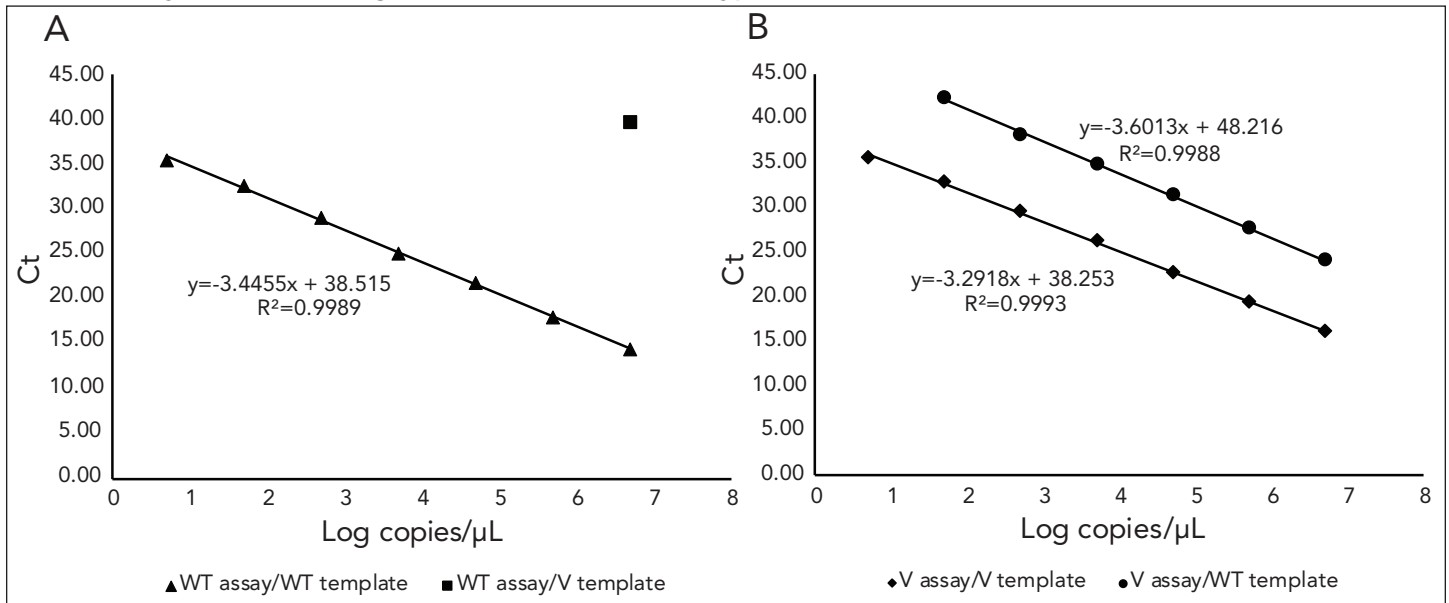

Abbreviations: Ct, cycle threshold; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; V, variant; WT, wild-type
[a] Standard curves for real-time quantitative reverse transcription polymerase chain reaction SARS-CoV-2 wild-type and D377Y variant B.1.617 assays against tenfold dilutions of DNA oligonucleotide controls in the presence of 100, 500 and 1,000 copies/μL of the alternate genotype for each allele

To test analytical specificity, both WT and V targets were tested in triplicate against serial dilutions from 10^6 cp/μL to 10^0 cp/μL of the alternate allele oligonucleotide. The WT assay showed negligible cross-reactivity, with a 25 Ct delayed detection in the presence of 1 x 10^6 cp/μL WT. The V assay showed cross reactivity with the WT template; however, the amplification was delayed by ~8 Ct (**Figure 2**).

A total of 980 samples from 36 urban and remote WWTPs and lift stations across Canada were sampled from June 30 to December 1, 2021 (**Table A2**). Of these, 539 (55%) tested positive for the SARS-CoV-2 Delta variant D377Y mutation, 210 (21.4%) tested positive for N1/N2 only and 232 (23.6%) samples were negative for both N1/N2 and D377Y. Additionally, there were 8 (0.8%) samples in which SARS-CoV-2 was detected by the D377Y assays but not N1/N2, of which seven had D377Y detection in only one of two replicates and six had detection <10 cp/mL. The Delta variant was detected in all six major cities, with initial detection ranging from July 11 to August 30 in the larger cities and October 7 in NL11 (St. John's) (**Figure 3**). Delta variant signal was initially detected in the majority of cities between July 17 and 22. The peak signal (highest concentration of Delta detected) in the cities throughout the study period ranged from July 22 to October 18, averaging 32 days after initial detection (range: 0–66, IQR: 10–49). Following initial detection in five of the six cities, Delta signal rapidly increased, becoming roughly equivalent to the SARS-CoV-2 N1 + N2 signal throughout the remainder of the study period. This sharp increase is indicative of rapid displacement of other circulating variants by Delta, as seen in clinical cases by genomic surveillance of SARS-CoV-2 variants. In St. John's, Newfoundland and Labrador, Delta signal was detected only twice during the study period and SARS-CoV-2 signal remained low; a pattern typically seen in the more remote locations in this study.

Figure 2: Standard curves[a,b,c] for the D377Y real-time polymerase chain reaction assays performed with serial dilutions of synthetic DNA oligonucleotides for the wild-type and D377Y variant alleles

Abbreviations: Ct, cycle threshold; V, variant; WT, wild-type
[a] For all standard curves, equations for the lines and R^2 values are indicated
[b] (A) Standard curve for the WT assay using WT template and cross-reactivity with the V template
[c] (B) Standard curve for the V assay using V template (solid line) and cross-reactivity with the WT template (dashed line)

Figure 3: Detection of SARS-CoV-2 Delta variant in wastewater from Canadian cities and rural areas using real-time quantitative reverse transcription polymerase chain reaction[a,b]
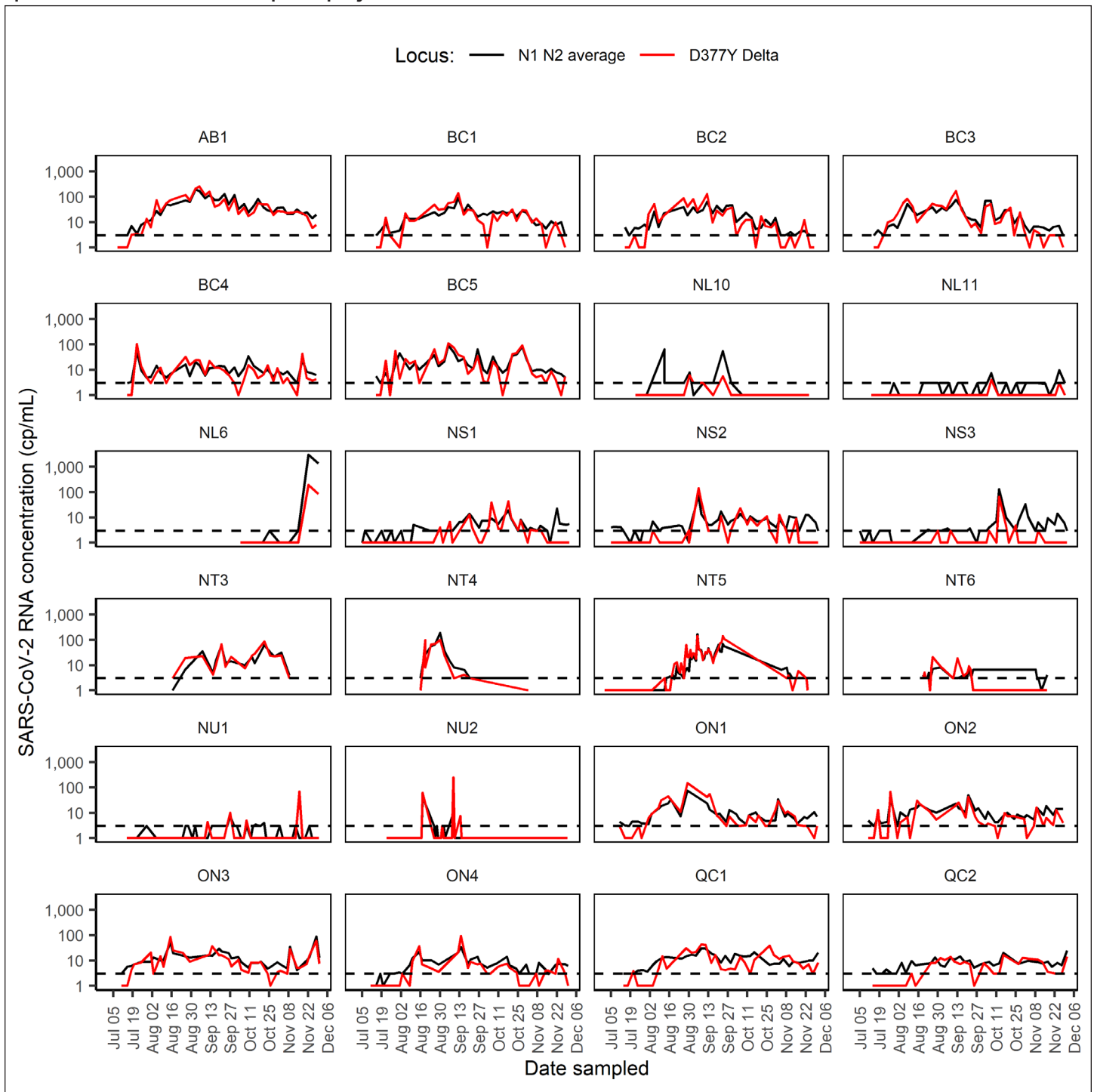
Abbreviations: AB, Alberta; BC, British Columbia; NL, Newfoundland and Labrador; NS, Nova Scotia; NT, Northwest Territories; NU, Nunavut; ON, Ontario; QC, Québec; RNA, ribonucleic acid; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2
[a] Red line depicts copies/ml (cp/mL) of D377Y mutation indicative of Delta variant presence
[b] Black line is the SARS-CoV-2 concentration using the average of the Centers for Disease Control and Prevention N1 and N2 cp/mL. Sites tested over a period of less than one month or with fewer than three samples in which SARS-CoV-2 was detected by N1 or N2 are not shown. Dashed line represents the limit of quantification of the assay. Sites are described in Table A2

The more sparsely populated regions investigated in this study showed less consistent detection of D377Y, with detection in 50% of remote sites in Newfoundland and Labrador (n=6/12) during at least one time point throughout the sampling period.

In Newfoundland and Labrador, D377Y was not detected in 4/6 sites prior to the last week of October 2021, whereas the remaining two sites had detection in July and September.

In the northern Territorial regions, Delta signal was detected in all six Northwest Territories (NWT) sites and both Nunavut (NU) sites during at least one time point. The NU1 variant had sporadic low-level detection in September, while D377Y was detected in NU2 in August to mid-September, peaking with a strong signal on September 8. The NT1 variant was sampled only in November, with high levels of detection throughout the month. Delta signal was first detected between August 12–19 in four of the remaining five NWT sites, and on September 20 for the final site. Four NWT sites (NT1,NT4–NT6) were not sampled during the month of October.

Discussion

This study describes the development of RT-qPCR assays to detect the N gene D377Y mutation associated with the SARS-CoV-2 Delta variant. The LODs of this assay were near the theoretical limit of RT-qPCR and sufficient for sensitive detection in wastewater, where SARS-CoV-2 RNA concentrations can be very low. The robustness and sensitivity of the V component of the assay allows for trending analysis, and where appropriate, early warning detection in communities and for monitoring the decline of the Delta wave.

The D377Y V assay is valuable for tracking the spread of the Delta variant in wastewater as it was used to monitor the spread of the Delta variant in eight major cities and 26 towns and rural locations across Canada over a four-month period. The Delta variant was detected in wastewater from all major Canadian cities, with a rapid increase in signal shortly following onset of detection, indicating rapid spread of Delta and displacement of other variants. Delta signal was also observed in approximately half of rural locations in Southern Canada, and all locations in Northern Canada. These data demonstrate the utility of this assay for tracking the spread of the SARS-CoV-2 Delta variant. These one-step RT-qPCR assays can be easily integrated into currently used wastewater surveillance programs to aid in SARS-CoV-2 surveillance.

Limitations
Assay limitations include the loss in sensitivity in the presence of the V allele, which limits the interpretation of the WT component of the assay during the onset of a Delta wave, where high levels of variant genomic material will attenuate the WT signal. This is consistent with previous studies that found a similar level of cross-reactivity between variant and WT assays (13,22). In wastewater samples, this delayed cross-reactivity is negligible, as the SARS-CoV-2 concentration is very low. Other limitations of this assay include: 1) inconsistent detection when RNA concentration in samples approaches the LOD of the assay or 2) the presence of inhibitors found in wastewater. Limitations of wastewater-based surveillance include testing being limited to populations present within the wastewater catchment area, variations in viral shedding between SARS-CoV-2 variants and

infected individuals, and variations in wastewater composition due to weather or industrial events.

The Delta variant of SARS-CoV-2 is defined by 27 mutations, which are commonly detected by whole genome sequencing (3,23–25). While detection of one mutation such as D377Y is not determinative of Delta variant presence, it is highly indicative as the N gene D377Y mutation is found very rarely in non-Delta strains (26).

Conclusion
Surveillance using RT-qPCR is a rapid and cost-effective method of screening for SARS-CoV-2 variants in both wastewater and clinical specimens. These assays provide a complement to SARS-CoV-2 variant detection assays as previously described (14) for surveillance of SARS-CoV-2 variants in wastewater. Wastewater-based surveillance is a valuable tool for tracking the spread of SARS-CoV-2 variants on a population level in regions where clinical testing is limited. The relative fraction of the Delta variant measured in wastewater using the assay developed in this work was communicated to public health decision-makers by weekly reporting across a network of surveillance sites throughout Canada. To our knowledge, these data were used as a complimentary public health intelligence stream and not directly actioned. Thus, over the course of the pandemic, this was principally the use for wastewater surveillance data amongst infectious control and public health leadership; likely because of a gap in trust arising from a lack of precedent and unfamiliarity with the data, in addition to the public scrutiny and pressure associated with pandemic. We hope that this work and the work of others will establish a base of use cases that will improve the action-ability of wastewater surveillance. A conservative use case could be to maintain wide-scale infectious control measures based on wastewater surveillance data. While more than a year has passed since the Delta wave, the relevance of the assay described here remains as sub-lineages of the Delta VOC has been observed in wild populations of white-tailed deer (27,28) and this work could contribute to the monitoring of the expanding host range of this virus. With the high number of asymptomatic COVID-19 cases and limited testing capacity worldwide, augmentation of surveillance capabilities by monitoring spread of SARS-CoV-2 variants in wastewater can aid in public health efforts.

