Infectious Disease Modelling 10 (2025) 325–337

Contents lists available at ScienceDirect

Infectious Disease Modelling

journal homepage: www.keaipublishing.com/idm

Evaluation of wastewater percent positive for assessing epidemic trends - A case study of COVID-19 in Shangrao, China

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A R T I C L E I N F O

Article history:
Received 22 May 2024
Received in revised form 10 November 2024
Accepted 10 November 2024
Available online 16 November 2024
Handling Editor: Dr. Raluca Eftimie

Keywords:
Wastewater-based epidemiology
Lag time
Correlation
Positive ratio
Wastewater percent positive
Epidemic trend

A B S T R A C T

Objective: This study aims to assess the feasibility of evaluating the COVID-19 epidemic trend through monitoring the positive percentage of SARS-CoV-19 RNA in wastewater.

Method: The study collected data from January to August 2023, including the number of reported cases, the positive ratio of nucleic acid samples in sentinel hospitals, the incidence rate of influenza-like symptoms in students, and the positive ratio of wastewater samples in different counties and districts in Shangrao City. Wastewater samples were obtained through grabbing and laboratory testing was completed within 24 h. The data were then normalized using Z-score normalization and analyzed for lag time and correlation using the xcorr function and Spearman correlation coefficient.

Results: A total of 2797 wastewater samples were collected. The wastewater monitoring study, based on sampling point distribution, was divided into two phases. Wuyuan County consistently showed high levels of positive ratio in wastewater samples in both phases, reaching peak values of 91.67% and 100% respectively. The lag time analysis results indicated that the peak positive ratio in all wastewater samples in Shangrao City appeared around 2 weeks later compared to the other three indicators. The correlation analysis revealed a strong linear correlation across all four types of data, with Spearman correlation coefficients ranging from 0.783 to 0.977, all of which were statistically significant.

Conclusion: The positive ratio of all wastewater samples in Shangrao City accurately reflected the COVID-19 epidemic trend from January to August 2023. This study confirmed the lag effect of wastewater percent positive and its strong correlation with the reported incidence rate and the positive ratio of nucleic acid samples in sentinel hospitals, supporting the use of wastewater percent positive monitoring as a supplementary tool for infectious disease surveillance in the regions with limited resources.

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Peer review under responsibility of KeAi Communications Co., Ltd.

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https://doi.org/10.1016/j.idm.2024.11.001

1. Introduction

COVID-19 is an infectious disease caused by the SARS-CoV-2 virus. Studies have shown that SARS-CoV-2 RNA can be detected in various biological specimens from cases (Makhmalbaf et al., 2022; Wang, Xu, et al., 2020; Zhang et al., 2020), such as respiratory nasopharyngeal swabs, fecal samples, urine samples. Moreover, SARS-CoV-2 can infect gastrointestinal cells (Xiao et al., 2020), prompting gastrointestinal symptoms like diarrhea and vomiting (Cheung et al., 2020; Mackulak et al., 2021; Mao et al., 2020; Wang, Hu, et al., 2020). This highlights the potential for conducting wastewater epidemiological research on the SARS-CoV-2 virus. Wastewater based epidemiology, an emerging field, investigates the transmission of infectious disease by analyzing markers in wastewater, and has swiftly advanced during the COVID-19 epidemic.

Research on the detection of SARS-CoV-2 RNA in feces has shown that infected individuals can excrete pathogens through feces even before the onset of symptoms (Tang et al., 2020). Wastewater studies conducted in Italy and Brazil (Fongaro et al., 2021; La et al., 2021) indicated that SARS-CoV-2 was already spreading in those countries before the first reported cases. A wastewater monitoring study in on-campus dormitory areas (Karthikeyan et al., 2021) found that around 85% of individuals produced positive wastewater samples before their clinical diagnosis. Therefore, utilizing wastewater monitoring as an auxiliary epidemic monitoring tool appears feasible, offering early warning signals compared to clinical testing. Additionally, since many asymptomatic or mildly symptomatic individuals may not seek medical care, but can still excrete SARS-CoV-2 through feces (Park et al., 2021), wastewater monitoring may have higher sensitivity. In April 2020, Wastewater-Based Epidemiology (WBE) was recommended as a detection method for tracking the novel coronavirus, and since then, wastewater-based monitoring of the COVID-19 pandemic has been widely implemented in many countries (Fahrenfeld et al., 2022; Fernandez-de-Mera et al., 2021; Tomasino et al., 2021; Westhaus et al., 2021). Numerous studies have confirmed that the concentration of SARS-CoV-2 in wastewater can reflect the scale of the COVID-19 outbreak in the study area. Existing research (Hillary et al., 2021; Karthikeyan et al., 2022; Lu et al., 2022; Rodriguez et al., 2022) shows the strong correlation between the quantitative monitoring data of SARS-CoV-2 viral RNA levels in wastewater and the number of cases, indicating that changes in virus concentration in wastewater reflect changes in the total number of cases in the study area.

To effectively manage the prevention and control of the epidemic after the implementation of "manage COVID-19 with measures against Class B infectious diseases", the Chinese government issued a policy on urban wastewater monitoring in January 2023. However, prevailing research (Aberi et al., 2021; Jeng et al., 2023; McMahan et al., 2021; Morvan et al., 2022; Phan et al., 2023) on infection scale analysis based on wastewater monitoring has focused on the quantitative detection of SARS-CoV-2 RNA copy number in wastewater, the high demand for facilities and economic resources for quantitative detection poses significant challenges to wastewater monitoring in limited-resource areas. During the epidemic, most health services in China were equipped with PCR instruments capable only of qualitative analysis, and to carry out quantitative analysis, each region would need to be equipped with a series of additional quantitative testing equipment and reagents. For example, the price of nucleic acid quantitative PCR instrument may range from 7000 dollars (ABI Quantstudio™ DX quantitative PCR instrument) to 230000 dollars (AutoMolec 3000 automatic nucleic acid extraction and fluorescence quantitative PCR instrument); Quantitative kits may also be more expensive due to processes such as reverse transcription. Adjusting wastewater monitoring methods suitable for resource-constrained areas provides an opportunity for monitoring the epidemic and responding to future large-scale infectious diseases.

