

Basic sanitation: a new indicator for the spread of COVID-19?

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Background: Basic sanitation could be a potential indicator of the spread of coronavirus disease-2019 (COVID-19) and, in this context, space-time patterns are important tools with which to elucidate the spread of disease and identify risk factors. The aim of this study was to assess a possible association between basic sanitation indices and COVID-19 rates in all the 5570 municipalities of Brazil and its spatial distribution.

Methods: Data of COVID-19 cases registered in Brazil from 28 February until 31 May 2020 and independent variables associated with basic sanitation were included.

Results: High incidence rates were significantly associated with precarious water service index (0–25% coverage) and offstandard faecal coliforms index for tap water (5–50% and 75–100% of samples tested). A significant association between high mortality rates and sewage collection (0–25% coverage)/treatment (25–50% coverage) indices was also verified. In addition, clusters with significant spatial autocorrelation were identified mainly in the North and Northeast regions for mortality and incidence rates (high-high risk areas) and for offstandard faecal coliforms index. Those regions are considered the poorest in Brazil, presenting with low incomes, human agglomerations, as well as a poor basic sanitation system, which also hinder the implementation of COVID-19-preventative measures.

Conclusions: A precarious basic sanitation infrastructure could potentially be associated with the high transmission of severe acute respiratory syndrome coronavirus-2 in Brazil.

Keywords: Brazil, COVID-19, faecal coliforms, SARS-CoV-2, sewage service, water service

Introduction

Coronavirus disease-2019 (COVID-19) has caused a pandemic and is an emerging public health concern. The infection is estimated to have an average incubation period of 5.2 d and the most commonly reported signs and symptoms are fever, cough, myalgia, fatigue, pneumonia and complicated dyspnoea, whereas less common symptoms include headache, diarrhoea, haemoptysis, rhinorrhoea and phlegm producing cough.^{1–3} To date, only appropriate symptomatic treatment, supportive care and strict prevention/control strategies have been recommended, as no specific antiviral treatment has been confirmed as effective against COVID-19.^{1,3}

Severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2), the aetiological agent of COVID-19, is widely transmitted via respiratory droplets and fomites from infected people to the respiratory systems of susceptible individuals.^{3,4} However, it has been reported that the virus replicates in gut enterocytes and it was already detected in stool from patients with severe or mild COVID-19, as well as from presymptomatic and asymptomatic individuals.^{2,4–7} Virus RNA has also been found in sewage, which raises the hypothesis of faecal-oral transmission.^{8,9}

COVID-19 patients can shed the virus in faeces for days after all respiratory symptoms have disappeared,^{9,10} although the persistence of viable SARS-Cov-2 in water and sewage has yet to be determined. A study with other coronaviruses demonstrated a 99.9% die-off of 10 d in tap water at 23°C and over 100 d at 4°C.

In sewage, the time to achieve a 99.9% die-off ranged from 2 to 3 d at 23°C.¹¹

There are different environmental viral routes from faeces to mouth, water, surfaces or places where insect vectors are present. Through these pathways, viruses may reach the mouth and infect both the intestinal and the respiratory tracts of a susceptible host.⁹ In high-risk settings, basic sanitation distribution could help balance sampling biases and contact tracing-based human testing for COVID-19,^{4,12,13} and it would also preemptively indicate epidemic foci and ascertain the exact risk of community transmission via the faecal-oral route.⁴

Approximately 2.2 billion people worldwide lack access to potable drinking water and 4.2 billion to basic sanitation,⁹ most of them in underdeveloped countries. In Brazil, according to the National Sanitation Information System,¹⁴ the average rate of sewage system coverage in 2018 was 53.2%, while the rate of sewage collection in urban areas was 60.9%, with a sewage treatment rate of 46.3%. Handling of COVID-19 and preventing its rapid and hazardous spread remains a global challenge. Wastewater monitoring could be a potential indicator of the spread of COVID-19 and, in this context, space-time patterns are important tools with which to elucidate the spread of disease and identify risk factors.¹⁵ Therefore, the aim of this study was to assess a possible statistical association between basic sanitation indices (sewage services and piped water supply) and COVID-19 rates (incidence and mortality) in all the municipalities of Brazil and its spatial distribution, to identify possible high-risk transmission clusters.

Materials and Methods

Study type and area description

We carried out an epidemiological and ecological study using spatial analysis tools. It included all confirmed COVID-19 cases and deaths in Brazil from 28 February to 31 May 2020 and basic sanitation data from all the 5570 municipalities of Brazil.

