



Remiero

Wastewater-Based Epidemiology for Cost-Effective Mass Surveillance of COVID-19 in Low- and Middle-Income Countries: Challenges and Opportunities

Sadhana Shrestha ^{1,2,*}, Emi Yoshinaga ¹, Saroj K. Chapagain ¹, Geetha Mohan ^{1,2}, Alexandros Gasparatos ^{1,2} and Kensuke Fukushi ^{1,2,*}

- Institute for the Advanced Study of Sustainability, United Nations University, 5 Chome-53-70 Jingumae, Shibuya City, Tokyo 150-8925, Japan; yoshinaga@unu.edu (E.Y.); saroj@unu.edu (S.K.C.); geetha.mohan@unu.edu (G.M.); gasparatos@g.ecc.u-tokyo.ac.jp (A.G.)
- ² Institute for Future Initiatives, The University of Tokyo, 7-3-1 Hongo, Bunkyo-ku, Tokyo 113-0033, Japan
- * Correspondence: sadhana.shrestha@unu.edu or gosadhana@gmail.com (S.S.); kensuke-fukushi@unu.edu (K.F.)

Abstract: Wastewater-based epidemiology (WBE) is an approach that can be used to estimate COVID-19 prevalence in the population by detecting severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) RNA in wastewater. As the WBE approach uses pooled samples from the study population, it is an inexpensive and non-invasive mass surveillance method compared to individual testing. Thus, it offers a good complement in low- and middle-income countries (LMICs) facing high costs of testing or social stigmatization, and it has a huge potential to monitor SARS-CoV-2 and its variants to curb the global COVID-19 pandemic. The aim of this review is to systematize the current evidence about the application of the WBE approach in mass surveillance of COVID-19 infection in LMICs, as well as its future potential. Among other parameters, population size contributing the fecal input to wastewater is an important parameter for COVID-19 prevalence estimation. It is easier to back-calculate COVID-19 prevalence in the community with centralized wastewater systems, because there can be more accurate estimates about the size of contributing population in the catchment. However, centralized wastewater management systems are often of low quality (or even non-existent) in LMICs, which raises a major concern about the ability to implement the WBE approach. However, it is possible to mobilize the WBE approach, if large areas are divided into sub-areas, corresponding to the existing wastewater management systems. In addition, a strong coordination between stakeholders is required for estimating population size respective to wastewater management systems. Nevertheless, further international efforts should be leveraged to strengthen the sanitation infrastructures in LMICs, using the lessons gathered from the current COVID-19 pandemic to be prepared for future pandemics.

Keywords: mass surveillance; poor sanitation coverage; SARS-CoV-2 variants; prevalence of infection; wastewater management system; policy making



Citation: Shrestha, S.; Yoshinaga, E.; Chapagain, S.K.; Mohan, G.; Gasparatos, A.; Fukushi, K. Wastewater-Based Epidemiology for Cost-Effective Mass Surveillance of COVID-19 in Low- and Middle-Income Countries: Challenges and Opportunities. *Water* 2021, 13, 2897. https://doi.org/10.3390/w13202897

Academic Editors: Masaaki Kitajima, Eiji Haramoto and Abasiofiok Mark Ibekwe

Received: 16 August 2021 Accepted: 12 October 2021 Published: 15 October 2021

Publisher's Note: MDPI stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.



Copyright: © 2021 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https://creativecommons.org/licenses/by/4.0/).

1. Background

Wastewater-based epidemiology (WBE) is a new and rapidly developing field for identifying and quantifying endogenous and exogenous chemical and biological markers in wastewater, and then this information is utilized to estimate in real-time certain quantitative indicators about public health in general population (e.g., prevalence of disease, proportion of population using illicit drugs) [1]. WBE has been identified as a rare bridge between the environmental surveillance (ES) of wastewater and the health information of the society [2]. So far, it has been used extensively to understand the patterns and trends of illicit drug use in communities [3–10], becoming a convenient new complementary method to more complex and lengthy survey approaches [4].