This is the first wastewater monitoring study conducted in Shangrao city, which lacks the economic foundation for large-scale quantitative wastewater monitoring. The current study based in Shangrao analyzed the correlation between clinical testing or field investigation data (reflecting the scale of the epidemic) and the positive percentage of SARS-CoV-19 RNA in wastewater. It validated the feasibility of wastewater percent positive monitoring in assessing dynamic trends in COVID-19, and would provide valuable insights into wastewater surveillance in resource-limited settings.

2. Materials and methods

2.1. Sampling points and sample collection

This WBE monitoring study was conducted in Shangrao City, Jiangxi Province, China. Different wastewater sampling points were set up in 12 counties and districts and at the city level (the city level refers to large-scale facilities such as Shangrao Railway Station, Shangrao People's Hospital, Shangrao Wastewater Treatment Plant) of Shangrao in this study. The sampling point arrangement was divided into two phases. In the first phase, from January 20, 2023 (week 3) to April 24, 2023 (week 17), sampling points were established at the inlet of one wastewater treatment plant and the wastewater discharge outlet of one hospital in each designated area. In Guangxin District, lacking a wastewater treatment plant, sampling points were designated at the wastewater discharge outlets of one hospital and the wastewater pond of one scenic area. Subsequently, the second phase, from May 1, 2023 (week 18) to July 31, 2023 (week 31), involved the expansion of sampling locations by the Shangrao Center for Disease Control and Prevention. During this phase, the wastewater sampling points included the inlet of one wastewater treatment plant, the wastewater discharge outlet of one hospital, one school, three sentinel apartment complexes, and one station, and the wastewater pond of one market and one scenic area in each area. If a specific sampling location was unavailable in an area, no sampling point was established there. Sampling points were set up in two wastewater treatment plants of Wuyuan County due to the presence of two wastewater treatment plants and in two hospitals of Yanshan District due to its hospital-division. Additionally, due to coordination factors with relevant departments, there were fluctuations in sampling point distribution on May 1, May 8, and June 5. The types and quantities of sampling points are detailed in Fig. 1 and Supplementary Table.A1 and Supplementary Table.A2.

At each sampling point, wastewater samples were collected once a week on Mondays or Tuesdays. Due to sampling condition constraints, wastewater samples were collected on Friday of the third week, Tuesday and Sunday of the fifth week, and no sample was collected in the fourth and sixth weeks. The sampling method involved obtaining instantaneous wastewater samples by grabbing, that positioned a 500 ml sampling container at a water depth of 0.5 m at each point. Subsequently, a 500 ml wastewater sample was obtained at each sampling point. Following collection, the samples were promptly dispatched for laboratory testing, a process completed within 24 h.

2.2. Sample testing

The collected wastewater samples were concentrated and enriched by polyethylene glycol precipitation, then the SARS-CoV-2 nucleic acid was extracted by the Tianlong nucleic acid extraction kit, and finally the RT-PCR detection was performed in accordance with the operating instructions of the BioGerm nucleic acid detection kit. Detailed information about the operating procedures and quality control measures is provided in Appendix B.

2.3. Data Source of incidence information

The Shangrao Center for Disease Control and Prevention provided epidemiological data in the study area (number of reported cases, the positive ratio of nucleic acid samples in sentinel hospitals) and the incidence rate of influenza-like symptoms in students. The number of reported cases of each county and district in Shangrao was obtained from the weekly reports on the China Information System for Disease Control and Prevention (CISDCP). The positive ratio of nucleic acid samples in sentinel hospitals referred to the proportion of positive specimens among the samples collected from outpatient (emergency)-department influenza-like cases (body temperature ≥38 °C, accompanied by cough or sore throat) in 13 sentinel hospitals of Shangrao City every week. The incidence rate of flu-like symptoms in students referred to the proportion of flu-like symptoms detected through daily morning checks and monitoring of illness-related absence in the selected student population from 47 schools in Shangrao City, including the selection of 1 middle school, 1 primary school and 1 kindergarten selected from each region (12 counties and districts and 3 economic development zones or management committees), as well as the selection of 2 colleges. Additionally, the total population of Shangrao City in 2022, as provided by the statistical department, was 6.437 million.

2.4. Statistical analysis

The reported incidence rate (RIR) was calculated using the equation.

$$RIR = \frac{n}{N} \times 100\%$$

Where n represented the number of weekly reported cases, and N represented the total population of Shangrao City.

The various statistical data were standardized using Z-score normalization. The normalized index was calculated by the following formula:

$$Z_{i,j} = \frac{x_{i,j} - \overline{x}_j}{S_j}$$

Where $Z_{i,j}$ was the normalized index of sample *i* in data category *j*, $x_{i,j}$ was the value of sample *i* in data category *j*, $\overline{x}_j$ and S_j were the mean and standard deviation of data category *j*. This standardization process ensured comparability between different data categories.