Brazil occupies an area of 8.51 million km², almost 50% of South America, and has a total population of 210.1 million inhabitants.¹⁶ Brazil is the fifth largest country of the world with the sixth largest population.¹⁶

Data were collected from the website covid.saude.gov.br, a public domain platform supplied daily by the Brazilian Ministry of Health, considering the municipality of residence and excluding those cases that did not present localisation records. Likewise, basic sanitation data were available in the public domain at the National Sanitation Information System.¹⁴

Variables and measures

The COVID-19-dependent variables selected were:

1. The incidence rates of the municipalities, calculated as the total number of COVID-19 cases (on 31 May 2020) divided by the size of the population, expressed per 100 000 inhabitants.
2. The mortality rates of the municipalities, calculated as the total number of COVID-19 deaths (on 31 May 2020) divided

by the size of the population, expressed per 100 000 inhabitants.

The basic sanitation independent variables selected were:

1. The total water service index, defined as the total number of urban and rural populations served with piped water by the service provider on the last day of the reference year (2018), expressed as a percentage.
2. The total sewage service index, defined as the total number of urban and rural populations served with a sanitary sewer service by the service provider on the last day of the reference year (2018), expressed as a percentage.
3. The sewage treatment index (wastewater treatment), corresponding to the annual volume of sewage collected in the service provider's area of operation that was submitted for treatment, also on the last day of the reference year (2018).
4. The faecal coliforms index for tap water, used as an indicator of faecal contamination and representing the hygienic sanitary conditions of the determined area on the last day of the reference year (2018), calculated by dividing the number of offstandard analyses by the total number of analyses performed and expressed as a percentage. The incidence of total coliforms in up to 5% of the samples examined was considered acceptable, as most Brazilian municipalities have more than 20 000 inhabitants.¹⁷

Basic sanitation indices were grouped according to the coverage into 0–25%, 25–50%, 50–75% and 75–100% to perform associations with COVID-19 mortality and incidence rates, except for the faecal coliforms index, which was stratified into 0–5%, 5–50%, 50–75% and 75–100% (offstandard samples). The faecal coliforms index was stratified into four different categories considering the cut-off point of 5% offstandard samples, established by the Brazilian Ministry of Health as acceptable for total coliforms detection. Descriptive and statistical data analysis was performed. D'Agostino and Pearson tests were applied to analyse the parametric distribution and the comparison between groups was performed by a Kruskal-Wallis test, to define the sanitation coverage cut-off point associated with COVID-19 rates. Data were considered statistically significant if $p < 0.05$.

Spatial cluster analysis

The incidence rates were represented in thematic maps stratified into five equal categories: (1) 0 (without case records or without information), (2) 0.1–200, (3) 200–400, (4) 400–600 and (5) ≥ 600 . Likewise, the mortality rates were also represented in thematic maps stratified into five equal categories: (1) 0 (without death records or without information), (2) 0.1–20, (3) 20–40, (4) 40–60 and (5) ≥ 60 . The total water service index (water supply) was stratified into satisfactory (95–100%) and unsatisfactory (0–95%); the total sewage service index (sewage collection) was stratified into satisfactory ($\geq 75\%$) and unsatisfactory (0–75%); the sewage treatment index was stratified into satisfactory ($\geq 75\%$) and unsatisfactory (0–75%); and faecal coliforms index was stratified into satisfactory (0–5%) and unsatisfactory (5–100%).¹⁷

Spatial autocorrelation of all variables was calculated by the index of the global Moran univariate, to verify if the spatial distribution occurs randomly in space. We elaborated on a spatial proximity matrix obtained by the contiguity criterion, adopting a significance level of 5%. This index varies from -1 to $+1$, where values close to zero indicate spatial randomness; values between 0 and $+1$ indicate positive spatial autocorrelation and, between -1 and 0 , negative spatial autocorrelation. The global Moran autocorrelation coefficient is based on the cross-products of the deviations from the mean, calculated for the observations as

$$I = \frac{\left[(n \sum_i^n \sum_j^n \omega_{ij} (y_i - \bar{y})(y_j - \bar{y})) \right]}{\left[\sum_i^n (y_i - \bar{y})^2 \sum_j^n \omega_{ij} \right]}$$

where ω_{ij} is a contiguity matrix element (ω); y_i is the incidence rate of municipality i ; y_j is the incidence rate of municipality j ; \bar{y} is the mean of sample; and the symbol n represents the total number of municipalities.