Water 2021, 13, 2897 2 of 14

Like chemical markers, biological markers such as DNA/RNA of bacteria, viruses, and fungi can also be detected through ES in wastewater, helping to identify human sources and provide information about disease outbreaks in communities [1]. The ES of biological markers in sewage and wastewater has been conducted worldwide, whenever outbreaks have occurred in the past. Some examples include studies on H1N1 sub-types of influenza A virus in sewage during influenza outbreaks [11]. Moreover, ES has been used to investigate the presence of enteropathogenic viruses in sewage (e.g., noroviruses and rotavirus) [12-14]. Most ES studies in sewage have compared the findings about pathogen data with health surveillance data [12]. The Global Polio Eradication Initiatives initiated by World Health Organization (WHO) have combined ES (wastewater was examined for polioviruses) with acute flaccid paralysis surveillance data in several countries for many years, aiming to identify the areas of increased risk as well as formulating or amending vaccination strategies [15]. However, a limited number of studies have translated the wastewater concentration of pathogens to the prevalence of disease in the population (proportion of the population infected by the disease at a specific time). In one of such examples, researchers used the norovirus GII concentration in sewage and the concentration of the virus shed by an infected individual in feces to estimate the number of infected individuals [12].

Since the onset of the global pandemic of the coronavirus disease 2019 (COVID-19), ES studies have been conducted in numerous countries to detect the etiological agent of the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) RNA in wastewater [16–19]. The most common practice for linking virus data from ES with health data was by establishing a relationship between SARS-CoV-2 concentration in wastewater and the prevalence data of COVID-19 reported by public health agencies and hospitals. For example, a study has compared a SARS-CoV-2 detection event in wastewater treatment plant (WWTP) samples with reported disease cases in the corresponding catchment in Japan and concluded that the virus RNA was detected when the disease cases were at the peak [17]. Moreover, the SARS-CoV-2 virus concentrations from ES have been significantly correlated with: (a) COVID-19 prevalence in the Netherlands [19]; (b) the available number of COVID-19-positive individuals in Israel [20]; and (c) local hospital admissions and the COVID-19 epidemiological curve in the USA [21]. Only a handful of studies that have translated the wastewater concentration of SARS-CoV-2 into the prevalence of COVID-19 in population [16,22–25] that can be applicable for public health decision making.

Such studies have estimated the COVID-19 prevalence in the general population utilizing the total number of SARS-CoV-2 RNA copies per liter of wastewater, viral RNA copies per gram shed by an infected individual, fecal load per person per day, wastewater flow per day, and population served by the WWTPs. Some studies have calculated the point estimates of the prevalence [22,23], whereas others provided the probability distribution and the range of number of the infected individuals [16,24,25]. While it is possible to calculate the COVID-19 prevalence, there are large uncertainties during the process. Some of the reasons for uncertainties are the limited data available on virus shedding (particularly by pre- and asymptomatic individuals), the fact that viral yield varies depending upon the selection of virus concentration and detection methods, especially for the variants, and the lack of appropriate methods of population normalization [26]. Certainly, there is a need to improve the components used for the prevalence estimation. Nevertheless, these back calculation methods have been proved to be beneficial for tackling the conditions of drug abuse [3] or estimating the prevalence of noroviruses in the general population [12]. Hence, the WBE approach can be used to complement health surveillance using prevalence estimates to guide decisions about resource allocation to contain the disease spreading. In addition, the detection of SARS-CoV2 RNA in wastewater samples prior to reported clinical cases of COVID-19 [19] indicates that wastewater surveillance can serve for an early identification of infectious diseases in the general population.

With global cases of the COVID-19 pandemic having reached more than 200 million by 15 August 2021, the pandemic has severely disrupted social and economic systems

Water 2021, 13, 2897 3 of 14

worldwide. Socioeconomic impacts of COVID-19 are harsher in lower- and middle-income countries (LMICs) [27], which had already faced many development challenges before the pandemic. Some of the more significant impacts of COVID-19 in LMICs include the loss of income, food insecurity, inability to access medicine, and loss of access to education [28]. Even though the vaccination against COVID-19 has been rolled out in at least 217 countries and territories [29], the emergence of new SARS-CoV-2 variants of concern (VOCs), such as B.1.1.7 (Alpha variant), B.1.351 (Beta variant), B.1.617.2 (Delta variant), and P.1 (Gamma variant) and their rapid spread worldwide further complicate the control of the COVID-19 pandemic. For example, by the time of writing this paper, the Alpha variant had been identified in 163 countries, the Beta variant had been identified in 105 countries, the Delta variant had been identified in 115 countries, and the Gamma variant had been identified in 66 countries [30]. Because of their higher transmission rate, higher disease severity, higher reinfection rate, reduction of therapy effectiveness, and reduction in the neutralization efficacy of sera from convalescent patients or vaccines [31,32], it is the utmost important issue for countries to be able to conduct the real-time surveillance and monitoring of emerging VOCs at the community level to control the pandemic.