Lagged time analysis was conducted using the xcorr function in MATLAB, the analysis index was based on the calculation of normalized cross-correlation (NCC), and the calculation formula was as follows:

$$NCC(X, Y) = \frac{Cov(\mathrm{X}, Y)}{sqrt(Var(\mathrm{X}) * Var(\mathrm{Y}))}$$

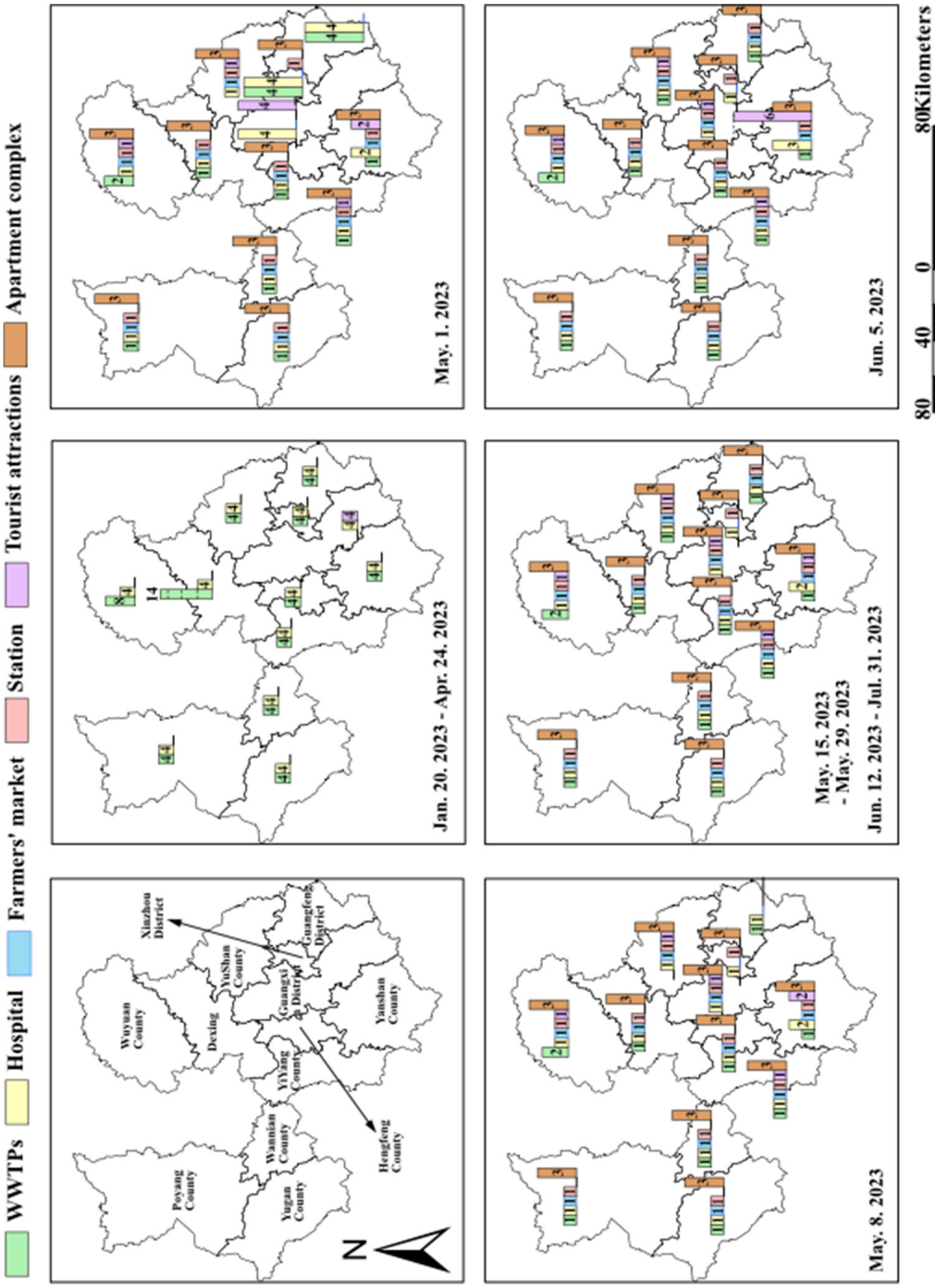

Fig. 1. Distribution Map of sampling points in Shangrao city.

Where *NCC(X,Y)* quantified the similarity between two sets of data *X* and data *Y*; Cov*(X,Y)* was the covariance of these two sets of data, and the denominator in the formula involved the variances of both sets, which were multiplied and then squared. NCC served as a key measure to gauge the correlation between different variables. A negative NCC indicated a negative correlation, a value of zero implied no correlation, and a positive NCC suggested a positive correlation. Larger absolute NCC values denoted greater similarity between the variables.

For further analysis, we selected the data with the highest NCC values for each statistical indicator, as well as the data from one week prior and one week subsequent to these values, for the normality test. A p-value >0.1 was considered indicative of a normal distribution. The results of the normality tests are detailed in Supplementary Table.A3. Due to the small sample size for each statistical indicator and the condition that not all sets of data conformed to a normal distribution, we used the Spearman correlation coefficient to assess the correlation, and the calculation formula was as follows:

$$r_s(X,Y) = c^2 = \frac{1}{n-1}\sum\left(\frac{X_i - \overline{X}}{S_X}\right)\left(\frac{Y_i - \overline{Y}}{S_Y}\right)$$

Where $r_s(X,Y)$ was the Spearman correlation coefficient for the two sets of data (X and Y), X_i and Y_i were the values of sample i in the two sets of data (X and Y), $\overline{X}$, $\overline{Y}$, and S_X, and S_Y were the means and standard deviations of the two sets of data (X and Y), respectively. A p-value <0.05 was considered statistically significant. On a statistically significant basis, the bigger the absolute value of r_s, the stronger the correlation.

Data entry and organization were performed using Excel 2020 software. Statistical analysis was conducted using MATLAB R2022a and IBM SPSS Statistics 26. Data plotting was accomplished using EXCEL 2020 and ArcMap 10.8 software.