The local Moran's index (Local Index of Spatial Association [LISA]) was used to compare the value of each municipality with their neighbours and also to verify the spatial dependence. Thus, to evaluate local spatial clustering, a measure of the association for each unit was used to verify whether the stationary process is local.

$$I = \frac{n \left[\left(Z_i \sum_j^n \omega_{ij} Z_j \right) \right]}{\left(\sum_j^n Z_j^2 \right)}$$

where $Z_i = y_i - \bar{y}$; $Z_j = y_j - \bar{y}$; ω_{ij} is the contiguous matrix element ω ; y_i is the incidence rate of municipality i ; y_j is the incidence rate of municipality j ; \bar{y} is the sample mean; and the symbol n represents the total number of cities.

A scatter plot was obtained with the following spatial quadrants: Q1 (high/high) and Q2 (low/low), which indicate municipalities with similar values to their neighbours, and represents concordance areas with positive spatial association aggregates; and Q3 (high/low) and Q4 (low/high), with different values that represent transition areas with negative spatial association aggregates. The significant results were visually expressed on Moran maps.

Software

We used Microsoft Office Excel 2010 (Microsoft Corporation; Redmond, Washington, United States) and GraphPad Prism 8 software (GraphPad Software; San Diego, California, United States) for descriptive analysis and data tabulation; QGIS 3.4.11 (QGIS Development Team; Open Source Geospatial Foundation Project) and TerraView 4.2.2 (Instituto Nacional de Pesquisas Espaciais, INPE, São José dos Campos, SP, Brazil) were used to perform spatial analysis and to create choroplethic maps.

Results

All the municipalities were categorised into four groups according to the percentages of piped water services, sewage collection, sewage (wastewater) treatment and faecal coliform index, as shown in Figure 1A-H. Interestingly, we observed higher inci-

dence and mortality rates for COVID-19 in municipalities with lower water service indices (0–25% [mean±SD 289.8±369.4 and 8.450±12.28, respectively; $p < 0.0001$; Figure 1A] and 25–50% [212.8±359.3 and 6.378±11.64, respectively; $p < 0.0001$; Figure 1B]) than in those with a higher water service index (50–75% and 75–100%, respectively). Similarly, higher incidence and mortality rates were identified in municipalities with lower sewage collection (sewerage) indices of 0–25% (198.1±318.6 and 7.013±11.26, respectively; $p < 0.0001$; Figure 1C) and 25–50% (160.5±227.0 and 7.871±12.37, respectively; $p < 0.0001$; Figure 1D) compared with municipalities with 75–100% sewage collection. No differences were observed between the incidence rate and sewage (wastewater) treatment index (Figure 1E). However, a higher mortality rate was observed in municipalities with 25–50% sewage treatment (7.465±10.87; $p < 0.001$; Figure 1F) than in those with 0–25%, 50–75% and 75–100% sewage treatment. Likewise, we observed a higher incidence rate in municipalities with offstandard faecal coliforms indices of 5–50% and 75–100% (197.6±243.7 and 301.0±455.6, respectively; $p < 0.01$; Figure 1G) than those of 0–5%.

The distribution and spatial analysis of incidence and mortality rates for COVID-19 throughout the Brazilian territory are shown in Figure 2A-D. SARS-CoV-2 cases were widely distributed throughout the country and a total of 512 553 cases were recorded in 73.76% ($n=4110$) of municipalities. However, the highest incidence rates ($\geq 600/100\ 000$ inhabitants) were observed in 208 municipalities, with 44.71% ($n=93$) located in the North and 40.86% ($n=85$) in the Northeast regions (Figure 2A). Moreover, a total of 29 281 deaths from COVID-19 were registered in 1711 (30.70%) municipalities in Brazil. In addition, the highest mortality rates ($\geq 40/100\ 000$ inhabitants) were registered in 102 municipalities: 44.11% ($n=45$) located in the North and 36.27% ($n=37$) in the Northeast regions (Figure 2B).

The prospective spatial analysis identified a significant spatial autocorrelation ($I=0.511$; $p=0.001$) for the COVID-19 incidence rate and 468 municipalities were categorised as risk areas for the disease (high/high). The largest clusters were observed in municipalities from the Northeast (53.20%; $n=249$) and North (35.04%; $n=164$) regions (Figure 2C). Similarly, significant spatial autocorrelation ($I=0.423$; $p=0.001$) was reported for deaths by COVID-19. In addition, we identified that 396 municipalities were considered risk areas for mortality by COVID-19 with high/high clusters distributed mainly in municipalities located in the Northeast (174; 43.93%) and North (142; 35.65%) regions (Figure 2D).