Authors' statement

SWP — Conceptualization, methodology, investigation, validation, writing, visualization
JD — Methodology, investigation, validation
CD — Methodology, investigation, validation
AN — Conceptualization, resources, project administration
MRM — Conceptualization, supervision, project administration
CSM — Conceptualization, writing, supervision, funding acquisition, project administration

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Appendix

Table A1: Positive control gBlock sequences for the SARS-CoV-2 Delta variant real-time quantitative reverse transcription polymerase chain reaction assays

Region	Allele	Sequence
N D377Y	WT	AAAGATCCAAATTTCAAAGATCAAGTCATTTTGCTGAATAAGCATATTGACGCATACAAAACATTCCCACCAACAGAGC-CTAAAAAGGACAAAAAGAAGAAGGCTGATGAAACTCAAGCCTTACCGCAGAGACAGAAGAAACAGCAAACTGT-GACTCTTCTTCCTGCTGCAGATTTGGATGATTTCTCCAAACAATTGCAA
	Variant	AAAGATCCAAATTTCAAAGATCAAGTCATTTTGCTGAATAAGCATATTGACGCATACAAAACATTCCCACCAACAGAG-CCTAAAAAGGACAAAAAGAAGAAGGCTTATGAAACTCAAGCCTTACCGCAGAGACAGAAGAAACAGCAAACTGT-GACTCTTCTTCCTGCTGCAGATTTGGATGATTTCTCCAAACAATTGCAA

Abbreviations: SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; WT, wild-type

Table A2: Location and number of samples tested from wastewater treatment plants and lift stations across Canada

Site code	Region	Sampling date range	Date of first detection	Date of peak signal	Number of samples
AB1	Edmonton, AB	2021-07-08 to 2021-11-28	2021-07-18	2021-09-05	40
BC1	Vancouver, BC	2021-07-15 to 2021-11-28	2021-07-22	2021-09-12	37
BC2	Vancouver, BC	2021-07-15 to 2021-11-28	2021-07-22	2021-09-12	37
BC3	Vancouver, BC	2021-07-15 to 2021-11-28	2021-07-22	2021-09-12	38
BC4	Vancouver, BC	2021-07-15 to 2021-11-28	2021-07-22	2021-07-22	37
BC5	Vancouver, BC	2021-07-15 to 2021-11-28	2021-07-22	2021-09-05	38
NL1	Newfoundland and Labrador	2021-10-04 to 2021-11-29	Not detected	Not detected	9
NL2	Newfoundland and Labrador	2021-10-04 to 2021-11-29	2021-11-29	2021-11-29	9
NL3	Newfoundland and Labrador	2021-10-04 to 2021-11-29	2021-11-16	2021-11-16	8
NL4	Newfoundland and Labrador	2021-07-19 to 2021-11-24	Not detected	Not detected	14
NL5	Newfoundland and Labrador	2021-10-06 to 2021-11-17	Not detected	Not detected	6
NL6	Newfoundland and Labrador	2021-10-04 to 2021-11-29	2021-11-22	2021-11-22	9
NL7	Newfoundland and Labrador	2021-07-14 to 2021-11-24	2021-07-21	2021-07-21	20
NL8	Newfoundland and Labrador	2021-09-16 to 2021-11-29	Not detected	Not detected	8
NL9	Newfoundland and Labrador	2021-10-14 to 2021-11-30	Not detected	Not detected	7
NL10	Newfoundland and Labrador	2021-07-22 to 2021-11-24	2021-08-30	2021-08-30	20
NL11	Newfoundland and Labrador	2021-07-13 to 2021-11-29	2021-09-02	2021-10-07	39
NL12	Newfoundland and Labrador	2021-10-04 to 2021-11-30	2021-10-25	2021-10-25	9
NL13	Newfoundland and Labrador	2021-10-14 to 2021-11-30	Not detected	Not detected	7
NS1	Halifax, Nova Scotia	2021-07-05 to 2021-12-01	2021-08-23	2021-10-18	42
NS2	Halifax, Nova Scotia	2021-07-05 to 2021-12-01	2021-08-04	2021-09-06	42
NS3	Halifax, Nova Scotia	2021-07-05 to 2021-12-01	2021-08-30	2021-10-13	42
NT1	Northwest Territories	2021-11-03 to 2021-11-24	2021-11-04	2021-11-10	15
NT2	Northwest Territories	2021-10-19 to 2021-11-17	2021-09-20	2021-09-20	11
NT3	Northwest Territories	2021-08-16 to 2021-11-08	2021-08-16	2021-09-20	16
NT4	Northwest Territories	2021-08-16 to 2021-11-01	2021-08-19	2021-08-30	12
NT5	Northwest Territories	2021-06-30 to 2021-11-23	2021-08-12	2021-09-23	53
NT6	Northwest Territories	2021-08-19 to 2021-11-16	2021-08-19	2021-08-26	23
NU1	Nunavut	2021-07-14 to 2021-11-29	2021-09-10	2021-11-15	54
NU2	Nunavut	2021-07-22 to 2021-11-29	2021-08-17	2021-09-08	56
ON1	Toronto, ON	2021-07-11 to 2021-11-30	2021-07-11	2021-08-29	38
ON2	Toronto, ON	2021-07-11 to 2021-11-28	2021-07-18	2021-07-27	37
ON3	Toronto, ON	2021-07-11 to 2021-11-30	2021-07-18	2021-08-15	37
ON4	Toronto, ON	2021-07-11 to 2021-11-30	2021-08-03	2021-09-14	38
QC1	Montréal, QC	2021-07-14 to 2021-12-01	2021-07-21	2021-09-08	36
QC2	Montréal, QC	2021-07-17 to 2021-12-01	2021-08-11	2021-10-16	36

Abbreviations: AB, Alberta; BC, British Columbia; NL, Newfoundland and Labrador; NS, Nova Scotia; NT, Northwest Territories; NU, Nunavut; ON, Ontario; QC, Québec

Device and surgical procedure-related infections in Canadian acute care hospitals, 2017–2021

Canadian Nosocomial Infection Surveillance Program[1]*

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Abstract

Background: Healthcare-associated infections (HAIs) are a significant healthcare burden in Canada. National surveillance of HAIs at sentinel acute care hospitals is conducted by the Canadian Nosocomial Infection Surveillance Program. This article describes device and surgical procedure-related HAI epidemiology in Canada from 2017 to 2021.

Methods: Data were collected from over 60 Canadian sentinel acute care hospitals between January 1, 2017, and December 31, 2021, for central line-associated bloodstream infections (CLABSIs), hip and knee surgical site infections (SSIs), cerebrospinal fluid shunt SSIs and paediatric cardiac SSIs. Case counts, rates, patient and hospital characteristics, pathogen distributions and antimicrobial resistance data are presented.

Results: Between 2017 and 2021, 2,898 device and surgical procedure-related infections were reported, with CLABSIs in intensive care units representing 69% (n=2,002) of all reported infections under surveillance. Significant rate increases were observed in adult mixed intensive care unit CLABSIs (1.08–2.11 infections per 1,000 line days, p=0.014) while decreases were observed in SSIs following knee arthroplasty (0.34–0.27 infections per 100 surgeries, p=0.05). No changes in trends were observed in the other reported HAIs. Of the 3,089 pathogens identified, the majority were gram-positive (66%), followed by gram negative (23%) and fungi (11%). Coagulase-negative staphylococci (22%) and *Staphylococcus aureus* (17%) were the most frequently isolated pathogens.

Conclusion: Epidemiological and microbiological trends among select device and surgical procedure-related HAIs are essential for benchmarking infection rates nationally and internationally, identifying any changes in infection rates or antimicrobial resistance patterns and helping inform hospital infection prevention and control and antimicrobial stewardship policies and programs.

Introduction

Healthcare-associated infections (HAIs) contribute to excess patient morbidity and mortality, leading to increased healthcare costs, longer hospital stays, and increased antimicrobial resistance (AMR) (1). Healthcare-associated infections may occur during the use of invasive devices and following surgical procedures (2). A 2017 point prevalence study in Canadian sentinel acute care hospitals found that device and surgical procedure-related infections accounted for 35.6% of all reported HAIs (3). Central line-associated bloodstream infections (CLABSIs) accounted for 21.2% of device and surgical procedure-related infections while 19.4% were associated with prosthetic implants (3). The risk of device and surgical procedure-related infections is associated with patient demographics and

comorbidities, in addition to the type of hospital in which the patient received care (4–6).

Understanding the epidemiology of device and surgical procedure-related HAIs is essential to provide benchmark rates over time, which help to inform effective antimicrobial stewardship and infection prevention and control measures. In addition, the collection and analysis of antimicrobial susceptibility data are important to inform the appropriate use of antimicrobials and help reduce AMR (7). This report provides an epidemiological overview of select device and surgical procedure-related HAIs from 2017 to 2021 in over 60 hospitals participating in the Canadian Nosocomial Infection Surveillance Program (CNISP).

Methods

Design
Since its establishment in 1994, CNISP has conducted national HAI surveillance at sentinel acute care hospitals across Canada, in collaboration with the Public Health Agency of Canada and the Association of Medical Microbiology and Infectious Disease Canada. Data are presented for the following device and surgical procedure-related HAIs: CLABSIs; hip and knee arthroplasty surgical site infections (SSIs); cerebrospinal fluid (CSF) shunt SSIs; and paediatric cardiac SSIs.

Case definitions
Device and surgical procedure-related HAIs were defined according to standardized protocols and case definitions (see **Appendix**). Complex infections, defined as deep incisional and organ/space, were included in hip and knee SSI surveillance, while CLABSIs identified in intensive care unit (ICU) settings were included in CLABSI surveillance. The adult mixed ICU, adult cardiovascular surgery intensive care unit (CVICU), paediatric intensive care unit (PICU) and neonatal intensive care unit (NICU) were included as eligible ICU settings. Adult mixed intensive care units included any adult ICU with a mix of patient types as part of the ICU patient mix (i.e. medical/surgical, surgical/trauma, burn/trauma, medical/neurosurgical).

Data source
Epidemiological data for device and surgical procedure-related infections identified between January 1, 2017, and December 31, 2021 (using surgery date for surgical site infections and date of positive blood culture for CLABSIs) were submitted by participating hospitals using standardized data collection forms. Data submission and case identification were supported by training sessions and periodic evaluations of data quality.

Statistical analysis
To calculate hip and knee SSI, CSF shunt SSI and paediatric cardiac SSI rates, the number of cases were divided by the number of surgical procedures performed (multiplied by 100). To calculate CLABSI rates, the number of cases was divided by line day denominators (multiplied by 1,000). To calculate proportions of pathogens, the number of pathogens were divided by the total number of identified pathogens. Denominators may vary, as missing and incomplete data were excluded from analyses. Median and interquartile ranges (IQR) were calculated for continuous variables. Trends over time were tested using the Mann-Kendall test. Significance testing was two-tailed and differences were considered significant at a *p*-value of ≤0.05. Analyses were conducted using R version 4.1.2 and SAS 9.4.

Results

Over 60 hospitals contributed device and surgical procedure-related infection data to CNISP between 2017 and 2021 (**Table 1**), with medium (201–499 beds) adult hospitals (n=18 sites, 29%) being the most common (data not shown). Overall, 2,898 device and surgical procedure-related infections were reported. Among all reported HAIs, CLABSIs were the most common, representing 69% (n=2,002) of all device and surgical procedure-related HAIs under surveillance. Among all SSIs reported (N=910), hip and knee infections represented 71% (n=648) of these types of infections.

A total of 3,089 pathogens were identified from device and surgical procedure-related HAI cases between 2017 and 2021. Of the identified pathogens, 66% were gram-positive, 23% were gram-negative and 11% were fungal. Coagulase-negative staphylococci (CoNS) and *Staphylococcus aureus* were the most frequently reported pathogens (**Table 2**).

Central line-associated bloodstream infections
A total of 2,002 CLABSIs were reported between 2017 and 2021, with the majority occurring in adult mixed ICUs (n=1,184, 59.1%) and NICUs (n=468, 23.4%). Overall, NICUs had the highest rates of CLABSIs between 2017 and 2021 (1.75 infections per 1,000 line days), followed by PICUs (1.71 per 1,000 line days), adult mixed ICUs (1.53 per 1,000 line days) and adult CVICUs (0.68 per 1,000 line days) (**Table A1**).

From 2017 to 2021, CLABSI rates fluctuated in NICUs and PICUs, while CLABSI rates in adult mixed ICUs nearly doubled (1.08–2.11 infections per 1,000 line days, *p*=0.014) (**Figure 1**). Though rates of CLABSI in adult CVICUs were low overall, adult CVICU CLABSI rates increased 179% from 2017 to 2020 (0.34–0.95 infections per 1,000 line days), before decreasing 10% to 0.86 infections per 1,000 line days in 2021.

During the coronavirus disease 2019 (COVID-19) pandemic, trends in CLABSI rates have varied across ICU settings. Adult mixed ICU CLABSIs continued to increase in 2020 and 2021 while CLABSIs in paediatric and NICUs decreased in 2020 and were lower overall in 2020 and 2021 compared with pre-pandemic years.