3. Results

Wastewater monitoring in Shangrao City, Jiangxi Province, commenced on January 20, 2023 (Week 3) and continued until July 31, 2023 (Week 31). A total of 2797 wastewater samples were collected through 28 grabbing events, representing a population of 6.437 million in Shangrao City.

3.1. Analysis of wastewater monitoring results in 12 counties and districts of Shangrao City

The results of the first phase (Weeks 3–17) of wastewater monitoring are depicted in Fig. 2A. Throughout this monitoring period, the positive ratio of wastewater samples in each county and district of Shangrao City exhibited a gradual decrease. Wuyuan County, Poyang County, and Hengfeng County consistently recorded the highest positive ratios, while Yanshan District, Xinzhou District, and Guangxin District exhibited the lowest positive ratios. Wuyuan County maintained a consistently high positive ratio of wastewater samples from Week 3 to Week 12, ranging from a minimum of 66.67% to a maximum of 91.67%. From Weeks 13–17, excluding Week 14 with a positive ratio of 8.33%, all other weeks reported a 0% positive ratio. Poyang County displayed a positive ratio of wastewater samples of ≥75% from Week 3 to Week 8, 25.00% in Week 9 and Week 10, and 0% for the subsequent seven weeks. Hengfeng County reported a positive ratio of 75.00% in Week 3 and Week 12, a low positive ratio (≤37.50) in Weeks 7, 10, and 14 to 16, and moderate positive ratios for the remaining five weeks, mainly at 50.00% and 62.50%. Yanshan District consistently reported a 0% positive ratio for wastewater samples, Xinzhou District exhibited a positive ratio of 25.00% only in Week 11, and 0% for the rest of the weeks, while Guangxin District reported a positive ratio of 12.5% only in Week 3 and 0% for the other weeks.

The results of the second phase (Weeks 18–31) of wastewater monitoring are presented in Fig. 2B. During this monitoring phase, the positive ratio of wastewater samples in each county and district of Shangrao City gradually increased and then decreased. The peak positive ratio in each county and district generally occurred between Week 21 and Week 23. Yanshan District, Hengfeng County, Poyang County, Yushan District, and Wannian County maintained a low positive ratio (≤37.5%) throughout the second phase of monitoring. Guangfeng District, Guangxin District, and Yiyang County exhibited moderate positive ratios during the peak period (more than 37.50% and less than or equal to 62.50%) and low positive ratios for the remaining time. The positive ratio distribution in these three counties during the moderate level was outlined as follows: Guangfeng District reported positive ratios ranging from 42.86% to 57.14% during Weeks 19–22, Guangxin District reported a positive ratio of 42.86% in Week 22, and Yiyang County reported a positive ratio of 50.00% in Weeks 19, 22, and 23.

Moreover, the counties and districts with a high positive ratio (more than 62.50%) during the peak period were Xinzhou District, Yugan County, Wuyuan County, and Dexing City. The changes in the positive ratio of wastewater samples in these areas were detailed as follows: Xinzhou District reported a positive ratio of 80.00% from Week 20 to Week 22, a moderate level in Weeks 19, 24, and 25, and a low level for the rest of the time; Yugan County reported a positive ratio of 71.43% in Weeks 22 and 23, 28.57% in Week 21, and 0% for the remaining time; Wuyuan County reported a positive ratio of 66.67% in Weeks 21 and 22, 100% in Week 23, 44.44% in Week 24, 55.56% in Week 25, and a low level for the rest of the time; Dexing City reported a high positive ratio of 71.43% in Weeks 21 and 22, a moderate level in Weeks 19 and 20, and a low level for the rest of the time.