Accordingly, Figure 3A-H shows the spatial distribution of the four main indicators of basic sanitation. The total water service index presented a heterogeneous spatial distribution across the country and it was considered satisfactory only in 14.88% (829/5571) of municipalities (Figure 3A). We also observed significant spatial autocorrelation ($I=0.224$; $p=0.001$) and a high-high cluster (with 697 municipalities), mainly in the Southeast region (Figure 3E). Interestingly, the highest sewage collection indices (Figure 3B) were identified in the Southeast and Midwest regions. There was significant spatial autocorrelation ($I=0.408$; $p=0.001$) and the formation of a large cluster with 624 municipalities, mainly in the Southeast region (Figure 3F). Similarly, the sewage treatment index showed a heterogeneous distribution in Brazil (Figure 3C). However, clusters with significant autocorrelation ($I=0.265$; $p=0.001$), comprising 524 municipalities from

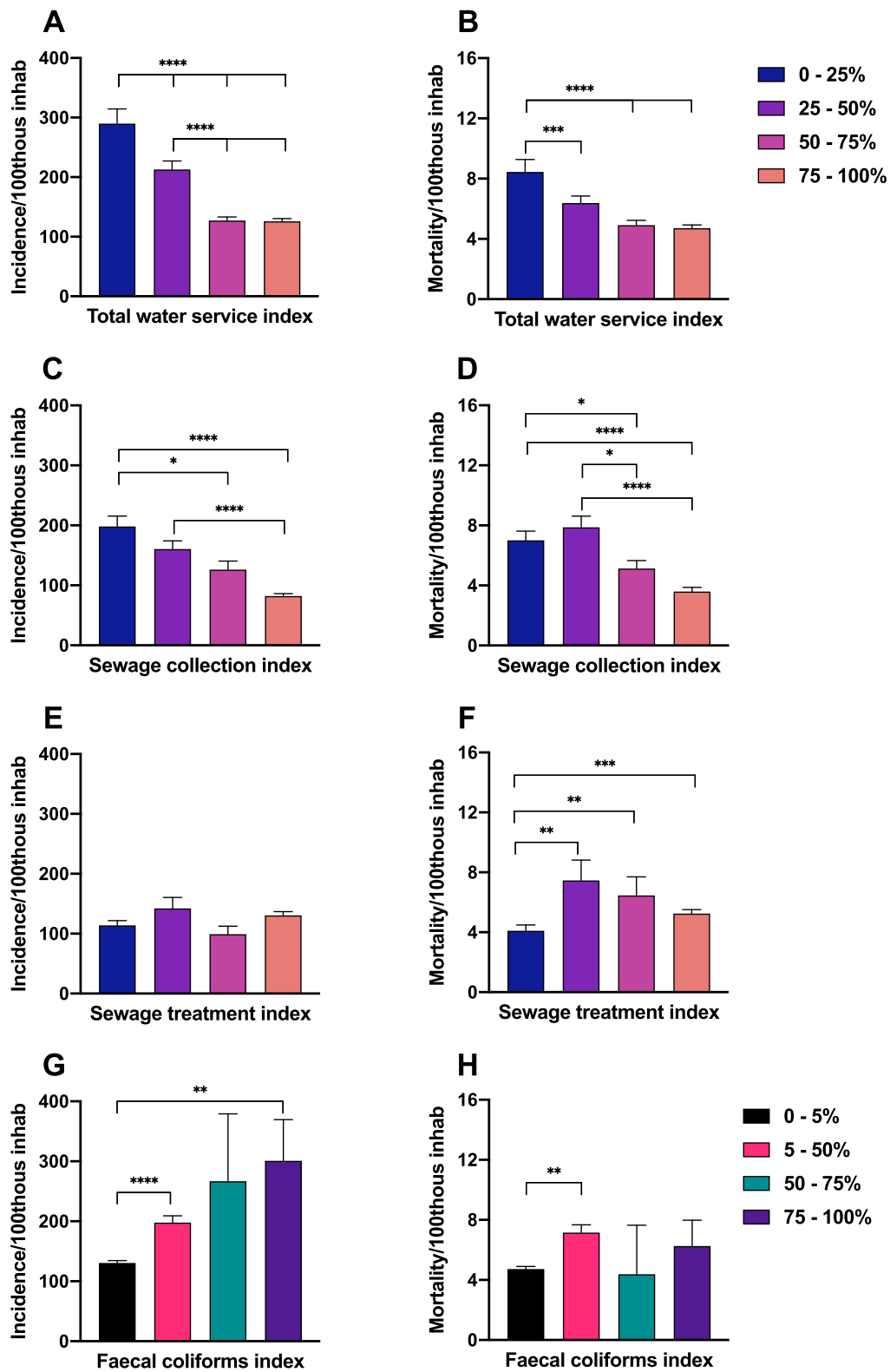


Figure 1. Incidence and mortality rates per COVID-19 in all the municipalities of Brazil according to the four main indicators of basic sanitation. Incidence per total water service index (A), mortality per total water service index (B), incidence per sewage collection index (C), mortality per sewage collection index (D), incidence per sewage treatment index (E), mortality per sewage treatment index (F), incidence per faecal coliforms index (G) and mortality per faecal coliforms index (H).