Table 1: Characteristics of acute care hospitals participating in device and surgical procedure-related healthcare-associated infection surveillance, 2021

Characteristic of hospitals	CLABSI-adult mixed ICU	CLABSI-adult CVICU	CLABSI-PICU	CLABSI-NICU	CSF shunt SSI	Paediatric cardiac SSI	Hip and knee SSI	Total unique hospitals
Total number of participating hospitals	38	7	12	16	14	6	28	62
Hospital type								
Adult	29	6	N/A	3[a]	4	N/A	14	32
Mixed	9	1	4	6	2	N/A	14	21
Paediatric	N/A	N/A	8	7	8	6	N/A	9
Hospital size								
Small (1–200 beds)	2	1	8	8	6	3	4	17
Medium (201–499 beds)	24	3	3	5	5	3	16	31
Large (500+ beds)	12	3	1	3	3	N/A	8	14

Abbreviations: CLABSI, central line-associated bloodstream infection; CSF, cerebrospinal fluid; CVICU, cardiovascular surgery intensive care unit; ICU, intensive care unit; N/A, not applicable; NICU, neonatal intensive care unit; PICU, paediatric intensive care unit; SSI, surgical site infection
[a] Three hospitals classified as "adult" also had a NICU

Table 2: Distribution and rank of the five most frequently reported gram-negative, gram-positive and fungal pathogens, 2017–2021[a]

Pathogen category	Rank	Pathogen	CLABSI N=2,002		Hip and knee N=599		CSF shunt N=126		Paediatric cardiac N=171		Total pathogens	
			n	%	n	%	n	%	n	%	n	%
Gram-positive	1	Coagulase-negative staphylococci[b]	481	22.1	120	18.5	52	39.4	21	16.2	674	21.8
	2	*Staphylococcus aureus*[c]	198	9.1	213	32.9	32	24.2	67	51.5	510	16.5
	3	*Enterococcus* spp.	396	18.2	39	6.0	6	4.5	1	0.8	442	14.3
	4	*Streptococcus* spp.	37	1.7	63	9.7	4	3.0	8	6.2	112	3.6
	5	Methicillin-resistant *S. aureus*	39	1.8	35	5.4	4	3.0	4	3.1	82	2.7
		Other gram-positive[d]	145	6.7	45	6.9	11	8.3	1	0.8	202	6.5
		Total gram-positive	1,296	59.5	515	79.5	109	82.6	102	78.5	2,022	65.5
Gram-negative	1	*Klebsiella* spp.	126	5.8	10	1.5	5	3.8	3	2.3	144	4.7
	2	*Escherichia coli*	112	5.1	20	3.1	7	5.3	1	0.8	140	4.5
	3	*Enterobacter* spp.	93	4.3	27	4.2	1	0.8	5	3.8	126	4.1
	4	*Pseudomonas* spp.	54	2.5	25	3.9	3	2.3	4	3.1	86	2.8
	5	*Serratia* spp.	50	2.3	13	2.0	2	1.5	0	0.0	65	2.1
		Other gram-negative[e]	121	5.6	35	5.4	2	1.5	5	3.8	163	5.3
		Total gram-negative	556	25.5	130	20.1	20	15.2	19	146	724	23.4
Fungi	1	*Candida albicans*	148	6.8	0	0.0	1	0.8	0	0.0	149	4.8
	2	Other *Candida* spp.[f]	166	7.6	3	0.5	1	0.8	9	6.9	179	5.8
		Other fungi[g]	13	0.6	0	0.0	1	0.8	1	0.8	15	0.5
		Total fungal	327	15.0	3	0.5	3	2.3	10	7.7	343	11.1
Total			2,179	N/A	648	N/A	132	N/A	130	N/A	3,089[h]	N/A

Abbreviations: CLABSI, central line-associated bloodstream infections; CSF, cerebrospinal fluid; *S. aureus, Staphylococcus aureus*
[a] Frequency distribution percentage rounded to the nearest tenth decimal
[b] Coagulase-negative staphylococci included *S. lugdunensis, S. haemolyticus, S. epidermidis, S. capitis, S. hominis* and *S. warneri*
[c] *Staphylococcus aureus* includes methicillin-susceptible *S. aureus* and unspecified *S. aureus*
[d] Other gram-positive pathogens included anaerobic gram-positive cocci, *Finegoldia magna, Clostridioides* spp., *Lactobacillus* spp. and others
[e] Other gram-negative pathogens included *Stenotrophomonas* spp., *Morganella morganii, Proteus mirabilis, Pantoea* spp., *Prevotella* spp., *Bacteroides fragilis* and others
[f] Other *Candida* spp. included *C. dubliniensis, C. glabrata, C. krusei, C. lusitaniae, C. parapsilosis* and *C. tropicalis*
[g] Other fungi included *Aspergillus* spp., *Trichophyton tonsurans* and unspecified fungi
[h] Up to three pathogens per device and surgical procedure-related infection were included in the analysis and exceeded the number of total reported infections overall

Figure 1: Rate of central line-associated bloodstream infection per 1,000 line days by intensive care unit type, 2017–2021
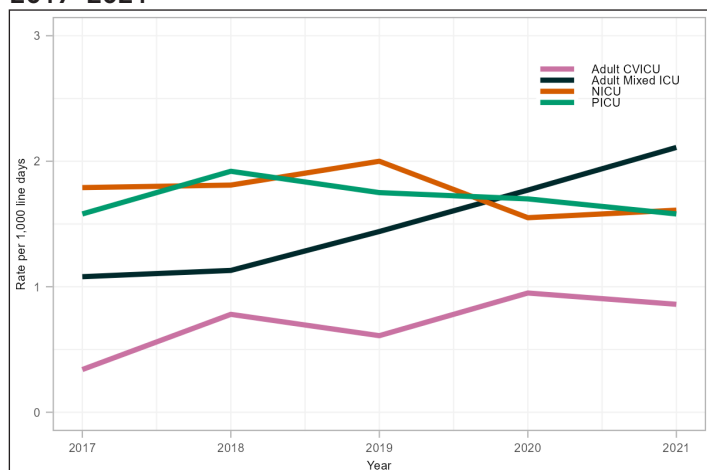
Abbreviations: CVICU, cardiovascular intensive care unit; ICU, intensive care unit; NICU, neonatal intensive care unit; PICU, paediatric intensive care unit

Among CLABSIs identified in adult mixed ICUs, the median age was 60 years (IQR=48–69 years), with males representing the majority of cases (66%). All-cause mortality within 30 days following the first positive culture, for adult mixed ICU CLABSI patients was 31.6% (n=374/1,183). Among CLABSIs identified in adult CVICUs, the median age was 65 years (IQR=50–72 years), with males representing 71% of cases. Within 30 days following the first positive culture, all-cause mortality for adult CVICU CLABSI patients was 29.6% (n=32/108). Among CLABSIs identified in PICUs, the median age was seven months (IQR=3–29 months), with males representing 60% of cases. Within 30 days following the first positive culture, all-cause mortality for PICU CLABSI patients was 10.4% (n=25/243). Among CLABSIs identified in NICUs, the median age at first positive culture was 17 days (IQR=9–38 days). Males represented 59% of NICU cases and all-cause mortality within 30 days of positive culture was 13% (n=61/468).

The most commonly identified pathogens among CLABSIs overall were CoNS and *Enterococcus* spp. (22.1% and 18.2%, respectively), which aligned with the most commonly identified pathogens among PICUs, adult mixed ICUs and adult CVICUs. Among NICU CLABSIs, CoNS and *S. aureus* were the most commonly identified pathogens.

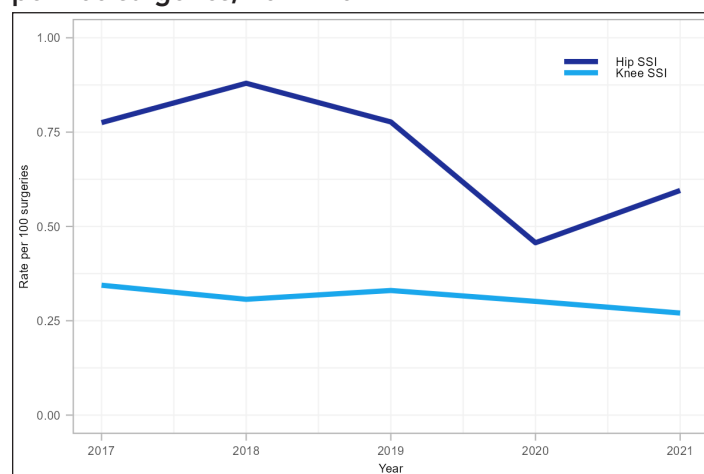## Hip and knee surgical site infections

A total of 599 complex hip and knee SSIs were reported between 2017 and 2021, of which the majority were hip arthroplasties (n=400, 67%). Among hip and knee SSIs, 53% (n=318) were organ/space infections and 47% (n=281) were deep incisional infections (**Table 3**). From 2017 to 2021, knee SSI rates decreased significantly (20.6%, 0.34 to 0.27 infections per 100 surgeries, *p*=0.05) while hip SSI rates fluctuated between 0.46 and 0.88 infections per 100 surgeries (*p*=0.33) (**Figure 2**). During the COVID-19 pandemic in 2020, knee SSI rates remained stable compared to 2019 while hip SSI rates decreased by 41%.

Table 3: Frequency of hip and knee surgical site infections by year and infection type, 2017–2021

Year	Deep incisional SSI		Organ/space SSI		All cases
	n	%	n	%	n
Hip arthroplasty					
2017	47	58.0	34	42.0	81
2018	64	65.3	34	34.7	98
2019	52	50.5	51	49.5	103
2020	25	53.2	22	46.8	47
2021	33	47.1	38	52.9	71
Overall	221	55.3	179	44.8	400
Knee arthroplasty					
2017	23	56.1	18	43.9	41
2018	18	45.0	22	55.0	40
2019	25	48.1	27	51.9	52
2020	19	57.6	14	42.4	33
2021	12	38.7	21	61.3	33
Overall	97	48.7	102	51.3	199

Abbreviation: SSI, surgical site infection

Figure 2: Rate of hip and knee surgical site infections per 100 surgeries, 2017–2021

Abbreviation: SSI, surgical site infection

In 2021, hip SSI rates increased by 30% to 0.60 infections per 100 surgeries, partially returning to rates observed in the pre-pandemic period (Figure 2 and **Table A2**).

The median patient age was 67 years (IQR=58–75 years) for hip SSIs and 66 years (IQR=59–73 years) for knee SSIs. The median time from procedure to hip and knee infections was 20 days (IQR=14–31 days) and 23 days (IQR=15–35 days), respectively. For data collected between 2018 and 2021, the median length of stay was 3 days (IQR=2–6 days) for complex SSIs following hip and knee arthroplasties. Most patients (86%, n=410/475) with an SSI following hip or knee arthroplasty were readmitted and 64% (n=296/465) required revision surgery. Within 30 days after

first positive culture, five all-cause deaths (1.6%, n=5/309) were reported among patients with a complex SSI following a hip arthroplasty while zero all-cause deaths were reported among patients with a knee arthroplasty SSI. Among hip and knee SSI cases, *S. aureus* and CoNS were the most commonly identified pathogens at 33% and 19%, respectively, and did not differ by deep or organ/space infection type (data not shown).

Cerebrospinal fluid shunt surgical site infections

Between 2017 and 2021, 126 CSF shunt SSIs were reported, with an overall rate of 2.9 infections per 100 surgeries (range: 1.7–3.4 infections per 100 surgeries, **Table A3**). Paediatric and adult/mixed hospitals infection rates were not significantly different at 3.2 and 2.5 infections per 100 surgeries, respectively (p=0.17). CSF shunt SSI rates in adult and mixed hospitals decreased throughout the COVID-19 pandemic in 2020 and 2021 (**Figure 3**), while paediatric hospital CSF shunt SSI rates initially decreased by 49% in 2020 before increasing to 3.7 infections per 100 surgeries in 2021, in keeping with the fluctuating rate trend observed since 2011 (data not shown).

Figure 3: Cerebrospinal fluid shunt surgical site infection rates per 100 surgeries by hospital type[a], 2017–2021
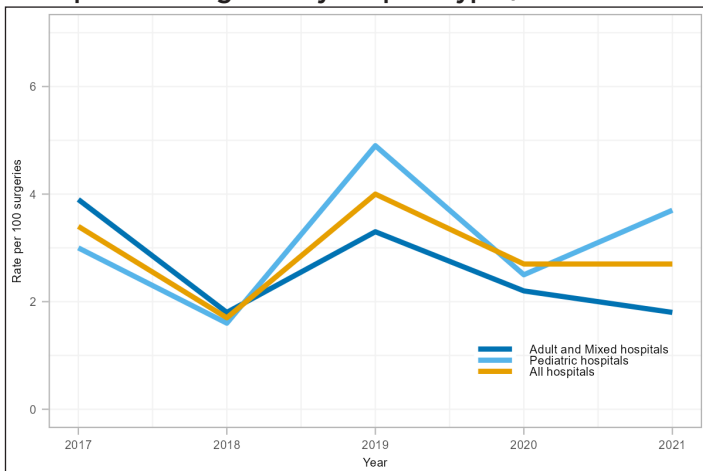
[a] All hospitals include adult, mixed, and paediatric hospitals participating in cerebrospinal fluid shunt surgical site infection surveillance

More than half of CSF shunt SSIs (53.6%, n=67/125) were identified from new surgeries while 46.4% (n=58/125) were identified from revision surgeries. The median age was 44 years (IQR=36–60 years) for adult patients and two years (IQR=0.3–7 years) for paediatric patients. Females represented 56% (n=70/125) of cases and median time from surgery to infection was 19 days (IQR=10–39 days). The most commonly identified pathogens from CSF shunt SSIs were CoNS and *S. aureus* (40% and 24% of identified pathogens, respectively). Outcome data were not collected for CSF shunt SSI surveillance.
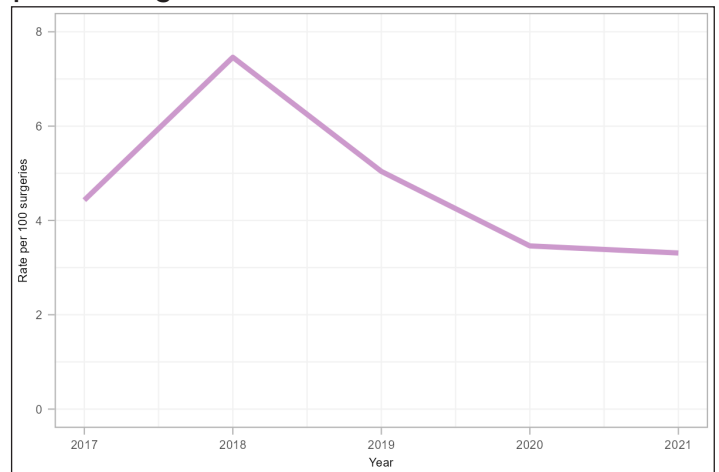
Paediatric cardiac surgical site infections

A total of 171 paediatric cardiac SSIs were reported between 2017 and 2021 (**Table 4**), most of which were superficial infections (62%). Organ/space infections accounted for 29% of these SSIs. Overall, the average paediatric cardiac SSI rate was 4.4 infections per 100 surgeries (**Table A4**). While rates remained generally consistent over the surveillance period, there was a significant increase in 2018 (7.5 infections per 100 surgeries, p<0.001) compared to the rate in 2017 (4.4 infections per 100 surgeries) (**Figure 4**). This increase was caused by outlier cases attributable to two hospitals. Since 2018, the rate decreased by 56% from 7.5 to 3.3 infections per 100 surgeries in 2021, returning to rates observed prior to 2018.

Table 4: Paediatric cardiac surgical site infection rates by year and infection type, 2017–2021

Year	Superficial incisional SSI cases		Organ/space SSI cases		Deep incisional SSI cases		All cases[a]
	n	%	n	%	n	%	
2017	17	70.8	5	20.8	2	8.3	24
2018	18	46.2	15	38.5	6	15.4	40
2019	19	54.3	14	40.0	2	5.7	35
2020	29	78.4	6	16.2	2	5.4	37
2021	23	65.7	9	25.7	3	8.6	35
Overall	106	62	49	29	15	9	171

Abbreviation: SSI, surgical site infection
[a] Excludes cases with missing infection type information

Figure 4: Paediatric cardiac surgical site infection rates per 100 surgeries, 2017–2021

The median age of patients with a paediatric cardiac SSI was 38 days (IQR=7–259 days), and the median time from surgery to onset date of infection was nine days (IQR=3–19 days). Among the three deaths reported within 30 days of infection onset (1.8% of cases), one death was unrelated to the paediatric cardiac SSI, while two deaths were attributable to the paediatric cardiac SSI. *Staphylococcus aureus* and CoNS were the most commonly identified pathogens from paediatric cardiac SSIs (55% and 17% of identified pathogens, respectively) and did not differ by superficial, organ/space or deep infection type (data not shown).