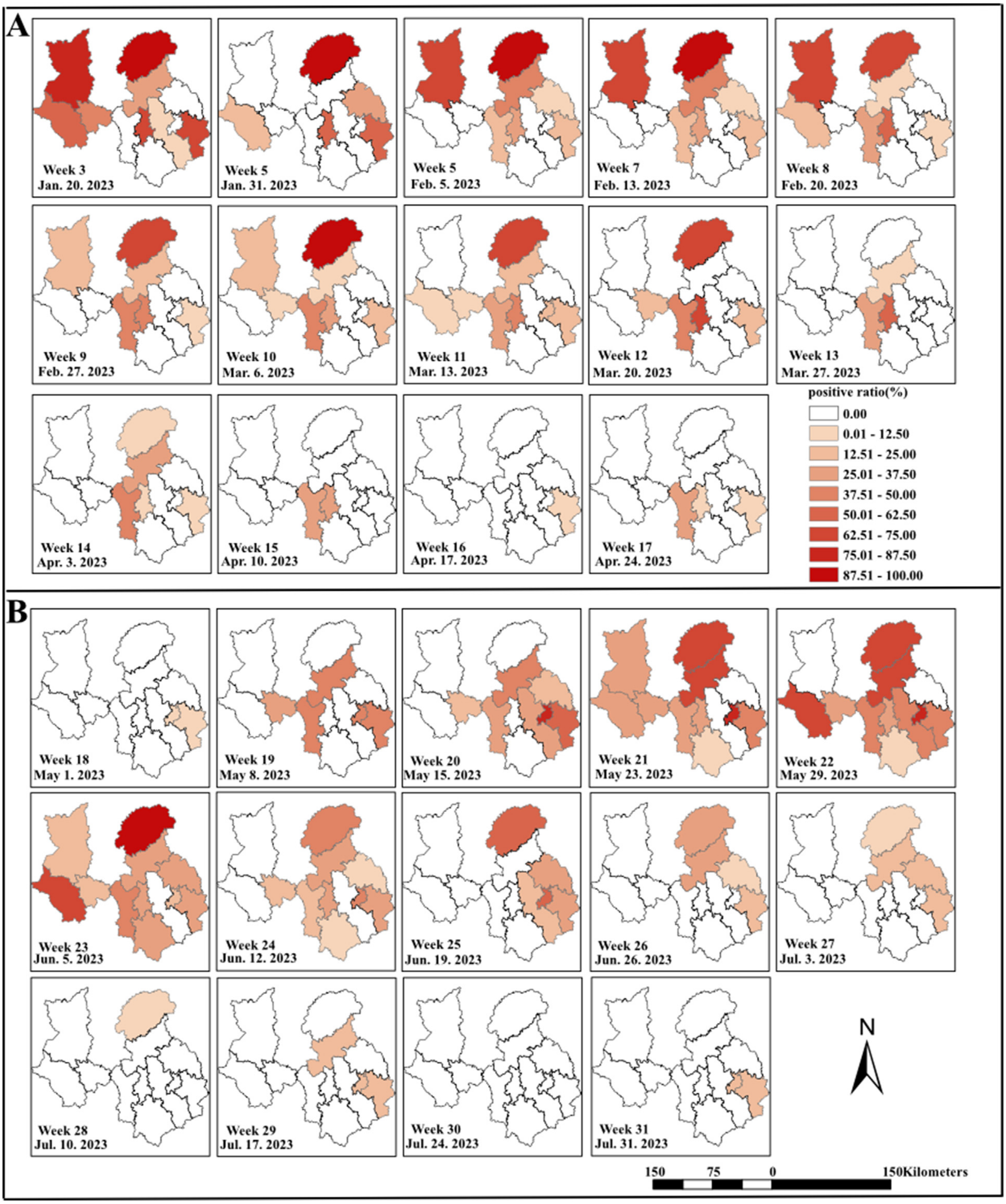

Fig. 2. The results of wastewater monitoring in each county and district of Shangrao City.

3.2. Long-term wastewater monitoring can identify the epidemic trend in Shangrao City

Since the wastewater monitoring was not carried out timely before every Tuesday from the 3rd week to the 6th week of the study, we presented separate descriptions for the positive ratio of all wastewater samples in Shangrao, the positive ratio of wastewater samples at the city level, the reported incidence rate, the positive ratio of nucleic acid samples in sentinel hospitals, and the incidence rate of influenza-like symptoms in students during these four weeks, as illustrated in Fig. 3A. The results of these data from the 7th week to the 31st week are presented in B. In both the early and late stages of the study, investigations of student populations were hindered by school holidays, resulting in available statistics on the incidence rate of influenza-like symptoms in students only from Week 7 to Week 26. Our primary analysis focused on various statistical indicators collected from Week 7 to Week 31.

Overall, compared to the results of city-level wastewater monitoring, the positive ratio of all wastewater samples in Shangrao provided a more accurate indication of the changing trend of the COVID-19 epidemic in Shangrao City. During the wastewater monitoring period from Week 7 to Week 17 in the first phase, the city experienced a declining phase of the epidemic wave, with the reported incidence rate and the positive ratio of nucleic acid samples in sentinel hospitals gradually decreasing at a low level. The incidence rate of influenza-like symptoms in students exhibited significant fluctuations, reaching a peak level from the 9th week to the 13th week (February to March), potentially linked to the influenza epidemic during this period. And the positive ratio of all wastewater samples in Shangrao also demonstrated a gradual decline. In the second phase (from the 18th week to the 31st week) of wastewater monitoring, the trends of the four statistical indicators gradually increased to peak values and then decreased. Notably, the peak value of the positive ratio of all wastewater samples in Shangrao occurred approximately 2 weeks later than the other three statistical indicators.

3.3. Correlation analysis of statistical indicators

The study undertook a correlation analysis between the positive ratio of all wastewater samples in Shangrao, the reported incidence rate, the positive ratio of nucleic acid samples in sentinel hospitals, and the incidence rate of influenza-like symptoms in students during the second phase of wastewater monitoring. Given the substantial disparities in the numerical values of these indicators, initial data processing utilized the z-score normalization method. The analysis results of lag-time are shown in Fig. 4A. The results of the NCC analysis indicated that the positive ratio of all wastewater samples in Shangrao had a lag effect. Specifically, changes in the wastewater indicator lagged the reported incidence rate by two weeks, lagged the positive ratio of nucleic acid samples in sentinel hospitals by one week, and lagged the rate of influenza-like symptoms in students by three weeks. Conversely, the reported incidence rate demonstrated synchronicity with the positive ratio of nucleic acid samples in sentinel hospitals and the incidence rate of influenza-like symptoms in students. Notably, the positive ratio of nucleic acid samples in sentinel hospitals lagged the incidence rate of influenza-like symptoms in students by one week.

Spearman correlation analysis results indicated optimal correlation when the positive ratio of wastewater samples lagged reported cases and the positive ratio of nucleic acid samples in sentinel hospitals by two weeks, and lagged the incidence rate of influenza-like symptoms in students by three weeks. Additionally, reported cases demonstrated synchronicity with the positive ratio of nucleic acid samples in sentinel hospitals and the incidence rate of influenza-like symptoms in students, and the positive ratio of nucleic acid samples in sentinel hospitals lagged the incidence rate of influenza-like symptoms in students by one week. Fig. 4B and Supplementary Table.A4 provide a comprehensive presentation of these results. The study highlighted a robust linear correlation among all four statistical indicators, as evidenced by Spearman correlation coefficients ranging from 0.783 to 0.977, all of which were statistically significant.