Already in some developed nations, there have been recommendations for the systematic inclusion of wastewater surveillance in the national testing strategies for the detection of SARS-CoV-2 and its variants [33]. The WBE approach, in addition, has a good potential for estimating prevalence and tracking infections clusters and trends in general population, as well as hot spots in large cities or areas, hence being an inexpensive approach to mass surveillance [22,34,35]. However, almost all the studies have used the WBE approach to estimating the COVID-19 prevalence in high-income nations such as USA [22,23], France [36], Australia [16], and Qatar [25]. WBE studies in low-income countries are very scanty. One of the few exceptions is a study in India [24] that has successfully implemented this approach utilizing WWTP samples and estimated the COVID-19 prevalence in a large urban center, Hyderabad. However, studies that have included onsite wastewater treatment systems (OWTSs), which are predominant wastewater management systems in LMICs, are rare.

This review paper aims to systematize the current evidence about the application of the WBE approach in LMICs, including the ways to incorporate predominant and diverse wastewater management systems and to conduct the mass surveillance of COVID-19 infections. We discuss the benefits the WBE approach has and the challenges to mobilize it effectively in the context of LMICs. This paper focuses on wastewater surveillance in diverse wastewater management systems existing in LMICs coupled with the possibility of translating the obtained SARS-CoV-2 concentration data into the prevalence estimation/trend identification/status of infection in the general population. We provide a series of policy and practice recommendations to execute the WBE approach that can contribute positively in the ongoing efforts in LMICs to control the COVID-19 pandemic.

2. Potential of WBE for COVID-19 Mass Surveillance in LMICs

WBE can play an important role in efforts to identify the status of spread of COVID-19 and VOCs in LMICs as a complementary approach to health surveillance. The WHO strongly recommends testing COVID-19 suspect cases and individuals who are in close contact to avoid further infections [37]. However, the recommended testing protocol requires costly kits and equipment, trained technicians, and laboratories of at least a biosafety level 2 [38,39]. There are fewer laboratory facilities and trained workforce that could meet the identification of viral infections following the WHO guidelines in LMICs [40]. The establishment of fully equipped laboratories requires a huge investment, which poses substantial economic burden in such resource-constrained countries [41]. Besides, the essential increment in medical supplies and personal protective equipment will also likely increase significant costs and the associated financial burden [41]. However, limiting the number of tests will consequently increase the probability of the infection spread. Conversely, the WBE approach uses pooled/composite samples. In fact, multiple samples must be collected for the successful implementation of the WBE approach. For example, Centers for

Water 2021, 13, 2897 4 of 14

Disease Control and Prevention [42] has advised for frequent wastewater sample collection (a minimum of three samples in a certain period), to track trends (development or change) of infection over time. Although the cost for reagents is identical for clinical testing and WBE testing (a mean of 15 USD per test kit) [43], the cost per sample for WBE testing is higher due to concentration steps. Nevertheless, WBE is more cost-effective than clinical testing, because WBE can essentially test thousands of people through one sample whereas clinical testing requires thousands of individual tests for a given area. Therefore, WBE is one viable method to conduct mass surveillance that will be economical for resource-poor settings [43].

The economic advantages of WBE can become even more important when considering the possible spread of VOCs in LMICs. Up to the writing of this paper, there have been relatively few LMICs reporting VOC spread, compared to in high-income countries. However, there have been sporadic findings about mutations in SARS-CoV-2 in wastewater samples from LMICs [44], which show that the spread of VOCs in these countries is happening and might be underreported. A possible underreporting of VOCs in LMICs is of major public health concern, given the effects of these variants on vaccine efficacy [31]. Most of the LMICs are anticipating the start of widespread vaccination campaigns soon, but the rapid spread of SARS-CoV-2 variants could adversely impact the outcomes of vaccination campaigns. Although the real-time surveillance of VOCs has been already recommended for some countries [33], the technology involved for next-generation sequencing (NGS) is prohibitively expensive, cumbersome, time-consuming and requires additional specialized infrastructures and standardized protocols [45] and high technical expertise for data interpretation. Thus, individual testing could further escalate the significant costs of managing the pandemic in already resource-constrained LMICs. In this sense, the WBE approach that uses pooled samples from the study population can become a strong complementary approach to the identification of the SARS-CoV-2 variants circulating in the general population.