Antibiogram

Results of antimicrobial susceptibility testing for the most frequently identified gram-positive, gram-negative and fungal pathogens from device and surgical procedure-related HAIs are listed in **Table 5** and **Table 6**. The *S. aureus* isolates were resistant to cloxacillin/oxacillin (methicillin-resistant *S. aureus* [MRSA]) in 17% (n=31/179) of CLABSIs and 11% (n=34/300) of SSIs. Meropenem resistance ranged from 2%–8% in gram-negative pathogens identified from CLABSIs. No meropenem resistance was observed among pathogens isolated from SSIs. Fifty-seven vancomycin-resistant *Enterococci* were identified among CLABSIs (19%).

Table 5: Antibiogram results[a] from pathogens identified from central line-associated bloodstream infections, 2017–2021

Antibiotic	Number of resistant/number tested and %																	
	Gram-positive						Gram-negative						Fungi					
	Coagulase-negative staphylococci[b]		*S. aureus*[c]		*Enterococcus* spp.		*Klebsiella* spp.		*E. coli*		*Enterobacter* spp.		*C. albicans*		*Candida* spp. other[d]			
	# resistant/ # tested	%	# resistant/ # tested	%	# resistant/ # tested	%	# resistant/ # tested	%	# resistant/ # tested	%	# resistant/ # tested	%	# resistant/ # tested	%	# resistant/ # tested	%		
Ampicillin	16/17	94	N/A	N/A	130/350	37	99/99	100	67/95	71	55/59	93	N/A	N/A	N/A	N/A		
Cefazolin	147/176	84	18/119	15	N/A	N/A	33/81	41	27/79	34	48/48	100	N/A	N/A	N/A	N/A		
Ceftriaxone	9/10	90	3/6	50	N/A	N/A	19/86	22	18/78	23	33/59	56	N/A	N/A	N/A	N/A		
Clindamycin	108/146	74	33/116	28	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A		
Ciprofloxacin	4/11	36	N/A	N/A	10/19	53	10/85	12	27/66	41	1/74	1	N/A	N/A	N/A	N/A		
Cloxacillin/ oxacillin	222/259	86	31/179	17	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A		
Erythromycin	62/71	87	21/79	27	14/14	100	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A		
Gentamicin[e]	16/33	48	1/33	3	21/155	14	14/102	14	11/98	11	6/74	8	N/A	N/A	N/A	N/A		
Meropenem	8/9	89	N/A	N/A	N/A	N/A	4/52	8	2/41	5	1/55	2	N/A	N/A	N/A	N/A		
Piperacillin-tazobactam	N/A	N/A	N/A	N/A	3/11	22	12/80	15	16/82	20	21/60	35	N/A	N/A	N/A	N/A		
Penicillin	56/57	98	41/48	85	19/40	48	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A		
Rifampin	3/71	4	0/26	0	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A		
Trimethoprim-sulfamethoxazole	95/170	56	5/106	5	N/A	N/A	13/94	14	39/83	47	N/A	N/A	N/A	N/A	N/A	N/A		
Tobramycin	N/A	N/A	N/A	N/A	N/A	N/A	8/81	10	8/80	10	3/60	5	N/A	N/A	N/A	N/A		
Vancomycin	1/274	0	1/98	1	57/295	19	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A		
Amphotericin B	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	0/25	0	0/20	0		
Caspofungin	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	0/36	0	1/52	2		
Fluconazole	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	1/99	1	19/89	21		

Abbreviations: *C. albicans*, Candida albicans; *E. coli*, Escherichia coli; N/A, not available; *S. aureus*, Staphylococcus aureus
[a] Antibiotic/organism combinations with fewer than six tests were excluded
[b] Coagulase-negative staphylococci included *S. lugdunensis*, *S. haemolyticus*, *S. epidermidis*, *S. capitis*, *S. hominis* and *S. warneri*
[c] Included methicillin-susceptible *S. aureus* and methicillin-resistant *S. aureus* (MRSA)
[d] Other *Candida* spp. included *C. dubliniensis*, *C. glabrata*, *C. krusei*, *C. lusitaniae*, *C. parapsilosis*, and *C. tropicalis*
[e] Gentamicin synergy for gram-positive organisms

Table 6: Antibiogram results[a] from pathogens identified from hip and knee, cerebrospinal fluid shunt, and paediatric cardiac surgical site infections, 2017–2021

Antibiotic	Number of resistant/number tested and %																
	Gram-positive						Gram-negative						Fungi			
	Coagulase-negative staphylococci[b]		S. aureus[c]		Enterococcus spp.		Klebsiella spp.		E. coli		Enterobacter spp.		C. albicans		Candida spp. other[d]	
	# resistant/ # tested	%	# resistant/ # tested	%	# resistant/ # tested	%	# resistant/ # tested	%	# resistant/ # tested	%	# resistant/ # tested	%	# resistant/ # tested	%	# resistant/ # tested	%
Ampicillin	N/A	N/A	N/A	N/A	1/37	3	15/15	100	9/20	45	18/21	86	N/A	N/A	N/A	N/A
Cefazolin	49/73	67	17/171	10	N/A	N/A	4/9	44	3/17	18	20/20	100	N/A	N/A	N/A	N/A
Ceftriaxone	N/A	N/A	N/A	N/A	N/A	N/A	0/13	0	2/10	20	8/16	50	N/A	N/A	N/A	N/A
Clindamycin	16/79	20	46/220	21	0/7	0	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Ciprofloxacin	2/8	25	4/26	15	N/A	N/A	0/11	0	5/17	29	0/24	0	N/A	N/A	N/A	N/A
Cloxacillin/oxacillin	93/148	63	34/300	11	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Erythromycin	16/41	39	30/94	32	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Gentamicin[e]	N/A	N/A	1/15	7	4/10	40	1/17	6	2/20	10	1/28	4	N/A	N/A	N/A	N/A
Meropenem	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	0/6	0	0/8	0	N/A	N/A	N/A	N/A
Piperacillin-tazobactam	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	0/6	0	7/14	50	N/A	N/A	N/A	N/A
Penicillin	16/18	89	42/45	93	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Rifampin	0/33	0	0/50	0	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Trimethoprim-sulfamethoxazole	22/72	31	2/203	1	N/A	N/A	0/12	N/A	2/15	N/A	1/20	5	N/A	N/A	N/A	N/A
Tobramycin	N/A	N/A	N/A	N/A	N/A	N/A	1/14	N/A	0/16	N/A	1/26	4	N/A	N/A	N/A	N/A
Vancomycin	0/79	0	1/101	1	0/22	0	N/A	N/A	N/A	N/A	0/6	0	N/A	N/A	N/A	N/A
Amphotericin B	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Caspofungin	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Fluconazole	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A

Abbreviations: *C. albicans*, *Candida albicans*; *E. coli*, *Escherichia coli*; N/A, not available; *S. aureus*, *Staphylococcus aureus*
[a] Antibiotic/organism combinations with fewer than six tests were excluded
[b] Coagulase-negative staphylococci included *S. lugdunensis*, *S. haemolyticus*, *S. epidermidis*, *S. capitis*, *S. hominis* and *S. warneri*
[c] Included methicillin-susceptible *S. aureus* and methicillin-resistant *S. aureus* (MRSA)
[d] Other *Candida* spp. included *C. dubliniensis*, *C. glabrata*, *C. krusei*, *C. lusitaniae*, *C. parapsilosis*, and *C. tropicalis*
[e] Gentamicin synergy for gram-positive organisms

Discussion

This report summarizes 2,898 device and surgical procedure-related HAIs identified over five years of surveillance (2017 to 2021) from 62 hospitals across Canada. Rates of device and surgical procedure-related HAIs have nearly doubled for adult mixed ICU CLABSIs, while knee SSI rates have decreased significantly. The COVID-19 pandemic has had a varied impact on the rates of device and surgical procedure-related HAIs (8). In Canada, preliminary investigations suggest that the COVID-19 pandemic had an immediate but unsustained impact on HAI rate trends (9). Rates of SSIs in the CNISP network initially decreased in 2020 during the COVID-19 pandemic, when elective surgeries were postponed, before increasing towards pre-pandemic levels in 2021. Ongoing investigations continue to assess the influence of pandemic-related factors such as changes in infection control practises, screening, laboratory testing and antimicrobial stewardship on the observed rates of HAIs.

Central line-associated bloodstream infections

Where comparable data were available, the rates of CLABSI in adult ICUs (overall rate: 0.7 and 1.5 infections per 1,000 line days for CVICUs and mixed ICUs, respectively) were lower than those in the United Kingdom but higher than those in Western Australia (10,11). In the United Kingdom, 2020/2021 rates of CLABSI in the adult and cardiac ICU were 4.4 and 5.5 infections per 1,000 line days, respectively (10). In Western Australia, CLABSI rates in adult ICU settings ranged from 0.0 to 0.8 infections per 1,000 line days between 2016 and 2020, and may be lower than levels in Canada due to differences in surveillance methodologies including the number and type of hospitals under surveillance (11).

Rates of CLABSIs in the NICU and PICU fluctuated from 2017 to 2021 but were higher overall (1.75 and 1.71 infections per 1,000 line days, respectively) compared to CLABSI rates in adult mixed ICUs and adult CVICUs (1.53 and 0.68 infections per

1,000 line days, respectively). Data available from the United States from 2017 to 2021 indicate the standardized incidence ratios (defined as the ratio of observed number of infections compared to the 2015 baseline) have reported similar fluctuating trends (12–16). Higher rates of CLABSIs have been seen in other limited resource settings compared to those observed in the CNISP network; a large surveillance study of ICU in 45 countries from Latin America, Europe, Eastern Mediterranean, Southeast Asia and Western Pacific World Health Organization regions reported pooled mean CLABSI rates of 11.2 per 1,000 line days in PICUs and 4.45 in medical/surgical adult ICUs (between January 2013 and December 2018) (17).

Surgical site infections

Among SSIs included in this surveillance report, hip and knee SSIs were the most prevalent. Hip SSI rates fluctuated across reporting years, while knee SSI rates decreased significantly. Surveillance from United Kingdom indicates similar trends where hip SSI rates fluctuated and knee SSI rates decreased from 2016/2017 to 2020/2021 (18). Compared to CNISP data, hip and knee SSI rates reported in Southern Australia were higher overall; hip SSI rates increased from 2017 to 2020 (1.32 to 1.91 infections per 100 procedures), while knee SSI rates decreased by 26% (0.91 to 0.67 infections per 100 procedures) during the same time period. In accordance with results from other regions, the most common pathogens among hip and knee SSIs were *S. aureus* and CoNS, likely attributed to the contamination of implant devices by the patient's endogenous skin flora (7,18,19). Higher median age of hip and knee SSIs relate to the older age of patients requiring joint replacements and the increased likelihood of surgical complications (20). Our data indicate that frequent readmission and revision surgeries are required for SSIs, both of which place high economic and resource burdens on the Canadian healthcare system (21).

The overall rate of surgical site infections from CSF shunts was 2.9 per 100 surgeries from 2017 to 2021. Stratification of CSF shunt SSI data by paediatric and adult/mixed hospitals showed that from 2017 to 2021, adult rates (2.5 infections per 100 surgeries) and paediatric rates (3.2 infections per 100 surgeries) were not significantly different. Data from a previous CNISP surveillance indicated a fluctuating trend in CSF shunt SSI rates from 2011–2020 (22). Compared to historical data, CSF shunt SSI rates among paediatric patients from 2017 to 2021 (3.0%) were lower than those from 2000 to 2002 (4.9%), signifying a decrease in SSI rates among paediatric populations (23). Meanwhile, the rate of CSF shunt SSI among adult patients from 2017 to 2021 (2.8%) remained relatively unchanged compared to that of 2000–2002 (3.2%) (23).

The overall rate of paediatric cardiac SSI between 2017 and 2021 was 4.4 per 100 surgeries. The 2018 paediatric cardiac SSI rate should be interpreted with caution, as rates may fluctuate due to the limited number of annual cases. Literature regarding paediatric cardiac SSI rates is limited; however, a pre and post-intervention study from 2013–2017 has reported successful reduction in paediatric cardiac SSI rates from 3.4 to 0.9 per 100 surgeries in a quaternary, paediatric academic center in California following the implementation of a postoperative SSI reduction care bundle (24).

Antibiogram

The percentage of *S. aureus* isolates that were MRSA among SSIs (11%) and CLABSIs (17%) (Table 5 and Table 6) was lower in the CNISP network compared to data reported by Centers for Disease Control and Prevention where 45% and 38% of *S. aureus* isolates were MRSA for CLABSIs and SSIs, respectively (25).

Of the identified *Enterococcus* spp. in CLABSIs, 19% were vancomycin-resistant *Enterococci,* which is less than the 30.9% identified as resistant in ICUs in Poland (26). From National Healthcare Safety Network surveillance in the United States, 73% of *Enterococcus faecium* and 4% of *Enterococcus faecalis* pathogens identified from CLABSIs in ICUs were vancomycin-resistant *Enterococci* in 2020 (27). Meropenem resistance was low in gram-negative pathogens identified among CLABSIs and SSIs (0%–8%) in the CNISP network, and similar to carbapenem resistance levels reported in the United States in 2020 (1.7%–7.5% among *Klebsiella* spp.; 4.4%–6.6% among *Enterobacter* spp.; and 0.6%–2.1% among tested *E. coli* isolates) (27). Overall, antibiogram patterns observed in the CNISP network may differ compared to other countries due to differences in surveillance methodologies, antimicrobial stewardship practises, types of hospitals or patient populations under surveillance, and differences in circulating molecular strain types.

Strengths and limitations

The main strength of CNISP surveillance is the standardized collection of detailed epidemiological and molecular linked data from a large network of sentinel hospitals across Canada. There have been continued efforts to continue to increase the representativeness of CNISP, especially among northern, community, rural and Indigenous populations. From 2017 to 2021, CNISP coverage of Canadian acute care beds has increased from 32% to 35%. To further improve representativeness, CNISP and Association of Medical Microbiology and Infectious Disease Canada have launched a simplified dataset accessible to all acute care hospitals across Canada to collect and visualize annual HAI rate data. The number of hospitals participating in each HAI surveillance project differed and epidemiologic data collected were limited to the information available in the patient charts. For CLABSI surveillance, data were limited to infections occurring in the ICU settings, and as such may only represent a subset of CLABSIs occurring in the hospital. Further, differences in surveillance protocols and case definitions limit comparison with data from other countries. The CNISP continues to support the national public health response to the COVID-19 pandemic. Studies are ongoing to assess the impact of the COVID-19 pandemic on device and surgical procedure-related HAIs and AMR.