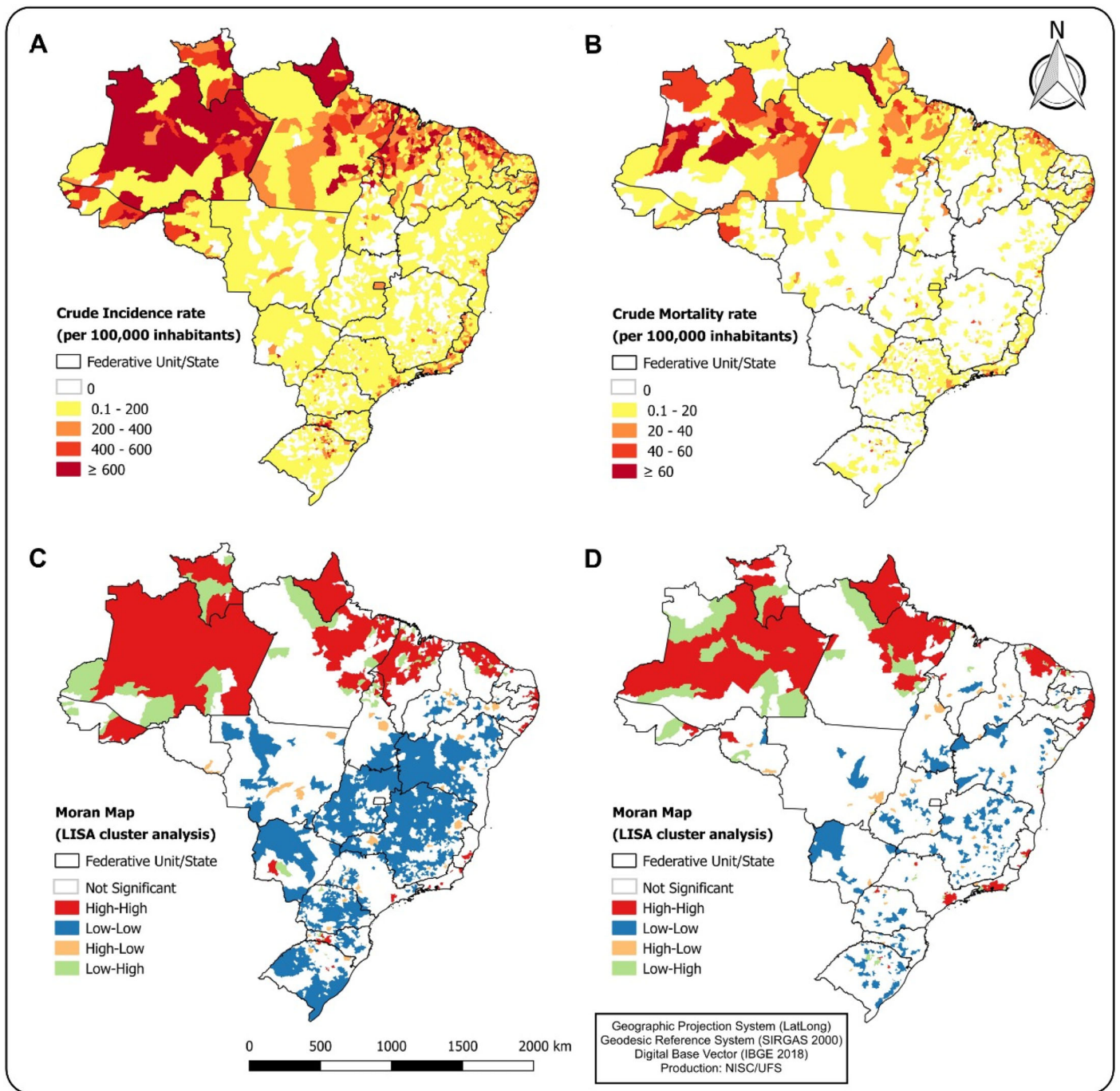


Figure 2. Spatial distribution of coronavirus disease in all municipalities of Brazil. Crude incidence rates (A), crude mortality rates (B), LISA cluster analysis of incidence rates (C) and LISA cluster analysis of mortality rates (D).