4. Discussion

This study with the qualitative testing for SARS-CoV-2 in 2797 wastewater samples obtained from Shangrao City, Jiangxi Province, demonstrated the feasibility and utility of Shangrao's wastewater monitoring for SARS-CoV-2 RNA. In this study, the cost of the testing reagents used to analyze whether COVID-19 is present in the wastewater samples was approximately $1.40 (comprising 0.28 dollars for the Tianlong nucleic acid extraction kit and 1.13 dollars for the BioGerm nucleic acid detection kit). In contrast, single sample testing can cost between $6 and $18 for quantitative testing (Ma et al., 2022). Therefore, the low-cost qualitative testing provided new insights into wastewater surveillance in resource-constrained settings.

Following the refinement and standardization of sampling sites in the second phase, the variations in Shangrao's positive ratio of all wastewater samples did not exhibit significant differences compared to the first phase which was at a low level of the epidemic. This lack of distinction may be attributed to the predominant placement of sampling sites in the first phase at wastewater treatment plants and hospitals, where larger populations or a higher proportion of key populations often resulted in reaching the positive limit for wastewater testing more readily. Overall, our findings suggest that wastewater monitoring accurately reflects the epidemic trend in Shangrao City, and the enrichment and standardized processing of wastewater sampling sites enhance the ability to indicate the local epidemic level more effectively. The positive ratio of all wastewater samples in Shangrao exhibited a gradual decrease from January to April, followed by an upward trend in May, and ultimately decreased again from June to August. Notably, sustained elevated levels of wastewater sample positive ratio were observed in Wuyuan County during two sampling periods. On December 7, 2022, China concluded the zero-dynamic policy, discontinued

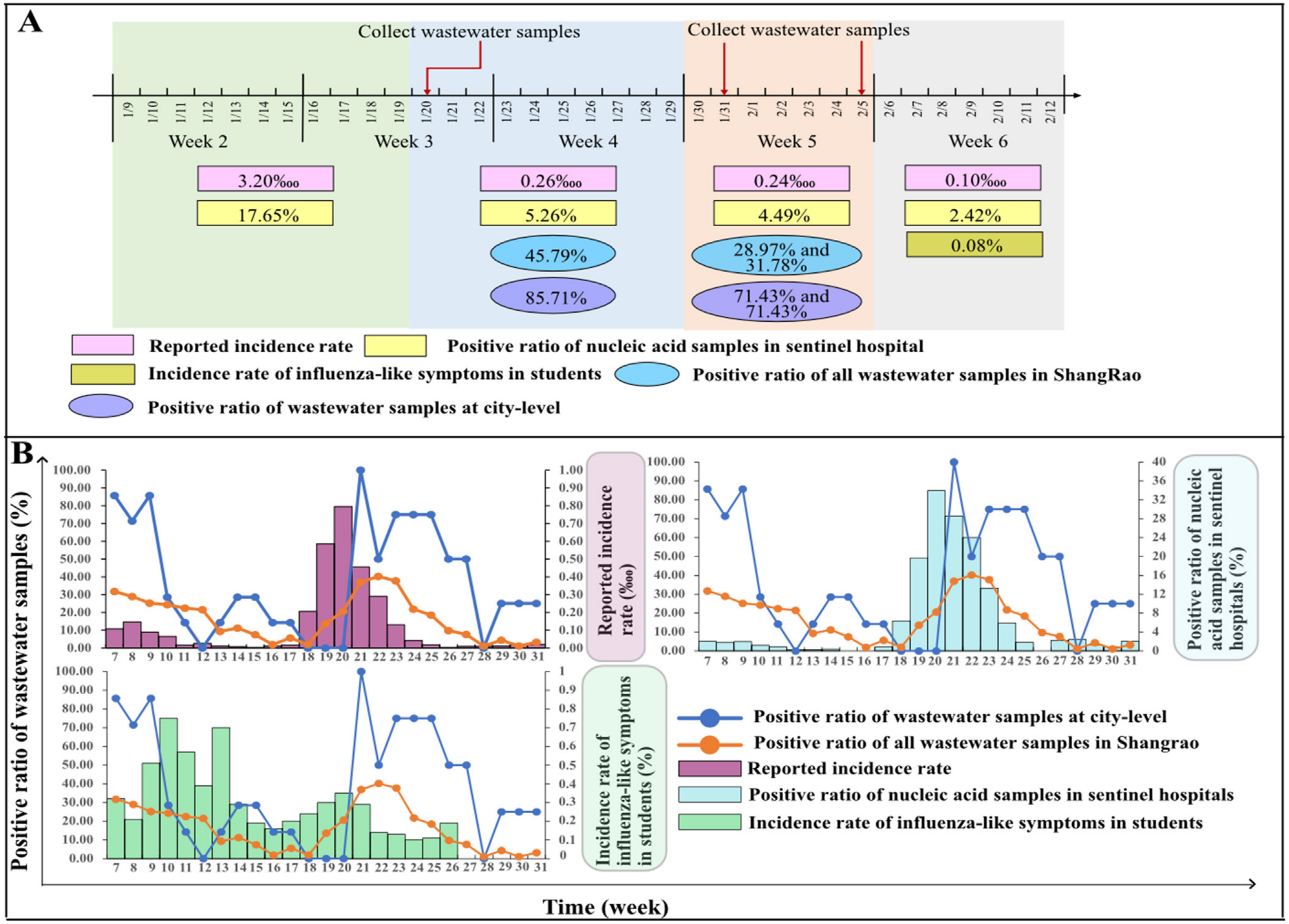

Fig. 3. Comparison of various monitoring indicators from Week 3 to Week 31 in Shangrao City.