Conclusion

This report provides an updated summary of rates, pathogen distributions and antimicrobial resistance patterns among select device and surgical procedure-related HAIs and relevant pathogens. The collection and analysis of national surveillance data are important to understanding and reducing the burden of device and surgical procedure-related HAIs. These data provide benchmark rates for national and international comparison and inform antimicrobial stewardship and infection prevention and control programs and policies.

Authors' statement

Competing interests

None.

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Appendix: Case definitions

Central line-associated bloodstream infection
Only central line-associated bloodstream infections (CLABSIs) related to an intensive care unit (ICU) admission were included in surveillance.

Bloodstream infections case definition:

Bloodstream infection is **NOT** related to an infection at another site and it meets one of the following criteria:

Criterion 1: Recognized pathogen cultured from at least one blood culture, unrelated to infection at another site.

OR

Criterion 2: At least one of: fever (higher than 38°C core), chills, hypotension; if aged younger than 1 year, fever (higher than 38°C core), hypothermia (lower than 36°C core), apnea or bradycardia **AND** common skin contaminant (see list below) cultured from at least two blood cultures drawn on separate occasions or at different sites, unrelated to infection at another site. Different sites may include peripheral veins, central venous catheters or separate lumens of a multilumen catheter. Different times include two blood cultures collected on the same or consecutive calendar days via separate venipunctures or catheter entries. The collection date of the first positive blood culture is the date used to identify the date of positive culture. Two positive blood culture bottles filled at the same venipuncture or catheter entry constitute only one positive blood culture.

Central line-associated bloodstream infection case definition:

A CLABSI must meet one of the following criteria:

Criterion 1: A laboratory-confirmed bloodstream infection (LCBSI) where a central line catheter (CL) or umbilical catheter (UC) was in place for more than two calendar days on the date of the positive blood culture, with day of device placement being Day 1.

OR

Criterion 2: A LCBSI where a CL or UC was in place more than two calendar days and then removed on the day or one day before positive blood culture was drawn.

Intensive care unit-related central line-associated bloodstream infection case definition:

A CLABSI is related to an ICU if it meets one of the following criteria:

Criterion 1: CLABSI onset after two days of ICU stay.

OR

Criterion 2: If the patient is discharged or transferred out of the ICU, the CLABSI would be attributable to the ICU if it occurred on the day of transfer or the next calendar day after transfer out of the ICU.

Note: If the patient is transferred into the ICU with the CL and the blood culture was positive on the day of transfer or the next calendar day, then the CLABSI would be attributed to the unit where the line was inserted.

Common skin contaminants:

Diphtheroids, *Corynebacterium* spp., *Bacillus* spp., *Propionibacterium* spp., coagulase-negative staphylococci (including *S. epidermidis*), viridans group streptococci, *Aerococcus* spp., *Micrococcus* spp. and *Rhodococcus* spp.

Hip and knee surgical site infection
Only complex surgical site infections (SSIs) (deep incisional or organ/space) following hip and knee arthroplasty were included in surveillance.

A deep incisional surgical site infection must meet the following criterion:

Infection occurs within 90 days after the operative procedure and the infection appears to be related to the operative procedure and involves deep soft tissues (e.g. facial and muscle layers) of the incision and the patient has at least **ONE** of the following:

- Purulent drainage from the deep incision but not from the organ/space component of the surgical site
- Deep incision that spontaneously dehisces or is deliberately opened by the surgeon and is culture-positive or not cultured when the patient has at least one of the following signs or symptoms: fever (higher than 38°C) or localized pain or tenderness (a culture-negative finding does not meet this criterion)
- An abscess or other evidence of infection involving the deep incision is found on direct examination, during reoperation or by histopathologic or radiologic examination
- Diagnosis of a deep incisional SSI by a surgeon or attending physician

An organ/space surgical site infection must meet the following criterion:

Infection occurs within 90 days after the operative procedure and the infection appears to be related to the operative procedure and infection involves any part of the body, excluding the skin incision, fascia or muscle layers, that is opened or manipulated

during the operative procedure and patient has at least **ONE** of the following:

- Purulent drainage from a drain that is placed through a stab wound into the organ/space
- Organisms isolated from an aseptically obtained culture of fluid or tissue in the organ/space
- An abscess or other evidence of infection involving the organ/space that is found on direct examination, during reoperation or by histopathologic or radiologic examination
- Diagnosis of an organ/space SSI by a surgeon or attending physician

Cerebrospinal fluid shunt surgical site infection

Only patients who underwent a placement or revision of a cerebrospinal fluid (CSF) shunting device and the infection occurred within one year of surgery were included in surveillance.

Cerebrospinal fluid shunt-associated surgical site infection case definition:

An internalized CSF shunting device is in place **AND** a bacterial or fungal pathogen(s) is identified from the cerebrospinal fluid **AND** is associated with at least **ONE** of the following:

- Fever (temperature 38°C or higher)
- Neurological signs or symptoms
- Abdominal signs or symptoms
- Signs or symptoms of shunt malfunction or obstruction

Paediatric cardiac surgery surgical site infection

Only surgical site infections following open-heart surgery with cardiopulmonary bypass among paediatric patients (younger than 18 years of age) were included in surveillance.

A superficial incisional SSI must meet the following criterion:

Infection occurs within 30 days after the operative procedure and involves only skin and subcutaneous tissue of the incision and meets at least **ONE** of the following criteria:

- Purulent drainage from the superficial incision
- Organisms isolated from an aseptically obtained culture of fluid or tissue from the superficial incision
- At least **ONE** of the following signs or symptoms of infection:
  - Pain or tenderness, localized swelling, redness or heat, and the superficial incision is deliberately opened by a surgeon, and is culture-positive or not cultured (a culture-negative finding does not meet this criterion)
  - Diagnosis of superficial incisional SSI by the surgeon or attending physician

A deep incisional SSI must meet the following criterion:

Infection occurs within 90 days after the operative procedure and the infection appears to be related to the operative procedure **AND** involves deep soft tissues (e.g. facial and muscle layers) of the incision **AND** the patient has at least **ONE** of the following:

- Purulent drainage from the deep incision but not from the organ/space component of the surgical site
- Deep incision spontaneously dehisces or is deliberately opened by the surgeon and is culture-positive or not cultured when the patient has at least one of the following signs or symptoms: fever (higher than 38°C) or localized pain or tenderness (a culture-negative finding does not meet this criterion)
- An abscess or other evidence of infection involving the deep incision is found on direct examination, during reoperation or by histopathologic or radiologic examination
- Diagnosis of a deep incisional SSI by a surgeon or attending physician

An organ/space SSI must meet the following criterion:

Infection occurs within 90 days after the operative procedure and the infection appears to be related to the operative procedure **AND** infection involves any part of the body, excluding the skin incision, fascia or muscle layers, that is opened or manipulated during the operative procedure **AND** the patient has at least **ONE** of the following:

- Purulent drainage from a drain that is placed through a stab wound into the organ/space
- Organisms isolated from an aseptically obtained culture of fluid or tissue in the organ/space
- An abscess or other evidence of infection involving the organ/space that is found on direct examination, during reoperation or by histopathologic or radiologic examination

Table A1: Rate of central line-associated bloodstream infection per 1,000 line days by intensive care unit type, 2017–2021

Year	Adult mixed ICU	Adult CVICU	NICU	PICU
2017	1.08	0.34	1.79	1.58
2018	1.13	0.78	1.81	1.92
2019	1.44	0.61	2.00	1.75
2020	1.77	0.95	1.55	1.70
2021	2.11	0.86	1.61	1.58
Overall	1.53	0.68	1.75	1.71

Abbreviations: CVICU, cardiovascular intensive care unit; ICU, intensive care unit; NICU, neonatal intensive care unit; PICU, paediatric intensive care unit

Table A2: Rate of hip and knee surgical site infections per 100 surgeries, 2017–2021

Year	Hip	Knee
2017	0.78	0.34
2018	0.88	0.31
2019	0.78	0.33
2020	0.46	0.30
2021	0.60	0.27
Overall	0.70	0.31

Table A3: Cerebrospinal fluid shunt surgical site infection rates per 100 surgeries by hospital type, 2017–2021

Year	Adult and mixed hospitals	Paediatric hospitals	All hospitals[a]
2017	3.9	3	3.4
2018	1.8	1.6	1.7
2019	3.3	4.9	4
2020	2.2	2.5	2.7
2021	1.8	3.7	2.7
Overall	2.5	3.2	2.9

[a] All hospitals include adult, mixed, and paediatric hospitals participating in cerebrospinal fluid shunt surgical site infection surveillance

Table A4: Paediatric cardiac surgical site infection rates per 100 surgeries, 2017–2021

Year	Rate
2017	4.43
2018	7.46
2019	5.04
2020	3.46
2021	3.31
Overall	4.39

Healthcare-associated infections and antimicrobial resistance in Canadian acute care hospitals, 2017–2021

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Abstract

Background: Healthcare-associated infections (HAIs) and antimicrobial resistance (AMR) continue to contribute to excess morbidity and mortality among Canadians. This report describes epidemiologic and laboratory characteristics and trends of HAIs and AMR from 2017 to 2021 (*Candida auris* 2012–2021) using surveillance and laboratory data submitted by hospitals to the Canadian Nosocomial Infection Surveillance Program (CNISP) and by provincial laboratories to the National Microbiology Laboratory (NML).

Methods: Data collected from 88 Canadian sentinel acute care hospitals between January 1, 2017, and December 31, 2021, for *Clostridioides difficile* infections (CDI, carbapenemase-producing *Enterobacterales* (CPE), methicillin-resistant *Staphylococcus aureus* (MRSA) bloodstream infections (BSIs) and vancomycin-resistant *Enterococcus* (VRE) BSIs. *Candida auris* (*C. auris*) surveillance was initiated in 2019 by CNISP and in 2012 by the NML. Case counts, rates, outcomes, molecular characterization and antimicrobial resistance profiles are presented.

Results: From 2017 to 2021, increased rates per 10,000 patient days were observed for MRSA BSIs (35%; 0.84–1.13), VRE BSIs (43%; 0.23–0.33) and CPE infections (166%, 0.03–0.08). CDI rates decreased 11% (5.68–5.05). Thirty-one *C. auris* isolates were identified in Canada from 2012 to 2021, with the majority from Western Canada (68%).

Conclusion: From 2017 to 2021, the incidence of MRSA and VRE BSIs, and CPE infections increased in Canadian acute care hospitals participating in a national sentinel network (CNISP) while CDI decreased. Few *C. auris* isolates were identified from 2012 to 2021. Reporting standardized surveillance data and the consistent application of infection prevention and control practises in acute care hospitals are critical to help decrease the burden of HAIs and AMR in Canada.

Introduction

Healthcare-associated infections (HAIs), including antimicrobial resistant organisms, continue to place a significant burden on the Canadian healthcare system, and cause excess morbidity and mortality (1–5). Point-prevalence studies conducted in Canada and across Europe in 2017 have estimated 6.5%–7.9% of patients in acute care facilities had at least one HAI (6,7). The United States Centers for Disease Control and Prevention estimated that one in 31 hospitalized patients were infected with an HAI, corresponding to 687,000 infections and 72,000 deaths each year (8).

Antimicrobial resistance (AMR) threatens the treatment of HAIs, and has been identified as a global health threat by the World Health Organization (9). A global burden study estimated that 1.27 million deaths were attributable to bacterial AMR in 2019 (10). In Canada, it was estimated that 14,000 deaths were associated with AMR in 2018, with an estimated cost to the healthcare sector of $1.4 billion per year, projecting to increase to $7.6 billion per year by 2050 (11). During the coronavirus disease 2019 (COVID-19) pandemic that was declared on March 11, 2020 (12), changes in hospital infection prevention and control and antimicrobial stewardship efforts had varied impacts on the rates of HAIs and AMR (13,14). Coordinated global public health action and improved antibiotic stewardship and public awareness are crucial to identify patterns of antimicrobial resistance and prevent and control emerging infections.

In Canada, the Public Health Agency of Canada collects national data on various HAIs and AMR through the Canadian Nosocomial Infection Surveillance Program (CNISP). Established in 1994, CNISP is a collaboration between the Public Health Agency of Canada, the Association of Medical Microbiology and Infectious Disease Canada and sentinel hospitals from across Canada. The goal of CNISP is to facilitate and inform the prevention, control and reduction of HAIs and antimicrobial resistant organisms in Canadian acute care hospitals through active surveillance and reporting.

In line with the World Health Organization's core components of infection prevention and control (14), CNISP performs consistent, standardized surveillance to reliably estimate HAI burden, establish benchmark rates for national and international comparison, identify potential risk factors and assess and inform specific interventions to improve patient health outcomes. Data provided by CNISP directly supports the collaborative goals outlined in the 2017 Pan-Canadian Framework for Action for tackling AMR and antimicrobial use (9).

In this report, we describe the most recent HAI and AMR surveillance data collected from CNISP participating hospitals between 2017 and 2021. Further, for the first time, we provide an epidemiological summary of *Candida auris* (*C. auris*) isolates identified from 2012 to 2021 to contextualize this emerging pathogen in Canada.

Methods

Design
CNISP conducts prospective, sentinel surveillance for HAIs (including antimicrobial resistant organisms).

Case definitions
Standardized case definitions for healthcare-associated (HA) and community-associated (CA) infections were used. Refer to **Appendix** for full case definitions.

Data sources
Between January 1, 2017, and December 31, 2021, participating hospitals submitted epidemiologic data and isolates for cases meeting the respective case definitions for *Clostridioides difficile* infection (CDI), methicillin-resistant *Staphylococcus aureus* bloodstream infections (MRSA BSI), vancomycin-resistant *Enterococcus* bloodstream infections (VRE BSI) and carbapenemase-producing *Enterobacterales* (CPE) infections. Eligible *Candida auris* isolates (infections or colonizations) were identified by provincial laboratories and participating hospital laboratories between January 1, 2012, and December 31, 2021, while CNISP surveillance for *C. auris* began on January 1, 2019. In 2021, 88 hospitals in 10 provinces and one territory participated in HAI surveillance and are further described in **Table 1** and **Supplemental material, Figure S1**. In 2021, patient admissions captured in CNISP HAI surveillance were distributed across hospitals categorized as small (1–200 beds, n=38 sites, 43%), medium (201–499 beds, n=36 sites, 41%) and large (500+ beds, n=14 sites, 16%) (Table 1).

Epidemiologic (demographic, clinical and outcomes) and denominator data (patient days and patient admissions) were collected and submitted by participating hospitals through the Canadian Network for Public Health Intelligence—a secure online data platform.