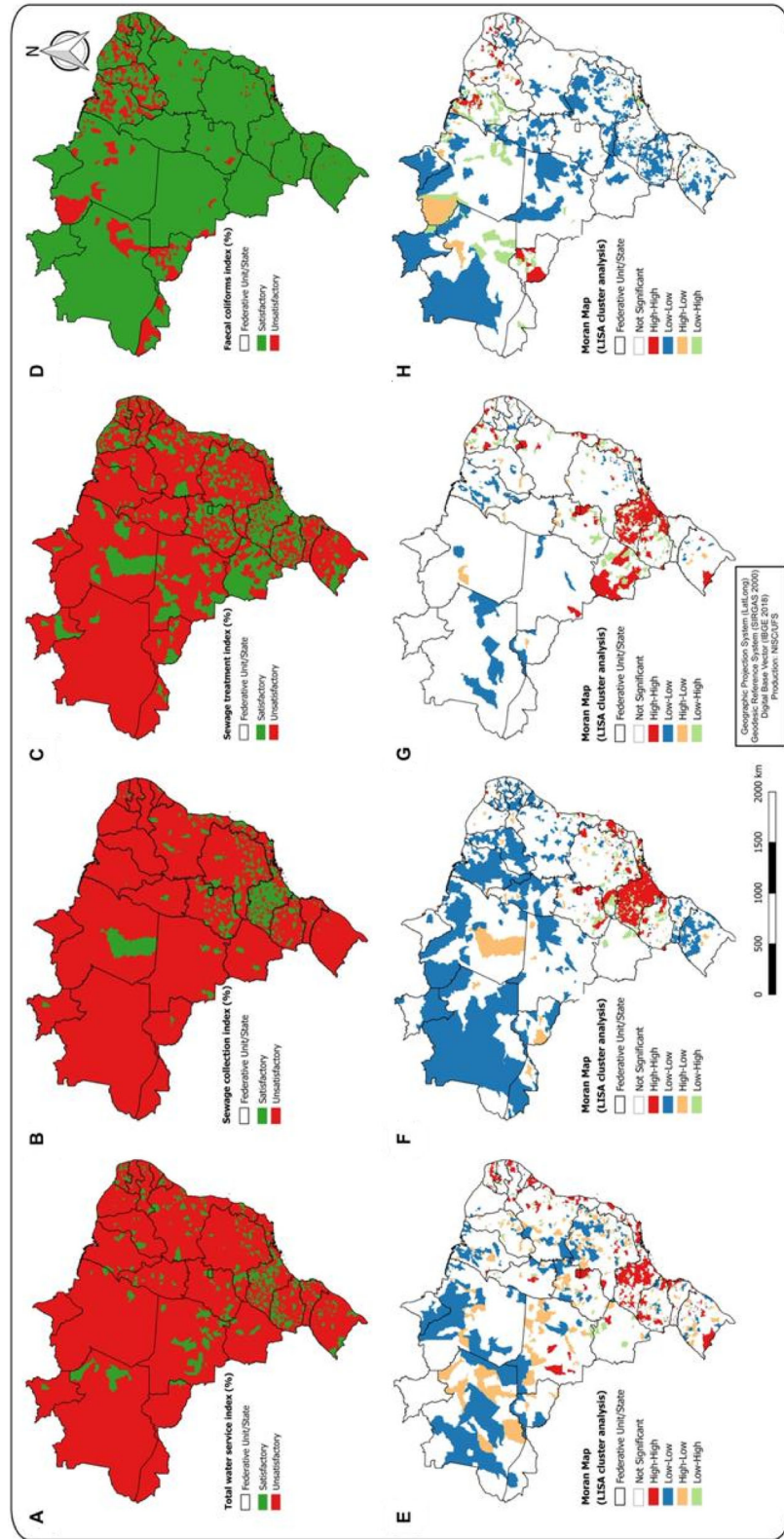


Figure 3. Spatial distribution of the four main indicators of basic sanitation in Brazilian municipalities. Total water service index (A), sewage collection index (B), sewage treatment index (C), faecal coliforms index (D), LISA cluster analysis of total water service index (E), LISA cluster analysis of total water service index (F), LISA cluster analysis of faecal coliforms index (G) and LISA cluster analysis of faecal coliforms index (H).

the Southeast and Midwest regions, were observed (Figure 3G). On the other hand, the faecal coliforms index was inadequate in 9.71% (541/5571) of municipalities, most of which were located in the Northeast and North regions (Figure 3D). We also observed clusters with spatial autocorrelation ($I=0.127$; $p=0.001$) in most of the municipalities ($n=109$) of the Northeast region (Figure 3H).