Many unknowns and uncertainties surrounding the COVID-19 pandemic have given rise to widespread fears, fueling in many cases stereotyping and stigmatization [46]. The stigmatization of COVID-19 patients, survivors, and frontline health workers [47–49] can lead to mental health problems and be counterproductive for curbing pandemics and attaining equitable development [50]. Stigma can also prevent the access of vulnerable groups to healthcare and social services, leading to social exclusion, discrimination, psychological distress, and violence in LMICs [51–53]. By being a non-invasive method [2], the WBE preserves the personal identity of patients by creating overall population infection profiles. Thus, it can offer a lot of potential in social contexts where stigmatization against COVID-19 patients and their families is prevalent. Therefore, the WBE approach could prove to be an effective mass screening approach to dealing with social stigma during the pandemic.

Asymptomatic COVID-19 infection has been reported to range from 18% to 31% [54–56], which are unaccounted by health surveillance except in the case of contact tracing. A recent study in clinical testing data to determine the abundance of asymptomatic versus symptomatic cases revealed that 79.2% of the COVID-19-positive individuals were asymptomatic [57]. However, many symptomatic and asymptomatic people shed the virus in stool that can be detected in wastewater. WBE embraces both symptomatic and asymptomatic groups.

3. Challenges in Mobilizing WBE for COVID-19 Mass Surveillance in LMICs

Wastewater production and collection are key aspects for ensuring the representativeness of WBE exercises [58]. The implementation of the WBE approach in a given area is much easier in the context with centralized wastewater management systems, which gather wastewater from many users through sewerage networks and treat it at one or several WWTPs [58]. The population size of a WWTP catchment is generally known for centralized wastewater management systems which can be used swiftly to obtain quantitative health information for the study population through back-calculation. Water 2021, 13, 2897 5 of 14

Wastewater management systems in LMICs are generally poor. LMICs contain the majority of the 2.4 billion people that still lack access to improved sanitation worldwide [59]. Moreover, over 95% of the wastewater produced in LMICs is being discharged untreated in the environment [60], polluting water bodies with chemicals and viruses [61]. While centralized wastewater management systems cover 91% of the population in most advanced economies [60,62], their coverage is much lower in many LMICs. Often, they cover only portions of larger urban areas and are often not planned for smaller towns and densely populated areas, low-income urban areas, and rural areas [63,64] (Figure 1). One of the reasons for not planning centralized wastewater management systems in these areas is the high expenditure associated with sewage collection, which often accounts for 60% of the total budget allocated for waste management [65,66].

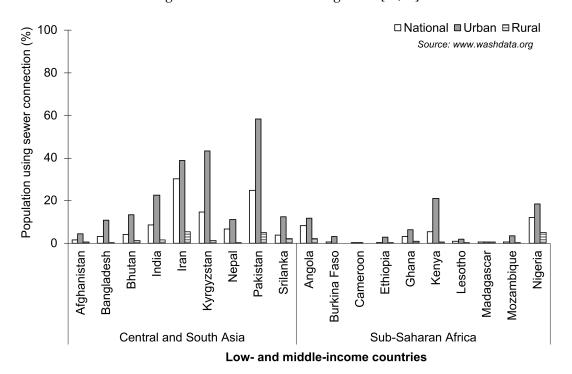


Figure 1. Populations with sewer connection in selected low- and middle-income countries (LMICs) [64].

Discouragingly, cities with centralized wastewater management systems often have low sewerage connection rates [67], sometimes lower than 10% of the population in many LMICs (Figure 1) [64]. In most cities and rural areas of many LMICs, individual households are not connected to sewerage systems and rely mostly on OWTSs [60,64] (Figure 2). Such systems consist of infrastructures in which excreta are stored or treated where generated, e.g., pit latrines and septic tanks [68]. This complies with the WHO/UNICEF definition of improved sanitation.

While the SARS-CoV-2 RNA virus concentration and detection methods in wastewater are evaluated as feasible enough to be performed in most of the microbiology and environmental engineering laboratories in LMICs [69], the major bottleneck of the WBE application lies in existing wastewater management systems. As mentioned in the previous section, the lack of central wastewater management systems in the majority of areas in LMICs prevents the easy application of the WBE approach in estimating COVID-19 prevalence in the catchment [70], which is otherwise being simply estimated in developed nations, for example in Australia [16]. In addition, poor sewerage connection rates, sewer oversizing, and leaks are some other problems [58], which affect the ability to estimate properly wastewater flow in WWTPs ultimately leading to the faulty back-calculation of COVID-19 infection status in the study population. Furthermore, widely employed OWTSs representing individual houses further complicate WBE applications in LMICs.