Reviews of standardized protocols and case definitions were conducted annually by established infectious disease expert working groups; training for data submission was provided to participating CNISP hospital staff as required. Data quality for surveillance projects was periodically evaluated; methodology has been published previously (15,16).

Laboratory data
Patient-linked laboratory isolates (stool samples for CDI cases) were sent to the Public Health Agency of Canada's National Microbiology Laboratory (NML) for molecular characterization and susceptibility testing. Isolates for MRSA BSI, VRE BSI, CPE, *C. auris* (2019–2021) and paediatric CDI were submitted year-round. Adult CDI isolates were submitted annually during a targeted two-month period (March 1 to April 30). Provincial laboratories have submitted *C. auris* isolates to NML since 2012.

Statistical analysis
Rates of HAI were calculated by dividing the total number of cases identified in patients admitted to CNISP participating hospitals by the total number of patient admissions (multiplied by 1,000) or patient days (multiplied by 10,000). The HAI rates are reported nationally and by region (Western: British Columbia, Alberta, Saskatchewan and Manitoba; Central: Ontario and Québec; Eastern: Nova Scotia, New Brunswick, Prince Edward Island and Newfoundland and Labrador; Northern: Nunavut). Sites that were unable to provide case data were excluded from rate calculations and missing denominator data were estimated using their previous years reported data, where

applicable. Missing epidemiological and molecular data were excluded from analysis. The Mann-Kendall test was used to test trends. Significance testing was two-tailed and differences were considered significant at $p \leq 0.05$.

Where available, attributable and all-cause mortality were reported for HAIs. Attributable mortality rate was defined as the number of deaths per 100 HAI cases where the HAI was the direct cause of death or contributed to death within 30 days of positive culture or histopathology specimen, as determined by physician review. All-cause mortality rate was defined as the number of deaths per 100 HAI cases 30 days following positive culture.

Results

Clostridioides difficile infection

Between 2017 and 2021, overall CDI rates decreased by 11% (5.68 to 5.05 infections per 10,000 patient days); however, this decreasing trend was not significant ($p=0.142$) (**Table 2**). Stratified by source of infection, the incidence of HA-CDI decreased significantly; by 15.5% from 4.19–3.54 infections per 10,000 patient days ($p=0.050$) (**Table S1.1**). Community-associated-CDI (Appendix) rates remained stable when comparing 2017 to 2021 rates per 1,000 patient admissions.

Table 1: Summary of hospitals participating in the Canadian Nosocomial Infection Surveillance Program, by region, 2021

Details of participating hospitals	Western[a]	Central[b]	Eastern[c]	Northern[d]	Total
Total number of hospitals	29	32	26	1	88
Hospital type					
Adult[e]	12	21	16	0	49
Mixed	13	7	9	1	30
Paediatric	4	4	1	0	9
Hospital size					
Small (1–200 beds)	11	8	18	1	38
Medium (201–499 beds)	10	18	8	0	36
Large (500+ beds)	8	6	0	0	14
Admissions and discharge					
Total number of beds	9,707	12,155	3,302	22	25,186
Total number of admissions	435,550	522,198	104,531	2,272	1,064,551
Total number of patient days	3,281,963	3,860,904	952,460	6,084	8,101,411

[a] Western refers to British Columbia, Alberta, Saskatchewan and Manitoba
[b] Central refers to Ontario and Québec
[c] Eastern refers to Nova Scotia, New Brunswick, Prince Edward Island and Newfoundland and Labrador
[d] Northern refers to Nunavut
[e] Seven hospitals classified as "adult" had a neonatal intensive care unit

Table 2: *Clostridioides difficile* infection data, Canada, 2017–2021[a]

C. difficile infection data	Year									
	2017		2018		2019		2020		2021	
Number of infections and incidence rates										
Number of *C. difficile* infection cases	4,018		3,850		3,600		3,654		3,572	
Rate per 1,000 patient admissions	4.29		4.15		3.70		3.97		3.94	
Rate per 10,000 patient days	5.68		5.42		4.90		5.35		5.05	
Number of reporting hospitals	68		68		73		82		80	
Attributable mortality rate per 100 cases (%)[b]	2.6		1.2		2.2		2.5		2.2	
Antimicrobial resistance[c]	n	%	n	%	n	%	n	%	n	%
Clindamycin	149	22.0	307	48.7	221	38.9	62	17.1	64	11.9
Moxifloxacin	114	16.9	70	11.1	66	11.6	24	6.6	49	9.1
Rifampin	14	2.1	10	1.6	6	1.1	3	0.8	9	1.7
Metronidazole	0	0.0	1	0.2	0	0.0	0	0.0	0	0.0
Total number of isolates tested[d]	676	N/A	631	N/A	568	N/A	363	N/A	538	N/A

Abbreviations: *C. difficile*, *Clostridioides difficile*; N/A, not applicable
[a] All *C. difficile* isolates from 2017 to 2021 submitted to National Microbiology Laboratory were susceptible to tigecycline and vancomycin
[b] Deaths where *C. difficile* infection was the direct cause of death or contributed to death 30 days after the date of the first positive lab specimen or positive histopathology specimen. Mortality data are collected during the two-month period (March and April of each year) for adults (age 18 years and older) and year-round for children (age one year to younger than 18 years old). Among paediatric patients, there was no death attributable to healthcare-associated *C. difficile* infection
[c] *C. difficile* infection isolates are collected for resistance testing during the two-month period (March and April of each year) for adults (age 18 years and older) and year-round for children (age one year to younger than 18 years old) from admitted patients only
[d] Total number reflects the number of isolates tested for each of the antibiotics listed above

Regionally, HA-CDI rates have decreased across all regions except in the East where rates have remained relatively consistent. For CA-CDI, Central region rates remain highest overall from 2017 and 2021 (range: 1.39–1.66), followed by the Western and Eastern region. Overall CDI attributable mortality remained low and fluctuated (range: 1.2–2.6 deaths per 100 cases) from 2017 to 2021 (*p*=0.801) (Table S1.1).

The proportion of *C. difficile* isolates resistant to moxifloxacin decreased by 7.8% between 2017 (16.9%, n=114/676) and 2021 (9.1%, n=49/538) (Table 2). Since 2017, moxifloxacin resistance decreased significantly among HA-CDI isolates (8.7%, *p*=0.050) while a smaller non-significant decrease was observed among CA-CDI (3.9%, *p*=0.327) (Table S1.2). All tested *C. difficile* isolates were susceptible to vancomycin and tigecycline. There was a single case of metronidazole resistance identified in 2018. From 2017 to 2021, the prevalence of ribotype 027 associated with NAP1 decreased for both HA and CA-CDI (by 7.7% from 15.4% to 7.7% and 4.6% from 14.7% to 11.0%, respectively) (Table S1.3).

Methicillin-resistant *Staphylococcus aureus* bloodstream infections

Between 2017 and 2021, overall MRSA BSI rates increased by 35% (0.84–1.13 infections per 10,000 patient days), with a peak rate observed in 2020 (1.16 infections per 10,000 patient days) (Table 3). Stratified by case type, a continued steady increase (80%, *p*=0.05) was observed from 2017 to 2021 in CA-MRSA BSI rates compared to HA-MRSA BSI rates, which remained stable over time (range: 0.43–0.50 infections per 10,000 patient days) (Table S2.1).

In 2021, HA-MRSA BSI and CA-MRSA BSI rates were highest in Western Canada (0.47 and 0.82 infections per 10,000 patient days, respectively) (Table S2.1). Among hospital types, HA and CA-MRSA BSI rates have generally remained highest among adult and mixed hospitals. Stratified by hospital size, rates of HA-MRSA BSI were highest among medium (201–499 beds) and large size hospitals (500+ beds) while CA-MRSA BSI rates have been highest in medium size hospitals since 2019. All-cause mortality remained relatively stable from 2017 to 2021 (range: 16.2%–18.8%) (Table 3). In 2021, 30-day all-cause mortality was higher among those with HA-MRSA (24.8%) compared to those with CA-MRSA (15.0%).

Clindamycin resistance among MRSA isolates decreased significantly by 13.8% between 2017 (42.4%, n=239/564) and 2021 (28.6%, n=185/646) (*p*=0.0143) (Table 3). Since 2017, the proportion of MRSA isolates with erythromycin and ciprofloxacin resistance decreased, yet remained high (68.1% and 64.1% in 2021, respectively) in relation to other antibiotics tested.

Table 3: Methicillin-resistant *Staphylococcus aureus* bloodstream infections data, Canada, 2017–2021

MRSA BSI data	Year									
	2017		2018		2019		2020		2021	
Number of infections and incidence rates										
Number of MRSA bloodstream infections	606		767		888		873		855	
Rate per 1,000 patient admissions	0.61		0.78		0.85		0.86		0.84	
Rate per 10,000 patient days	0.84		1.05		1.14		1.16		1.13	
Number of reporting hospitals	65		62		69		81		78	
All-cause mortality rate[a]										
Number of deaths	99		144		144		152		159	
All-cause mortality rate per 100 cases	16.4		18.8		16.2		17.4		18.6	
Antimicrobial resistance[b]	n	%	n	%	n	%	n	%	n	%
Erythromycin	455	80.7	527	75.6	602	75.6	501	72.2	440	68.1
Ciprofloxacin	432	76.6	503	72.2	560	70.4	454	65.4	414	64.1
Clindamycin	239	42.4	287	41.2	297	37.3	229	33.0	185	28.6
Tetracycline	35	6.2	49	7.0	62	7.8	46	6.6	51	7.9
Trimethoprim/sulfamethoxazole	8	1.4	13	1.9	15	1.9	16	2.3	27	4.2
Rifampin	9	1.6	6	0.9	7	0.9	6	0.9	8	1.2
Tigecycline	0	0	0	0	0	0	1	0.1	2	0.3
Daptomycin	5	0.9	0	0	3	0.4	5	0.7	5	0.8
Total number of isolates tested[c,d]	564	N/A	697	N/A	796	N/A	694	N/A	646	N/A

Abbreviations: MRSA, methicillin-resistant *Staphylococcus aureus*; MRSA BSI, methicillin-resistant *Staphylococcus aureus* bloodstream infection; N/A, not applicable
[a] Based on the number of cases with associated 30-day outcome data
[b] All MRSA isolates from 2017 to 2021 submitted to National Microbiology Laboratory were susceptible to linezolid and vancomycin
[c] In some years, the number of isolates tested for resistance varied by antibiotic
[d] Total number reflects the number of isolates tested for each of the antibiotics listed above

Between 2017 and 2021, daptomycin non-susceptibility was detected in 18 isolates. All submitted MRSA BSI isolates from 2017 to 2021 were susceptible to linezolid and vancomycin.

Comparing HA-MRSA isolates to CA-MRSA isolates, clindamycin resistance was consistently higher among HA-MRSA isolates each year from 2017 (47.3% vs. 36.6%) to 2021 (36.3% vs. 23.9%) (**Table S2.2**). There were no other notable differences in antibiotic resistance patterns by MRSA BSI case type.

Between 2017 and 2021, the proportion of spa types identified as t002 (CMRSA2) and most commonly associated with MRSA infections acquired in a healthcare setting continued to decrease; from 23.5% of all HA-MRSA isolates in 2017 to 15.6% in 2021. The proportion of spa types identified as t008 (CMRSA10) and most commonly associated with MRSA infections acquired in the community continued to increase and account for the largest proportion of CA-MRSA isolates from 2017 (45.3%) to 2021 (48.9%) (**Table S2.3**).

Vancomycin-resistant *Enterococcus* bloodstream infections

From 2017 to 2018, VRE BSI rates increased by 43%, from 0.23 to 0.33 infections per 10,000 patient days while rates remained elevated but stable from 2018 to 2021 (range: 0.30–0.33 infections per 10,000 patient days) (**Table 4**). Regionally, VRE BSI rates were highest in Western and Central Canada (0.42 and 0.34 infections per 10,000 patient days in 2021, respectively) with few VRE BSIs reported in Eastern Canada (range: 0–0.02 infections per 10,000 patient days) (**Table S3.1**). Stratified by hospital type, VRE BSI rates remained highest in adult hospitals from 2017 to 2021 (range: 0.29–0.45 infections per 10,000 patient days). From 2017 to 2021, VRE BSI rates in paediatric hospitals were low, with zero cases reported in 2021. In 2021, VRE BSI rates were 0.36 infections per 10,000 patient days in both medium (201–499 beds) and large (500+ beds) size hospitals while rates in small (1–200 beds) hospitals have decreased since 2019 (0.35 to 0.14 infections per 10,000 patient days).

Table 4: Vancomycin-resistant *Enterococcus faecium* bloodstream infections data, 2017–2021

VRE BSI data	Year									
	2017		2018		2019		2020		2021	
Vancomycin-resistant *Enterococcus* bloodstream infections data										
Number of VRE BSIs	154		242		241		223		246	
Rate per 1,000 patient admissions	0.16		0.25		0.23		0.22		0.25	
Rate per 10,000 patient days	0.23		0.33		0.30		0.30		0.33	
Number of reporting hospitals	59		62		70		80		76	
Antimicrobial resistance of *Enterococcus faecium* isolates	n	%	n	%	n	%	n	%	n	%
Ampicillin	115	100	180	100	173	100	130	98.5	142	99.3
Chloramphenicol	11	9.6	4	2.2	30	17.3	28	21.2	48	33.6
Ciprofloxacin	115	100	180	100	173	100	131	99.2	142	99.3
Daptomycin[a]	9	7.8	11	6.1	7	4.0	4	3.0	2	1.4
Erythromycin	107	93.0	172	95.6	166	96.0	126	95.5	135	94.4
High-level gentamicin	44	38.3	76	42.2	57	32.9	35	26.5	26	18.2
Levofloxacin	115	100	178	98.9	173	100	130	98.5	142	99.3
Linezolid	0	0.0	2	1.1	3	1.7	1	0.8	1	0.7
Nitrofurantoin	51	44.3	54	30.0	66	38.2	54	40.9	112	78.3
Penicillin	115	100	180	100	173	100	131	99.2	142	99.3
Quinupristin/dalfopristin	8	7.0	18	10.0	18	10.4	7	5.3	4	2.8
Rifampicin	109	94.8	162	90.0	160	92.5	114	86.4	131	91.6
High-level streptomycin	39	33.9	60	33.3	42	24.3	29	22.0	39	27.3
Tetracycline	65	56.5	107	59.4	119	68.8	88	66.7	114	79.7
Tigecycline	0	0.0	1	0.6	0	0.0	0	0.0	0	0.0
Vancomycin	110	95.7	175	97.2	170	98.3	128	97.0	138	96.5
Total number of isolates tested[b]	115	N/A	180	N/A	173	N/A	132	N/A	143	N/A

Abbreviations: N/A, not applicable; VRE BSI, vancomycin-resistant *Enterococcus* bloodstream infection
[a] Clinical and Laboratory Standards Institute (CLSI) resistance breakpoints came into effect in 2019 and was applied to all years
[b] Total number reflects the number of isolates tested for each of the antibiotics listed above
Note: Aggregate mortality data reported in-text due to fluctuations in the small numbers of VRE BSI deaths reported each year

Vancomycin-resistant *Enterococcus* BSI were predominantly HA, as 89.9% (n=994/1,106) of VRE BSI reported from 2017 to 2021 were acquired in a healthcare facility. All-cause mortality remained high (32.6%) from 2017 to 2021. The incidence rates by region, hospital type and hospital size are presented in **Table S3.2**.