Discussion

There is evidence indicating that SARS-CoV-2 results in enteric infection.⁵ In addition, mild symptoms such as diarrhoea and SARS-CoV-2 viral RNA segments in the stool of COVID-19 patients have been reported.^{1,2,5,7,10} The rapid and constant increase of COVID-19 cases in Brazil along with detection of the virus in faeces and wastewater has raised questions about the transmission routes.

Despite the implementation of preventative measures such as handwashing, social distancing practices and face mask-wearing, thousands of Brazilians were infected by COVID-19. In this study, we observed that most COVID-19 cases (incidence rate) and deaths (mortality rate) were distributed among municipalities with the lowest rates of access to safe water (50% coverage cut-off) or a sewerage system (50% coverage cut-off), as well as a higher incidence rate in municipalities with a offstandard faecal coliforms index for tap water ($>5\%$). Nonetheless, a significantly higher mortality rate was verified in municipalities with 25–50% sewage treatment compared with municipalities with 0–25% sewage treatment coverage. Considering that Brazil is a large country with social, educational and economic disparities, other factors related to COVID-19 mortality could be responsible for this finding, such as limited access to health services, no income with which to purchase medication and difficulties in following preventative/treatment measures. Although the faecal-oral transmission of SARS-CoV-2 and its survivability in water are still under investigation,¹⁸ the results presented herein reinforce the warning for universal access to safe water, along with other COVID-19 preventative measures in Brazil, through the expansion and monitoring of basic sanitation services.

Insufficient sanitisation of faecal infected drinking water enables viral transmission from consumption, inhalation or aspiration (water-bathing) or through contact with the skin or eyes (swimming).¹⁹ However, SARS-CoV-2 is a coated RNA coronavirus with a brittle exterior membrane and therefore is less environmentally stable, being deactivated by oxidants.¹⁹ Nevertheless, studies have reported that coronaviruses can remain in water or wastewater for days or weeks.²⁰ Despite the number of Brazilian households served with piped water and a sewerage system increasing over time, there are still areas not covered, in particular peripheral urban, rural and low-income clusters.²¹ Historically, several governmental strategies have been developed aiming for universal basic sanitation access in Brazil, such as PLANASA (National Sanitation Plan) in 1969 and Law n° 11.445 (2007).²² Despite these efforts, Brazil currently occupies 103rd place in the world rankings of access to basic sanitation, behind other South American countries such as Uruguay, Argentina and Venezuela.^{22,23} An improvement in quality of life is expected from

limited contact with pathogenic microorganisms.²⁴ However, a large number of people exposed to raw sewage (e.g. wastewater running through open-air ditches that bisect neighbourhoods, rivers used as 'open sewers' and the proximity of sewers or septic tanks to groundwater tables) remain in continuous exposure to several pathological agents.

Hereupon, the recommendation of handwashing with soap as a control measure against COVID-19 infection is not possible for part of the Brazilian population as access to water is not universal. According to Wolf et al.,²⁵ one in four people in the world do not have access to a place with soap and water to clean their hands after potential contact with faeces. This reality is reflected in the high rates of infection and death caused by enteroparasitosis. Historically, Brazil has had difficulties in controlling infectious diseases with established faecal-oral transmission, such as ascariasis, giardiasis and amebiasis.²⁶ In 2013, more than 16% of hospitalisations in Brazil due to water-borne diseases could have been avoided if the basic sanitation system was satisfactory.²⁷ Indeed, disruptions to improvements in healthcare are also associated with an absence of basic structures, poor health education, the sharing of news without any scientific basis, as well as no control or monitoring of preventative measures.

In general, both distribution and spatial analysis of COVID-19 incidence/mortality registered higher indices in the North and Northeast regions, with risk areas (high/high) for the disease shown in the spatial correlation analysis. Not surprisingly, most of the municipalities with a faecal coliforms index considered inadequate ($>5\%$) are located in those regions. The main preventative measures recommended to contain the new coronavirus pandemic are not available to many residents of communities and peripheries in Brazilian cities. The outstanding inequality in Brazil results in an asymmetric impact on the population and, therefore, different measures are mandatory. The heterogeneous distribution of the sewage treatment index across the country highlights the social and economic disparities. The poorest regions of Brazil are the North and Northeast, with a deficient sanitation system alongside other poverty factors such as low income, human agglomerations, precarious housing, a low educational level and a poor health system. Considered together, these factors hinder the implementation of preventative measures such as handwashing, social distancing and access to preventative information and healthcare, thus contributing to the spread of COVID-19. Areas without infrastructure are associated with poverty and carry a stigma. Conversely, we verified high/high clusters for COVID-19 in the Southeast region through the spatial correlation analysis. Despite presenting the highest sewage collection indices, those high/high clusters in the Southeast region, where major urban centres are located, are probably associated with slums, while poor areas without infrastructure, with human agglomerations and low income, are mainly located in urban peripheries. Considering the current knowledge about this disease, those factors probably influence COVID-19 mortality and incidence rates together with basic sanitation services, as previously indicated herein.