Water 2021, 13, 2897 6 of 14

Sampling each OWTS is labor-intensive, expensive and has strong unethical ramifications considering the sensitivity of the obtained information.

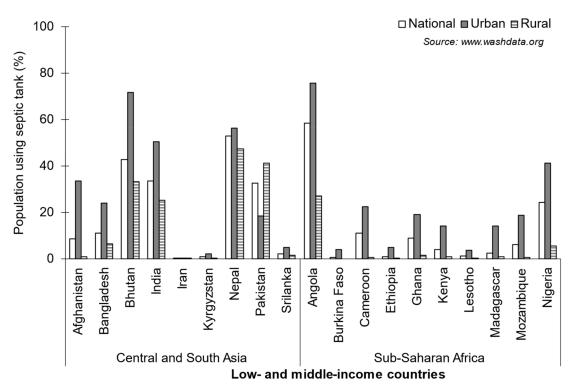


Figure 2. Populations using septic tanks in selected LMICs [64].

4. Mobilizing the WBE Approach for COVID-19 Mass Surveillance in LMICs

Despite the challenges outlined in the previous section, it is still possible to mobilize the WBE approach for COVID-19 applications in LMICs. Considering the issues related to centralized wastewater management systems outlined above, this would most likely entail downscaled applications from metropolitan cities/municipalities to small cities, towns, smaller communities, specific drainage areas, residences, schools, and other institutions, according to the need of the locality (Figure 3). An ES study conducted in a populous city of India taking wastewater samples from WWTPs [71] advocated for the WBE application in LMICs starting from larger cities. Below, we expand on some of the possible WBE application strategies to cover most of areas in LMICs.

A major problem in the WBE application in context with low sewerage connection rates is the uncertainty of the population size of the WWTPs catchment. In such a case, one possible approach would be to use population biomarkers to quantify the contributing population size in real time. Possible population biomarkers that are naturally excreted by humans in wastewater [72,73] include creatinine [74], cholesterol, coprostanol [72], nicotine [75], cortisol, androstenedione, and the serotonin metabolite 5-hydroxyndoleacetic acid (5-HIAA) [76]. Therefore, with some additional chemical analysis, population data could be obtained and used for the mass surveillance of COVID-19 infection. In the areas provided with completed centralized wastewater management systems, samples can be taken at the inlets of WWTPs. Moreover, areas with a sewer network but without WWTPs are also very common in LMICs where wastewater is being disposed directly to water bodies such as rivers. In such a case, wastewater samples can be collected from the outlets to these water bodies or from manholes (Figure 3) [44]. Here, municipalities/city offices/local governments should be coordinated and mobilized to delineate areas, identify wastewater sampling points and estimate population size corresponding to each wastewater outlet.

Water 2021, 13, 2897 7 of 14

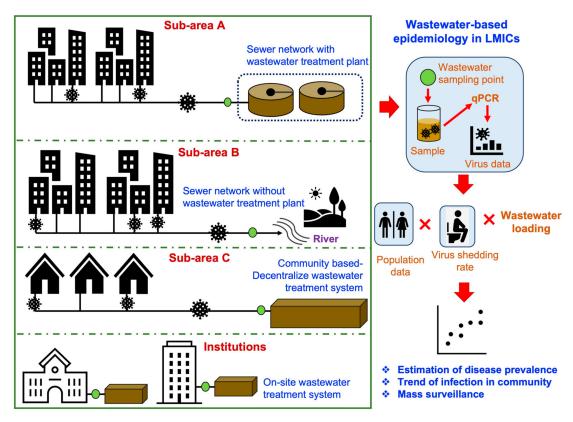


Figure 3. Strategies for mobilizing the wastewater-based epidemiology (WBE) approach for the mass surveillance of COVID-19 in LMICs.