Between 2017 to 2021, high-level gentamicin resistance among VRE BSI isolates (*Enterococcus faecium*) decreased from 38.3% to 18.2% (p=0.05) (Table 4). Daptomycin non-susceptibility, first identified in 2016, has decreased from 7.8% (n=9 isolates) in 2017 to 1.4% (n=2 isolates) in 2021 (p=0.0143). Since 2017, the majority (99.4%) of VRE BSI isolates were identified as *Enterococcus faecium*; however, three *E. faecalis* were identified in 2018 and one in 2020 (**Table S3.3**). Among *E. faecium* isolates, the proportion identified as sequence type (ST)1478 was highest in 2018 (37.2%, n=67/180) and decreased to 7.0% (n=10/143) in 2021 (p=0.0415) (**Table S3.4**). Furthermore, the proportion of ST17 isolates significantly increased from 2017 (6.1%, n=7/115) to 2021 (53.8%, n=77/143) (p=0.05) (Table S3.4).

Carbapenemase-producing *Enterobacterales*

From 2017 to 2021, CPE infection rates have remained low. A slight increase was observed from 2017 to 2018 (0.03 to 0.06 infections per 10,000 patient days, respectively) and rates have remained stable from 2018 to 2021 (**Table 5**).

From 2017 to 2021, the majority of CPE infections (97.5%) were identified in Central (50.0%, n=101/202) and Western Canada (47.0%, n=95/202) while few infections were identified in the East (3.0%; n=6/202) (**Table S4**). From 2017 to 2021, large hospitals (500+ beds) generally reported the highest rates of CPE infections (0.05–0.12 infections per 10,000 patient days). Thirty days all-cause mortality was 19.7% (n=38/193). From 2017 to 2021, 28.9% (n=48/166) of CPE infected patients reported travel outside of Canada and of those, 91.5% (n=43/47) received medical care while abroad.

From 2017 to 2021, the prevalence of amikacin and gentamicin resistance among CPE isolates decreased by 9.4% and 6.7%, respectively, while trimethoprim-sulfamethoxazole resistance increased by 11.4% (Table 5). The predominant carbapenemases identified in Canada were *Klebsiella pneumoniae* carbapenemase (KPC), New Delhi metallo-β-lactamase (NDM) and Oxacillinase-48 (OXA-48), accounting for 88.7% of identified carbapenemases in 2021. Among submitted isolates, the proportion of carbapenemase-producing pathogens identified as *Escherichia coli* has decreased 7.1% since 2019; however, they remain the most commonly identified pathogen from 2017 to 2021 (range: 23.1%–33.7%) (**Table S5**). From 2017 to 2021, carbapenemase-producing pathogens identified as *Klebsiella pneumoniae* decreased by 7.7% while *Citrobacter freundii* increased by 9%.

Candida auris

A total of 31 isolates (colonizations and infections) have been reported to NML from 2012 to 2021. Twenty-one cases were from Western Canada, nine cases were from Central Canada and one case was reported from Eastern Canada. Approximately, one third of isolates were resistant to amphotericin B (38.7%, n=12/31) and two thirds were resistant to fluconazole (58.1%, n=18/31). One third of isolates were multidrug-resistant (resistant to two classes of antifungals) (38.7%, n=12/31). Of the eight patients with travel information, two reported no travel (25%) while six reported international travel (75%). Of the six patients with reported history of travel, five had received healthcare abroad (83%). Of the six patients with reported travel, four had known carbapenemase-producing organism status and three were positive.

Discussion

CNISP surveillance data have shown that between 2017 and 2021 there was a decreasing trend for CDI infection rates (including both HA and CA-cases) in Canada, but rates of MRSA and VRE BSI increased by 35% and 43%, respectively. Rates of CPE infection increased, but remained stable from 2018 to 2021 and few *C. auris* isolates were identified from 2012 to 2021. The COVID-19 pandemic has had a varied effect on the rates of HAIs in Canada and in the United States (13,17). Modelling HAI rates before and during the COVID-19 pandemic showed evidence of an immediate increase in HA rates of CDI while MRSA BSI, CPE and VRE BSI rates immediately decreased; however, COVID-19 pandemic status was not associated with lasting impacts on monthly rate trends in these infections (18). Studies have suggested pandemic-related factors that may have contributed to the changes in observed rates of HAIs, such as public health measures implemented in both the hospital and the community, population travel and mobility, changes in infection control practises, screening, laboratory testing and antimicrobial stewardship (14).

Declining CDI rate trends observed in the CNISP network are like those reported globally; however, rates have been reported to be higher in North America than other regions (19). The overall reduction in CDI rates across Canada suggests improvements in infection prevention and control practises and quality-improvement initiatives such as hand hygiene compliance, environmental cleaning, improved laboratory diagnostic techniques and antibiotic stewardship (20,21). In 2020, during the COVID-19 pandemic, there was evidence of an immediate increase in rates of CDI in the CNISP network, in contrast with the United States where rates continued to decline (17); however, the COVID-19 pandemic was not associated with a lasting impact on CDI rate trends.

Table 5: Carbapenemase-producing *Enterobacterales* data, Canada, 2017–2021[a]

CPE data	Year									
	2017		2018		2019		2020		2021	
Number of infections and incidence rates										
Number of CPE infections	20		36		50		41		55	
Infection rate per 1,000 patient admissions	0.02		0.04		0.06		0.05		0.06	
Infection rate per 1,000 patient days	0.3		0.6		0.8		0.6		0.8	
Infection rate per 10,000 patient days	0.03		0.06		0.08		0.06		0.08	
Number of reporting hospitals	52		51		59		75		77	
Drugs tested for antimicrobial resistance										
Antibiotics[b,c]	n	%	n	%	n	%	n	%	n	%
Piperacillin-Tazobactam	159	85.0	210	92.1	237	90.8	230	93.9	262	92.3
Ceftriaxone	173	92.5	212	93.0	250	95.8	218	88.9	244	85.9
Ceftazidime	160	85.6	192	84.2	233	89.3	203	82.9	225	79.2
Meropenem	159	85.0	198	86.8	190	72.8	149	60.8	183	64.4
Ciprofloxacin	138	73.8	158	69.3	183	70.1	173	70.6	195	68.7
Amikacin	32	17.1	44	19.3	23	8.8	24	9.8	22	7.7
Gentamicin	64	34.2	80	35.1	86	33.0	76	31	78	27.5
Tobramycin	71	38.0	101	44.3	121	46.4	91	37.1	106	37.2
Trimethoprim-sulfamethoxazole	113	60.4	143	62.7	193	73.9	184	75.1	204	71.8
Tigecycline	18	9.6	30	13.2	36	13.8	0	0	1	0.4
Total number of isolates tested[d]	187	N/A	228	N/A	261	N/A	245	N/A	284	N/A
Carbapenemases identified										
KPC	86	46.0	122	53.0	127	48.5	98	40	133	46.8
NDM	53	28.3	59	25.7	74	28.2	80	32.7	74	26.1
OXA-48	33	17.6	30	13.0	40	15.3	48	19.6	45	15.8
SME[e]	2	1.1	4	1.7	1	0.4	2	0.8	1	0.4
NDM/OXA-48	5	2.7	6	2.6	10	3.8	9	3.7	11	3.9
GES	1	0.5	1	0.4	2	0.8	0	0	1	0.4
IMP	0	0.0	3	1.3	1	0.4	1	0.4	1	0.4
NMC	4	2.1	2	0.9	4	1.5	7	2.9	15	5.3
VIM	3	1.6	3	1.3	3	1.1	0	0	1	0.4
Other	0	0.0	0	0.0	0	0.0	0	0	2	0.7
Total number of isolates tested[f]	187	N/A	230	N/A	262	N/A	245	N/A	284	N/A

Abbreviations: CPE, carbapenemase-producing *Enterobacterales*; GES, Guiana extended-spectrum β-lactamase; IMP, active-on-imipenem; KPC, *Klebsiella pneumoniae* carbapenemase; NDM, New Delhi metallo-β-lactamase; NMC, not metalloenzyme carbapenemase; N/A, not applicable; OXA-48, Oxacillinase-48; SME, *Serratia marcescens* enzymes; VIM, Verona integron-encoded metallo-β-lactamase
[a] Includes data for all CPE isolates submitted
[b] All isolates were resistant to ampicillin, and all but one to cefazolin. All carbapenemase-producing organism isolates were screened for the *mcr*-type gene which is an acquired gene associated with colistin resistance
[c] The denominator for some drugs were adjusted as minimum inhibitory concentration values were not given in all cases due to VITEK® algorithms
[d] Total number reflects the number of isolates tested for each of the antibiotics listed above
[e] Only found in *Serratia marcescens*
[f] Some isolates contain multiple carbapenemases therefore the total number of isolates tested and the number of carbapenemases indicated may not match
Note: Aggregate mortality data reported in-text due to fluctuations in the small numbers of CPE deaths reported each year

In Canada, ribotype 027 continued to decrease in prevalence from 2017 to 2021, and coincided with a 7.8% decrease in moxifloxacin resistance during this time period. Furthermore, moxifloxacin resistance remained lower (9.1% in 2021) than previously published weighted pooled resistance data for North America (44.0%) and Asia (33.0%) (22,23). The decline in RT027 prevalence from 2017 to 2021 may also have influenced the decline in CDI rates among CNISP hospitals as this ribotype has been associated with increased virulence and fluoroquinolone resistance (24).

From 2017 to 2021, MRSA BSI rates continued to increase in the CNISP network, and is attributed to the increase in CA cases. Methicillin-resistant *S. aureus* BSI is associated with increased

morbidity and mortality, increased length of hospital stays and increased HA costs among admitted patients (25–28). The 13.8% decrease in clindamycin resistance among MRSA BSI isolates from 2017 to 2021 was likely associated with the decrease in the proportion of spa type t002 (CMRSA2 epidemic type) identified among tested isolates (29). Healthcare-associated-MRSA BSI rates observed in the CNISP network from 2017 to 2020 (range: 0.43–0.50 infections per 10,000 patient days) were lower compared to those reported in Australian public hospitals (range: 0.71–0.76 infections per 10,000 patient days) (30). Based on available data in 2017 and 2018, HA-MRSA BSI rates were higher in the United States (0.52 infections per 10,000 patient days) compared to Canada (0.43–0.45 infections per 10,000 patient days) (31).

The increasing number of patients identified with CA-MRSA who were admitted to hospital in the CNISP network may be associated with a growing CA-MRSA reservoir, both in Canada and globally (32,33). Increased rates of CA-MRSA BSI suggests that strategies that target the reduction and prevention of MRSA infections in the community, especially in populations with increased risk of contracting CA-MRSA (i.e. children, athletes, incarcerated populations, people who inject drugs), such as screening and eradication of the carriage of MRSA, may be effective in reducing the burden of MRSA BSI overall (34,35).

The increase in VRE BSI rates in Canadian acute care hospitals is concerning as vancomycin resistance related to this infection has been shown to be a principal predictor of mortality, and is associated with increased hospital burden (36–38). The increase in VRE BSI rates observed in the CNISP network may be linked to changes in infection control policies, including the discontinuation of VRE screening and isolation programs in some Canadian acute care hospitals (39). The ST17 sequence type has contributed to the increased burden of VRE BSI in CNISP-participating hospitals by emerging as the predominant clone, overtaking ST1478. The ST17 sequence type is a globally disseminated VRE clone endemic in many countries but previously observed in low numbers in Canada (40). Changes in the resistance profiles of VRE BSI coincide with changes in ST distributions. The ST17 sequence type is associated with nitrofurantoin and chloramphenicol resistance, and the increase in ST17 prevalence corresponds to the increasing trend in resistance detected for these antimicrobials while daptomycin and high-level gentamicin resistance, associated with ST1478, have decreased since 2017. Vancomycin-resistant *Enterococcus* BSI trends are further impacted by the number of high-risk patients admitted to hospital (e.g. bone marrow transplants, solid organ transplants, cancer patients, etc.) (41). Although there is a lack of recent data on VRE BSI rates in comparable jurisdictions, there have been increasing trends noted in Europe (42–45), which may be associated, in part, with the introduction and spread of a new clone and gaps in infection prevention practises (44–46).

Carbapenemase-producing *Enterobacterales* infections are a significant threat to public health due to their resistance to last line antimicrobials, limiting treatment options for patients with an infection due to pathogens that have the propensity to rapidly spread in healthcare settings (47–51). While the number of CPE infections increased from 2017 to 2021 in the CNISP network, incidence remained stable from 2018 to 2021. Data on the incidence of CPE infections in other countries, such as the United Kingdom, have noted increasing incidence of CPE infections (52,53). Similarly, the number of CPE isolates identified through laboratory surveillance associated with CPE infections has increased in Switzerland from 2013 to 2018 (54). Strict implementation of infection control measures, including screening for patient travel history, is essential to reduce the transmission of CPE in Canadian acute care hospitals.

Candida auris is an emerging multi-drug resistant fungus which has been detected across multiple countries and continents including Canada, since its first detection in 2009. *Candida auris* has been associated with outbreaks in healthcare settings in many countries, including Canada and the United States (55–58), and can cause both superficial and invasive infections with mortality ranging from 30%–60% (59). Though still relatively rare in Canada, the United States reported almost 8,000 clinical and screening cases in a recent one-year period (60). We evaluated *C. auris* preparedness within CNISP hospitals in 2018 and found that most hospitals did not yet have laboratory protocols or infection prevention and control policies in place for detecting and controlling *C. auris* (61). The identification of *C. auris* in routine microbiology laboratories requires identification of *Candida* to the species level, which may not be routinely performed due to challenges in balancing cost with value added for clinical decision-making. Treatment options are limited for patients as one third of identified *C. auris* isolates in Canada were multi-drug-resistant and additional resistance can develop during antifungal therapy (62). Therefore, rapid identification, screening for colonization in at-risk patients and strict implementation of infection prevention and control measures are required to reduce the transmission of *C. auris* in Canadian healthcare settings. Continued reporting on *C. auris* in Canada is important to assess and monitor risk of this pathogen, in addition to identifying epidemiological and microbiological trends (63).