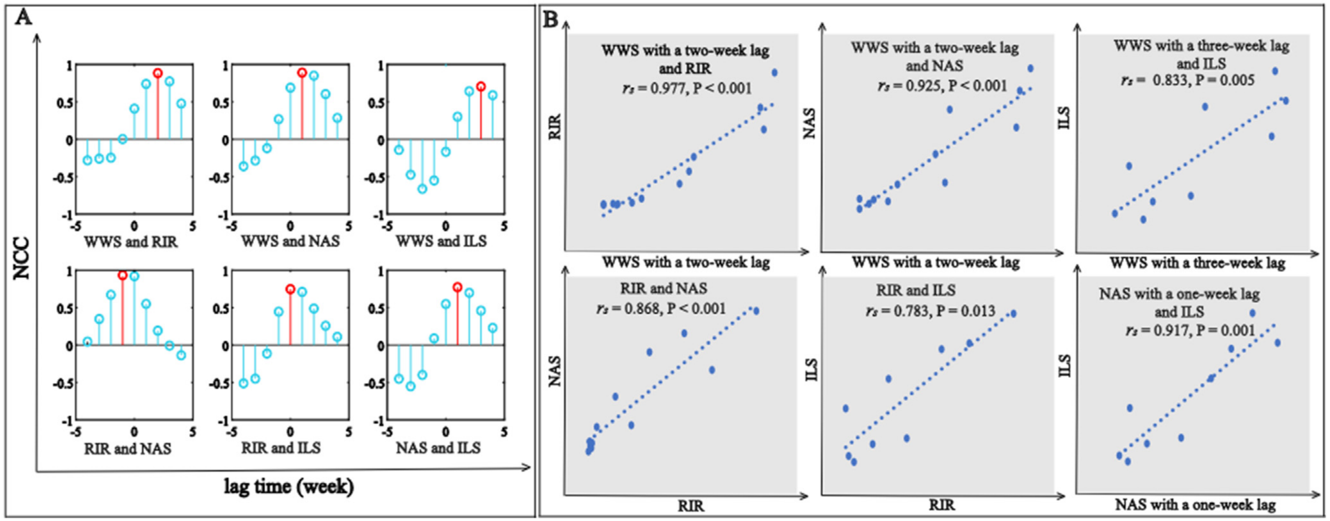

Fig. 4. Results of lag time and correlation analysis for various monitoring indicators.

isolation measures for COVID-19 patients, and adjusted testing strategies to "testing all who are willing to be tested." Consequently, there was a sharp increase in COVID-19 cases. On January 8, 2023, China managed COVID-19 with measures against Class B infectious diseases, instead of Class A infectious diseases, in a major shift of its epidemic response policies. These policy changes may account for the relatively high incidence of COVID-19 infections in Shangrao City during the early stages. Following this period, many residents in Shangrao City entered a recovery phase or achieved complete recovery with immunity.

Moreover, as a prominent tourist destination in China, Shangrao City witnessed significant population migration during the May Day holiday, involving large-scale and widespread population movement. This dynamic might have heightened the risk of COVID-19 transmission. These factors might have contributed to the large-scale detection of positive wastewater samples and a gradual decrease in the positive ratio in Shangrao City during the first phase of wastewater monitoring, the positive ratio of wastewater samples in Shangrao City gradually increased and the coverage of positive samples gradually expanded in the early second phase of wastewater monitoring (May). Additionally, factors such as Wuyuan County's favorable geographical environment and thriving tourism industry, coupled with its proximity to other popular tourist areas like Huangshan City in Anhui Province, Kaihua County in Zhejiang Province, and Jingdezhen City in Jiangxi Province, could have contributed to an elevated risk of COVID-19 transmission in Wuyuan County.

A primary objective of this study was to assess the viability of wastewater monitoring as a supplementary tool for early detection of the COVID-19 epidemic. To achieve this, lag time analysis was conducted, and Spearman correlation coefficients were calculated for the weekly standardized positive ratio of all wastewater samples in Shangrao, reported incidence rate, positive ratio of nucleic acid samples in sentinel hospitals, and incidence rate of influenza-like symptoms in students. The findings indicated a lag in wastewater data compared to the other indicators (Peccia et al., 2020; Weidhaas et al., 2021). Both analyses revealed that wastewater data lagged the reported incidence rate by 2 weeks, lagged the positive ratio of nucleic acid samples in sentinel hospitals by 1–2 weeks, and lagged the incidence rate of influenza-like symptoms in students by 3 weeks. The disparity in lag time for wastewater data and the positive ratio of nucleic acid samples in sentinel hospitals between the two analysis methods might be attributed to two factors: firstly, the Spearman correlation coefficient analysis was sensitive to changes in the number of research data as lag time varies, and secondly, the actual lag time between wastewater data and the positive ratio of nucleic acid samples in sentinel hospitals would be between one and two weeks, but our study analyzed lag time using integer weeks, which might have resulted in divergent outcomes due to the distinct principles of the two analysis methods.

We inferred that in theory, the reported incidence rate should precede the positive ratio of nucleic acid samples in sentinel hospitals by 0–1 week and lag the incidence rate of influenza-like symptoms in students by 1 week. Moreover, the positive ratio of nucleic acid samples in sentinel hospitals should lag the incidence rate of influenza-like symptoms in students by 1–2 weeks. Nevertheless, our research results showed that the incidence rate of influenza-like symptoms in students was synchronized with the reported incidence rate, and only preceded the positive ratio of nucleic acid samples in sentinel hospitals by one week. The reason for this discrepancy may be related to the limited data number of the incidence rate of influenza-like symptoms in students in the later stage of the study when schools were closed (making it impossible to collect this data). Among all the data, wastewater data demonstrated the highest correlation with the reported incidence rate and the positive ratio of nucleic acid samples in sentinel hospitals, with Spearman correlation coefficients exceeding 0.92, and the Spearman correlation coefficient between the reported incidence rate and the positive ratio of nucleic acid samples in sentinel hospitals reached 0.868. Consequently, although qualitative wastewater data was acknowledged as a lagging indicator, the study suggested that qualitative wastewater monitoring remained a valuable early warning tool for monitoring the epidemic situation in the study area. An increasing trend in wastewater positive ratio could indicate the gradual expansion of the COVID-19 epidemic, prompting the timely implementation of prevention and control measures. Furthermore, the study also substantiated the feasibility of inferring infectious disease prevalence based on sentinel hospital monitoring data (Das et al., 2021; May et al., 2024; Mazagatos et al., 2022).