In areas not served by centralized wastewater management systems, community-managed decentralized wastewater treatment systems (DEWATS) are increasingly used. Such DEWATS employ cluster systems in scattered and low-density urban communities and rural areas [60]. Community-managed DEWATS connected to simplified sewer systems or communal sanitation centers have the potential to close the gap between OWTSs and centralized wastewater management systems [63], with their prevalence growing in LMICs due to cost-effectiveness [65], e.g., in Indonesia [63], Nepal [77], India [78], Bangladesh [79], and South Africa [80]. Wastewater samples from community-managed DEWATS (Figure 3) which has a known population size would increase the feasibility of the WBE approach for COVID-19 applications.

The fact remains that large portions of urban areas and rural areas in LMICs still rely on OWTS, such as septic tanks (Figure 2). Hence, excluding households with OWTSs will neglect a large portion of urban and rural population, prohibiting a complete picture of disease spread. However, the population with OWTSs can be considered by randomly sampling a representative number of households (Figure 3). The aim of such an approach is not to identify individual households with COVID-19 cases, but to understand the status and trends of COVID-19 infection in the targeted population. Social stigma is a major concern where individual household can be unintentionally identified while applying WBE. Therefore, municipalities/city offices/local governments should be coordinated and mobilized to make local communities understand the purpose and importance of the WBE application for the mass surveillance of COVID-19 and to seek prior approval from the community. Separately, institutions that use OWTSs, such as schools, colleges, and offices, can use the WBE approach, as the population size is almost constant and known. However, the number of people visiting hospitals and health centers is largely undefined, and the population contributing to these institutions' OWTSs fluctuates. Hence, including these types of institutions are less useful for estimating the COVID-19 prevalence in the

Water 2021, 13, 2897 8 of 14

study area. Nevertheless, the ES of wastewater from hospitals with a known number of COVID-19 cases could provide crucial information, e.g., for model development.

Samplings in the cases outlined above should comply the CDC guidelines [41]. In case of OWTSs, composite samples serve as a representative sample which can be obtained by combining portions of multiple grab samples manually [81]. For example, samples from septic tanks should be grab samples of a sludge from multiple chambers and locations within the tanks and should be combined to generate representative composite samples [82].

To summarize, despite the possible challenges, the application of the WBE approach for the mass surveillance of COVID-19 could be possible in LMICs. This would require segregating larger areas into sub-areas corresponding to wastewater management systems (Figure 3), collecting representative wastewater samples separately and estimating the population size of the catchment of wastewater sampling point correctly.

5. Policy and Practice Recommendations

Instead of testing individuals through health surveillance, WBE can be applied as a complementary approach which is cost-effective and especially suitable for mass surveillance in resource-poor country and regions [43]. WBE is an essential tool for detecting the re-emergence of COVID-19 and to early warn future outbreaks [26]. The WBE approach provides a prevalence or trend of COVID-19 infection in the general population which is usually underreported in health surveillance. However, as discussed in the previous sections, the poor or a non-existent central wastewater management system is the major bottleneck of the WBE application for the mass surveillance of COVID-19 in LMICs. At the current expansion rates of central wastewater treatment systems, most people in Asia and Africa will still not experience safely managed sanitation by 2050 [83]. Therefore, we recommend applying the WBE approach in LMICs by considering the existing situations of diverse wastewater management systems to curb current progression.

For applying the WBE approach in LMICs, large areas, such as municipalities, can be divided into smaller sub-areas or individual communities corresponding to the existing wastewater management systems. Subsequently, wastewater samples can be collected and analyzed for each sub-area. At first, the COVID-19 prevalence in several smaller catchments can be estimated and then combined to obtain the single prevalence of the city or the municipality.

Wastewater is a pooled sample, and its analysis reduces the economic burden compared to testing individuals. Although SARS-CoV-2 detection methods in wastewater are reported to be feasible in LMICs [69] and this review paper does not discuss the technological aspect of virus detection, testing still can entail high cost in LMICs. In particular, setting up an appropriate laboratory (e.g., BSL-2 laboratories) and procuring necessary equipment for molecular analysis (e.g., NGS and variant-specific qPCR) can be very costly and pose challenges in LMIC settings.