Strengths and limitations

The main strength of CNISP is the collection of standardized and detailed epidemiological and laboratory-linked data from 88 sentinel hospitals across Canada for the purpose of providing national HAI and AMR trends for benchmarking and to inform hospital infection prevention and control practises. It is important to note that data in this report include those from the early years of the COVID-19 pandemic. Therefore, rates of HAIs and AMR in 2020 and 2021 may be impacted by changes in national, regional and municipal hospital-based infection prevention and control measures.

Epidemiological data collected by CNISP were limited to information available in patient charts. Hospital staff turnover may affect the consistent application of CNISP definitions when reviewing medical charts; however, these data were collected by experienced and trained infection prevention and control staff who receive periodic training with respect to CNISP methods and definitions. Furthermore, data quality assessments were conducted to maintain and improve data quality. Recruitment efforts have increased representation and coverage of Canadian acute care beds in the CNISP network from 32% to 35% from 2017 to 2021, notably among northern, rural communities and Indigenous populations.

Next steps

Recruitment of Canadian acute care hospitals to the CNISP network in all ten provinces and three territories is an ongoing effort to improve the quality and representativeness of Canadian HAI surveillance data. Furthermore, the enhanced hospital screening practices survey is conducted annually to better understand and contextualize changes in HAI rates in the CNISP network. To further improve representativeness and generalizability of national HAI benchmark rates, CNISP and Association of Medical Microbiology and Infectious Disease Canada have launched a simplified dataset accessible to all acute care hospitals across Canada to collect and visualize annual HAI rate data. In recent years, CNISP has implemented surveillance for new and emerging pathogens, including *C. auris* and COVID-19. Studies are ongoing to assess the impact of the COVID-19 pandemic on HAI rates and AMR.

Conclusion

Surveillance findings from a national sentinel network of Canadian acute care hospitals indicate that rates of MRSA BSI, VRE BSI and CPE infections have increased from 2017 to 2021 while rates of CDI have decreased. Few cases of *C. auris* were detected in Canada from 2012 to 2021. Consistent and standardized surveillance of epidemiologic and laboratory HAI data are essential to providing hospital practitioners with benchmark rates and informing infection prevention and control and antimicrobial stewardship policies to help reduce the burden of HAI and the impact of AMR in Canadian acute care hospitals.

Authors' statement

Canadian Nosocomial Infection Surveillance Program hospitals provided expertise in the development of protocols in addition to the collection and submission of epidemiological data and lab isolates. The National Microbiology Laboratory completed the laboratory analyses and contributed to the interpretation and revision of the paper. Epidemiologists from Public Health Agency of Canada were responsible for the conception, analysis, interpretation, drafting and revision of the article.

Competing interests
None.

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Supplemental material

These documents can be accessed on the Supplemental material file.

Figure S1: Number and proportion of patient admissions included in the Canadian Nosocomial Infection Surveillance Program by hospital type and size, 2021
Table S1.1: Cases and incidence rates of healthcare-associated and community-associated *Clostridioides difficile* infection by region, hospital type and hospital size, Canada, 2017–2021
Table S1.2: Antimicrobial resistance of healthcare-associated and community-associated *Clostridioides difficile* infection isolates, Canada, 2017–2021
Table S1.3: Number and proportion of common ribotypes of healthcare-associated and community-associated *Clostridioides difficile* infection cases, Canada, 2017–2021
Table S2.1: Cases and incidence rates of healthcare-associated and community-associated methicillin-resistant *Staphylococcus aureus* bloodstream infections by region, hospital type and hospital size, 2017–2021
Table S2.2: Antimicrobial resistance of healthcare-associated and community-associated methicillin-resistant *Staphylococcus aureus* bloodstream infection isolates, Canada, 2017–2021
Table S2.3: Number and proportion of select methicillin-resistant *Staphylococcus aureus* spa types (with corresponding epidemic types) identified
Table S3.1: Number of vancomycin-resistant *Enterococcus* bloodstream infections incidence rates by region, hospital type and hospital size, 2017–2021

Table S3.2: Number of healthcare-associated vancomycin-resistant *Enterococcus* bloodstream infections and incidence rates by region, hospital type and hospital size, 2017–2021
Table S3.3: Number and proportion of vancomycin-resistant *Enterococcus* bloodstream infections isolate types identified, 2017–2021
Table S3.4: Distribution of vancomycin-resistant *Enterococcus faecium* bloodstream sequence types, 2017–2021
Table S4: Number of carbapenemase-producing *Enterobacterales* infections and incidence rates by region, hospital type and hospital size, 2017–2021
Table S5: Number and proportion of main carbapenemase-producing pathogens identified

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Appendix: Surveillance case definitions and eligibility criteria, 2021

Clostridioides difficile infection

A "primary" episode of *Clostridioides difficile* infection (CDI) is defined either as the first episode of CDI ever experienced by the patient or a new episode of CDI that occurs greater than eight weeks after the diagnosis of a previous episode in the same patient.

A patient is identified as having CDI if:

- The patient has diarrhea or fever, abdominal pain and/or ileus AND a laboratory confirmation of a positive toxin assay or positive polymerase chain reaction (PCR) for *C. difficile* (without reasonable evidence of another cause of diarrhea)
OR
- The patient has a diagnosis of pseudomembranes on sigmoidoscopy or colonoscopy (or after colectomy) or histological/pathological diagnosis of CDI
OR
- The patient is diagnosed with toxic megacolon (in adult patients only)

Diarrhea is defined as one of the following:

- More watery/unformed stools in a 36-hour period
OR
- More watery/unformed stools in a 24-hour period and this is new or unusual for the patient (in adult patients only)

Exclusion:

- Any patients younger than one year
- Any paediatric patients (aged one year to younger than 18 years) with alternate cause of diarrhea found (i.e. rotavirus, norovirus, enema or medication, etc.) are excluded even if *C. difficile* diagnostic test result is positive

CDI case classification:

Once a patient has been identified with CDI, the infection will be classified further based on the following criteria and the best clinical judgment of the healthcare and/or infection prevention and control practitioner.

Healthcare-associated (acquired in your facility) CDI case definition:

- Related to the current hospitalization:
  - The patient's CDI symptoms occur in your healthcare facility three or more days (or 72 hours or longer) after admission

- Related to a previous hospitalization:
  - Inpatient: the patient's CDI symptoms occur less than three days after the current admission (or fewer than 72 hours) AND the patient had been previously hospitalized at your healthcare facility and discharged within the previous four weeks
  - Outpatient: the patient presents with CDI symptoms at your emergency room (ER) or outpatient location AND the patient had been previously hospitalized at your healthcare facility and discharged within the previous four weeks
- Related to a previous healthcare exposure at your facility:
  - Inpatient: the patient's CDI symptoms occur less than three days after the current admission (or fewer than 72 hours) AND the patient had a previous healthcare exposure at your facility within the previous four weeks
  - Outpatient: the patient presents with CDI symptoms at your ER or outpatient location AND the patient had a previous healthcare exposure at your facility within the previous four weeks

Healthcare-associated (acquired in any other healthcare facility) CDI case definition:

- Related to a previous hospitalization at any other healthcare facility:
  - Inpatient: the patient's CDI symptoms occur less than three days after the current admission (or fewer than 72 hours) AND the patient is known to have been previously hospitalized at any other healthcare facility and discharged/transferred within the previous four weeks
  - Outpatient: the patient presents with of CDI symptoms at your ER or outpatient location AND the patient is known to have been previously hospitalized at any other healthcare facility and discharged/transferred within the previous four weeks
- Related to a previous healthcare exposure at any other healthcare facility:
  - Inpatient: the patient's CDI symptoms occur less than three days after the current admission (or fewer than 72 hours) AND the patient is known to have a previous healthcare exposure at any other healthcare facility within the previous four weeks
  - Outpatient: the patient presents with CDI symptoms at your ER or outpatient location AND the patient is known to have a previous healthcare exposure at any other healthcare facility within the previous four weeks

Healthcare-associated CDI but unable to determine which facility:

The patient with CDI DOES meet both definitions of healthcare-associated (acquired in your facility) and healthcare-associated (acquired in any other healthcare facility), but unable to determine to which facility the case is primarily attributable to.

Community-associated CDI case definition:

- Inpatient: the patient's CDI symptoms occur less than three days (or fewer than 72 hours) after admission, with no history of hospitalization or any other healthcare exposure within the previous 12 weeks
- Outpatient: the patient presents with CDI symptoms at your ER or outpatient location with no history of hospitalization or any other healthcare exposure within the previous 12 weeks

Indeterminate CDI case definition:

The patient with CDI does NOT meet any of the definitions listed above for healthcare-associated or community-associated CDI. The symptom onset was more than four weeks but fewer than 12 weeks after the patient was discharged from any healthcare facility or after the patient had any other healthcare exposure.

Methicillin-resistant *Staphylococcus aureus* (MRSA) infection

MRSA bloodstream infection (BSI) case definition:

- Isolation of *Staphylococcus aureus* from blood
AND
- Patient must be admitted to the hospital
AND
- Is a "newly identified *S. aureus* infection" at a Canadian Nosocomial Infection Surveillance Program (CNISP) hospital at the time of hospital admission or identified during hospitalization

Infection inclusion criteria:

- Methicillin-susceptible *Staphylococcus aureus* (MSSA) or MRSA BSIs identified for the first time during this current hospital admission
- MSSA or MRSA BSIs that have already been identified at your site or another CNISP site but are **new** infections

Criteria to determine NEW MSSA or MRSA BSI:

- Once the patient has been identified with a MSSA or MRSA BSI, they will be classified as a new MSSA or MRSA if they meet the following criteria: more than 14 days since previously treated MSSA or MRSA BSI and in the judgment of infection control physicians and practitioners represents a new infection

Infection exclusion criteria:

- Emergency, clinic, or other outpatient cases who are **NOT admitted** to the hospital

Healthcare-associated (HA) case definition:

Healthcare-associated is defined as an inpatient who meets the following criteria and in accordance with the best clinical judgment of the healthcare and/or infection prevention and control practitioner:

- Patient is on or beyond calendar day 3 of their hospitalization (calendar day 1 is the day of hospital admission)
OR
- Has been hospitalized in your facility in the last 7 days or up to 90 days depending on the source of the infection
OR
- Has had a healthcare exposure at your facility that would have resulted in this bacteremia (using best clinical judgment)
OR
- Any patient who has a bacteremia not acquired at your facility that is thought to be associated with any other healthcare exposure (e.g. another acute-care facility, long-term care, rehabilitation facility, clinic or exposure to a medical device)

Healthcare-associated (HA) case definition (newborn):

- The newborn is on or beyond calendar day 3 of their hospitalization (calendar day 1 is the day of hospital admission)
- The mother was **NOT** known to have MRSA on admission and there is no epidemiological reason to suspect that the mother was colonized prior to admission, even if the newborn is fewer than 48 hours of age
- In the case of a newborn transferred from another institution, MSSA or MRSA BSI may be classified as HA your acute-care facility if the organism was **NOT** known to be present and there is no epidemiological reason to suspect that acquisition occurred prior to transfer

Community-associated case definition:

- No exposure to healthcare that would have resulted in this bacteremia (using best clinical judgment) and does not meet the criteria for a healthcare-associated BSI

Vancomycin-resistant *Enterococcus* (VRE) infection

VRE BSI case definition:

- Isolation of *Enterococcus faecalis* or *faecium* from blood
AND
- Vancomycin MIC **at least** 8 µg/ml
AND
- Patient must be admitted to the hospital
AND
- Is a "newly" identified VRE BSI at a CNISP facility at the time of hospital admission or identified during hospitalization

A newly identified VRE BSI is defined as a positive VRE blood isolate more than 14 days after completion of therapy for a previous infection and felt to be unrelated to previous infection in accordance with best clinical judgment by Infection Control physicians and practitioners.

Exclusion criteria:

- Emergency, clinic, or other outpatient cases who are **NOT admitted** to the hospital

Healthcare-associated (HA) case definition:

Healthcare-associated is defined as an inpatient who meets the following criteria and in accordance with the best clinical judgment of the healthcare and/or infection prevention and control practitioner:

- Patient is on or beyond calendar day 3 of their hospitalization (calendar day 1 is the day of hospital admission)
OR
- Has been hospitalized in your facility in the last 7 days or up to 90 days depending on the source of the infection
OR
- Has had a healthcare exposure at your facility that would have resulted in this bacteremia (using best clinical judgment)
OR
- Any patient who has a bacteremia not acquired at your facility that is thought to be associated with any other healthcare exposure (e.g. another acute-care facility, long-term care, rehabilitation facility, clinic or exposure to a medical device)

Carbapenemase-producing *Enterobacterales* (CPE) infection

Case eligibility:

- Patient is admitted to a CNISP hospital or presents to a CNISP hospital emergency department or a CNISP hospital-based outpatient clinic
- Laboratory confirmation of carbapenem resistance or carbapenemase production in *Enterobacterales* spp.

Following molecular testing, only isolates determined to be harbouring a carbapenemase are included in surveillance. If multiple isolates are submitted for the same patient in the same surveillance year, only the isolate from the most invasive site is included in epidemiological results (e.g. rates and outcome data). However, antimicrobial susceptibility testing results represent all CPE isolates (including clinical and screening isolates from inpatients and outpatients) submitted between 2016 and 2020; duplicates (i.e. isolates from the same patient where the organism and the carbapenemase were the same) were excluded.

Candida auris

Patients admitted to a participating hospital or presenting to a hospital emergency department or a hospital-based outpatient clinic with laboratory confirmation of *C. auris* from any specimen.

Included in this surveillance project are all clinical or screening samples that were positive for *C. auris* by any method. Currently, *C. auris* can be identified by rRNA sequencing, Vitek MS MALDI-TOF (with either the clinical database v3.2 or later or the RUO database), or Bruker MALDI-TOF (with either the clinical database v6903 or later or the RUO database). The project also includes potential *C. auris* misidentifications or "No identification" as outlined in the **Table A1** below.

Table A1: Laboratory identification of *Candida auris*

Identification method	Identification of suspect isolates
Vitek MS MALDI Clinical database older than v3.2	*C. haemulonii* No ID/low discrimination *C. rugosa* (not a problem for v3.0 or later) *C. pulcherrima* (not a problem for v3.0 or later)
Bruker MALDI Clinical database older than v6903	No ID
Vitek 2 version 8.01	*C. haemulonii* *C. duobushaemulonii* No ID/low discrimination
Vitek 2 version before 8.01	*C. haemulonii* *C. duobushaemulonii* *C. lusitaniae* *C. famata* No ID/low discrimination
API 20C AUX	*Rhodotorula glutinis* (characteristic red colour not present) *C. sake* No ID/low discrimination
API Candida	*C. famata*
BD Phoenix yeast identification system	*C. haemulonii* *C. catenulata* No ID

Abbreviations: *C., Candida*; MALDI, Matrix Assisted Laser Desorption Ionization; MS, mass spectrometry

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