The detection of SARS-CoV-2 in the faeces of infected patients raised concern regarding a faecal-oral transmission route. Soares et al.²⁸ proposed monitoring the presence of SARS-CoV-2 in sewers as a preventative strategy in places where mass testing is not possible and to also identify risk areas. In

Australia and Italy, sewage samples for the detection of SARS-CoV-2 were determined as an epidemiological screening method for the spread of COVID-19.²⁸ In France, sewer contamination was verified just before the peak of COVID-19 cases and was also associated with the number of deaths.²⁸ The detection of SARS-CoV-2 in frozen sewage samples collected in the first quarter of 2019 reinforces the importance of monitoring sewerage and tap water.²⁸ Likewise, it indicates that the virus was already circulating, with low or even without pathogenicity to humans. Nonetheless, hospitalisations due to respiratory syndromes without an apparent cause, during the referred to period, could have been poorly investigated, reflecting a possible under-reporting of COVID-19 cases. Anyway, the circulation of the virus in different hosts and environments may have contributed to the development of gene mutations associated with its pathogenicity.

Wastewater monitoring can be useful for early warning surveillance of the spread of SARS-CoV-2. When recurrent individual testing is not possible, aggregated information concerning the outbreak level might be useful for monitoring the evolution and effectiveness of the implemented measures.²⁹ Daughton³⁰ has already suggested that wastewater-based epidemiology could help to determine an increasing or decreasing trend of SARS-CoV-2 spread. Several research groups are monitoring wastewater for SARS-CoV-2 RNA specifically for this purpose. The level of infection and the temporal trends could be assessed by comparing/associating the viral load in wastewater with the overall population.²⁹

Evidence points to a strong relationship between COVID-19 cases and a poor basic sanitation system, reinforcing the likelihood of SARS-CoV-2 being spread through a faecal-oral transmission route. However, areas of poverty present other factors that contribute to the spread of infectious diseases, such as human agglomerations, precarious housing and a low educational level. Mass testing restrictions, asymptomatic individuals and a fragile basic sanitation system reinforce the importance of SARS-CoV-2 wastewater monitoring.²⁹ Until now, no cases of SARS-CoV-2 faecal-oral transmission have been reported, which suggests that this infection route is improbable in quarantined accommodation or hospitals. Conversely, potential faecal-oral transmission may pose a greater risk in enclosed living places such as hostels, camping facilities, nursing homes and cruise ships. Even so, more studies are needed to investigate this potential transmission path, through the detection of the infective virus in sewage, faeces, tap water and toilets, together with its survivability time. If COVID-19 faecal-oral transmission is attested, imperative consequences for public health and also for COVID-19 pandemic control measures will be perceived.

This study was conducted using secondary data reported during the pandemic and this source of data may have datasets missing and the number of COVID-19 cases in Brazil is probably under-reported. Additionally, other important data about comorbidities, level of education, socioeconomic and race inequities were not available. Therefore, further studies using spatial regression modelling are needed to clarify the intersection of these variables, sanitation conditions and COVID-19 mortality. Despite the limitations found here, our findings provide relevant data for decision-making and the planning of new public policies to face the epidemic in Brazil, considering the living conditions of the population.

Authors' contributions: TRM, DST and PLS: conception of the study. MBS, CJNR, TRM, ADS, DST and PLS: design of the study; RRS, CJNR, TRM, ADS, DST and PLS: analysis, interpretation of data and drafting of the article. All the authors provided final approval for the article to be submitted.

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Ethical approval: As this research study analysed data in the public domain available online without any kind of authorisation, the need for approval for scientific use is not mandatory.

Data availability: The datasets were derived from sources in the public domain: Coronavirus Brasil: <https://covid.saude.gov.br/> and National Sanitation Information System: <http://www.snis.gov.br/diagnostico-anual-agua-e-esgotos/diagnostico-dosservicos-de-agua-e-esgotos-2018>.

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