In order to overcome this financial problem, relevant stakeholders in LMICs should plan to implement the WBE approach initially in larger cities, because larger cities have ended up becoming the main hotspots of COVID-19 infections as they are characterized by higher population densities, extensive economic activities, and complexity of human mobility patterns [84]. Subsequently, wastewater surveillance should be institutionalized as municipal or prefectural testing strategies for the virus detection. Public or private academic and research institutions and profit-making clinical and environmental laboratories could become valuable collaborators in such efforts. For example, local governments can outsource wastewater surveillance to public or private environmental and clinical laboratories. LMICs should also seek and utilize fundings that are usually supported by international donors, for example resources that are made available for rapid quality COVID-19 tests [85]. Before planning wastewater sampling in an area, sewage lines (sewer networks) should be mapped, and the population size of the catchment should be known [44]. In addition, sub-areas with community-managed DEWATS and individual

Water **2021**, 13, 2897 9 of 14

OWTS should be mapped, and the respective population sizes should be estimated. Such a mapping can be performed in collaboration and coordination with stakeholders, for example water supply and sewerage system authorities, city development committee, public works and human settlement authorities, and local community leaders [86]. At the same time, environmental health experts should be mobilized to translate the virus data into public health estimates, while public health experts/authorities should utilize such data for public health actions. The overall coordination of above described strategies could be handled by entities such as health departments, home affairs departments, presidential taskforces on COVID-19, and committees for COVID-19 prevention and control [86].

The national governments of LMICs should take initiatives in forming a multidisciplinary team of scientists embracing environmental engineering and public health expertise to act extensively against growing COVID-19 infections. A nationwide wastewater surveil-lance campaign is advised to better understand the temporal and spatial dynamics of COVID-19 disease prevalence [87]. In addition, a combined national and international collective effort, such as "COVID-19 WBE Collaborative" consisting of scientists from multiple disciplines that aims to facilitate timely and high-impact WBE studies for public benefit [88], could also aid in increasing the applicability of WBE in LMICSs. Nevertheless, the international community should leverage further international efforts to help governments strengthen and hasten the accomplishment of sanitation and health infrastructures in LMICs, using the lessons gathered from the current COVID-19 pandemic to be prepared for future epidemics.

6. Conclusions

WBE is a wastewater surveillance approach, which can be used for quantitative surveillance of SARS-CoV-2 in an informal converging wastewater network in large cities in LMICs as an initial step. Many studies have shown that the quantitative detection of SARS-CoV-2 in ES over time correlates well with reported COVID-19 cases and mortality. In fact, several studies have shown that ES can serve as an earlier indicator as well. The quantitative assays can show temporal-spatial trends to identify hotspots of transmission. All of the information will be helpful for public health stakeholders even in the absence of the prevalence data. Preforming clinical testing for mass surveillance puts huge financial burden on LMICs, because WHO recommended testing protocols are costly to implement. In addition, the recent recommendation of the real-time surveillance of VOCs that need prohibitively expensive NGS technology is less affordable by LMICs. Furthermore, WBE uses a pooled sample, and hence, the non-invasive method could prove to be an effective to deal with social stigma during the pandemic. While useful for LMICs for the costeffective mass surveillance of COVID-19, the WBE application is challenging because a centralized wastewater management system is a key for this approach but is poor or non-existent in the majority of the countries. The strategy discussed here for mobilizing WBE in LMICs is downscaling its application from metropolitan cities or municipalities to small cities, towns, smaller communities, specific drainage areas etc., according to the existing wastewater management systems. For example, a large area can be divided into small sub-areas: (a) areas with a sewerage network with WWTPs; (b) areas with a sewerage network discharged into environmental water bodies; (c) areas with community-managed DEWATS; (d) areas with individual OWTS such as septic tanks; (e) institutions with an approximately constant population size such as schools and offices. At the early stage of the WBE implementation, large cities should be focused, instead of implementing it as a national strategy. We suggest that fostering institutional collaboration could further help reduce the financial burden. In more detail, considering both the current boom in clinical testing facilities and the expanding wastewater surveillance research in LMICs, local governments could alternatively opt to outsource sample collection and analysis to other relevant public and private academic and research institutions. Before planning for wastewater sampling, the mapping of sewage lines, areas with community-managed DEWATS, and individual OWTS should be prepared in coordination with city wastewater

Water 2021, 13, 2897 10 of 14

management authorities, city development committees, etc. Relevant authorities should be coordinated to estimate the population size in the respective catchments. After the translation of virus data to public health estimates by environmental scientists, public health experts should be collaborated to utilize the data for making public health decisions. To devise this strategy of implementing the WBE approach, health departments and national or regional COVID-19 taskforces could provide an overall coordination between the stakeholders. Further, collective efforts of national and international scientists could increase the applicability of WBE in LMICs.