Considering that the wastewater samples in the study were mainly collected during the initial days of the early week (Monday or Tuesday), combined with the results of the correlation analysis, it was suggested that, in contrasted to the positive nucleic acid ratio observed in samples from sentinel hospitals, wastewater data might exhibit a lag of 0–1 week. Conversely, when compared to the officially reported incidence rate, the wastewater data trends manifested a lag of 1–2 weeks. This observation aligned with findings from a quantitative wastewater monitoring study in the United States (Fahrenfeld et al., 2022). However, conflicting perspectives exist among scholars, some assert that wastewater data can proactively predict epidemic scale (Bibby et al., 2021; Galani et al., 2022; Wu et al., 2021). Several reasons contribute to wastewater data being a lagging indicator: (1) throat swabs yield earlier positive results than fecal samples, approximately 5 days earlier (Chen et al., 2020); (2) COVID-19 patients can excrete the virus in feces for 20–30 days post-symptom onset (Miura et al., 2021), contributing to prolonged signals in wastewater; (3) Feng's study (Feng et al., 2021) emphasizes the impact of sampling frequency on correlation analysis results, and this should be taken into consideration when analyzing the results, as weekly wastewater sampling may be too infrequent, which is also a limitation of this study; (4) following the easing of epidemic control measures, the pace of case detection and reporting by infected individuals may decelerate, influencing analysis outcomes; (5) one study (Zhao et al., 2023) has shown that the highest concentration of SARS-CoV-2 RNA in feces may also occur 2 days after the onset of symptoms; (6) The efficiency of clinical testing in the later stage is accelerated, which may weaken the leading effect of sewage in the early research to a certain extent.

This study has the following limitations. Firstly, the sampling method used in this study was grabbing. Although scholars who have conducted quantitative studies on the number of COVID-19 viral copies have indicated that the accuracy of grab samples may only differ by a few logarithmic units from composite samples (Kyle et al., 2021), the drawbacks of grabbing, such as poorer sample representativeness and the inability to capture virus signals in wastewater when the incidence rate of the disease to be monitored in the study area is very low, should also be considered. Secondly, sample collection in Shangrao City's counties was not uniformly executed by a single institution, but independently carried out by each county, potentially leading to variations in testing conditions and result accuracy among counties. Thirdly, for hospital and case data it was a percentage of the population that was positive, it might artificially deflate the value if there were a lot of asymptomatic people. Lastly, due to research limitations, this study was unable to conduct a quantitative testing of COVID-19 viral copies in wastewater samples. Consequently, only qualitative testing could be performed, limiting the ability to predict the scale of infection in the study area based on wastewater data. Nevertheless, the high correlation between wastewater percent positive and other data indicated that wastewater percent positive effectively reflected the changing trends of monitored diseases in the study area. Therefore, wastewater percent positive monitoring could serve as a supplementary tool for comprehending the characteristics of the COVID-19 epidemic, issuing early warning signals, and assisting decision-makers in managing the epidemic at the local or regional level. This provided valuable insights for regions without the resources to conduct wastewater percent positive monitoring research.

5. Conclusion

The positive ratio of all wastewater samples in Shangrao accurately reflected the epidemic trends of COVID-19 in Shangrao City. And the results of wastewater monitoring showed that Wuyuan County had the highest risk of COVID-19 transmission among the 13 counties and districts in Shangrao city and could be identified as a key area for monitoring COVID-19 or other infectious diseases that are prone to spreading among the entire population. In addition, both the reported incidence rate and the positive ratio of nucleic acid samples in sentinel hospitals had the strong correlation with wastewater data, this further confirmed the feasibility and scientific validity of conducting infectious disease surveillance through sentinel hospitals in the past. It also supported the notion that wastewater percent positive monitoring could be used as a supplementary tool for infectious disease surveillance in the regions with limited resources.

CRediT authorship contribution statement

Jing Wang: Writing – review & editing, Writing – original draft, Visualization, Methodology, Formal analysis. **Haifeng Zhou:** Writing – original draft, Validation, Investigation, Formal analysis, Conceptualization. **Wentao Song:** Writing – original draft, Validation, Methodology, Conceptualization. **Lingzhen Xu:** Visualization, Resources, Investigation, Data curation. **Yaoying Zheng:** Resources, Investigation, Data curation. **Chen You:** Resources, Investigation, Data curation. **Xiangyou Zhang:** Resources, Investigation, Data curation. **Yeshan Peng:** Resources, Investigation, Data curation. **Xiaolan Wang:** Supervision, Conceptualization. **Tianmu Chen:** Writing – review & editing, Project administration, Conceptualization.

Ethics approval and consent to participate

This effort of investigation was part of CDC's routine responsibility in Shangrao City, China. Therefore, institutional review and informed consent were not required for this study. All data analysed were anonymised.

Consent for publication

Not applicable.

Availability of data and materials

The data that support the findings of this study are available from Shangrao Center for Disease Control and Prevention, and these data are published in the local epidemic surveillance report. Data can also be obtained directly from the authors with the permission of Dr. Xu (Email: srcdczyws@126.com).

Funding

This study was supported by the Guangzhou Laboratory Project (SRPG22-007), the Guangzhou Laboratory Project (GZNL2024A01004) and the National Key Research and Development Program of China (2021YFC2301604). The funder had no role in study design, data collection, data analysis, data interpretation, or writing of the report.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

Not applicable.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.idm.2024.11.001.

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