Author Contributions: S.S., conceptualization, writing original draft, art design, and corresponding author; E.Y., G.M. and S.K.C., editing and reviewing; A.G., editing, reviewing, and funding; K.F., reviewing, funding, and corresponding author. All authors have read and agreed to the published version of the manuscript.

Funding: This research was supported by the Japan Society for the Promotion of Science (JSPS) and Japan Science and Technology Agency (JST).

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: The data for Figures 1 and 2 have been obtained from Joint Monitoring Programme for Water Supply, Sanitation and Hygiene—Estimates on the Use of Water, Sanitation, and Hygiene by country (2000–2020) that can be found in then link: https://data.unicef.org/topic/water-and-sanitation/covid-19/ (Accessed on 25 March 2020).

Acknowledgments: We would like to thank Bikash Malla, a postdoctoral researcher in University of Yamanashi, Japan, who is currently working in different aspect of SARS-CoV-2 detection in wastewater, for sharing his views on the applicability of WBE in LMICs and overall issues. We would like to thank Japan Society for the Promotion of Science (JSPS) for the financial support. A.G. acknowledges the support of the Japan Science and Technology Agency (JST) for project SASSI (JPMJBF18T3).

Conflicts of Interest: The authors declare no conflict of interest.

References

- Sims, N.; Kasprzyk-Hordern, B. Future Perspectives of Wastewater-Based Epidemiology: Monitoring Infectious Disease Spread and Resistance to the Community Level. *Environ. Int.* 2020, 139, 105689. [CrossRef]
- 2. Daughton, C.G.; Jones-Lepp, T.L. (Eds.) *Pharmaceuticals and Care Products in the Environment: Scientific and Regulatory Issues*; ACS Symposium Series; American Chemical Society: Washington, DC, USA, 2001; Volume 791, ISBN 978-0-8412-3739-1.
- 3. Zuccato, E.; Chiabrando, C.; Castiglioni, S.; Calamari, D.; Bagnati, R.; Schiarea, S.; Fanelli, R. Cocaine in Surface Waters: A New Evidence-Based Tool to Monitor Community Drug Abuse. *Environ. Health* **2005**, *4*, 14. [CrossRef]
- 4. Zuccato, E.; Chiabrando, C.; Castiglioni, S.; Bagnati, R.; Fanelli, R. Estimating Community Drug Abuse by Wastewater Analysis. *Environ. Health Perspect.* **2008**, *116*, 1027–1032. [CrossRef]
- 5. Andrés-Costa, M.J.; Rubio-López, N.; Morales Suárez-Varela, M.; Pico, Y. Occurrence and Removal of Drugs of Abuse in Wastewater Treatment Plants of Valencia (Spain). *Environ. Pollut.* **2014**, 194, 152–162. [CrossRef] [PubMed]
- 6. Daglioglu, N.; Guzel, E.Y.; Kilercioglu, S. Assessment of Illicit Drugs in Wastewater and Estimation of Drugs of Abuse in Adana Province, Turkey. *Forensic Sci. Int.* **2019**, 294, 132–139. [CrossRef] [PubMed]
- 7. Bijlsma, L.; Botero-Coy, A.M.; Rincón, R.J.; Peñuela, G.A.; Hernández, F. Estimation of Illicit Drug Use in the Main Cities of Colombia by Means of Urban Wastewater Analysis. *Sci. Total Environ.* **2016**, *565*, 984–993. [CrossRef] [PubMed]
- 8. Archer, E.; Castrignanò, E.; Kasprzyk-Hordern, B.; Wolfaardt, G.M. Wastewater-Based Epidemiology and Enantiomeric Profiling for Drugs of Abuse in South African Wastewaters. *Sci. Total Environ.* **2018**, *625*, 792–800. [CrossRef] [PubMed]
- Foppe, K.S.; Hammond-Weinberger, D.R.; Subedi, B. Estimation of the Consumption of Illicit Drugs during Special Events in Two Communities in Western Kentucky, USA Using Sewage Epidemiology. Sci. Total Environ. 2018, 633, 249–256. [CrossRef] [PubMed]
- 10. Lai, F.Y.; O'Brien, J.; Bruno, R.; Hall, W.; Prichard, J.; Kirkbride, P.; Gartner, C.; Thai, P.; Carter, S.; Lloyd, B.; et al. Spatial Variations in the Consumption of Illicit Stimulant Drugs across Australia: A Nationwide Application of Wastewater-Based Epidemiology. *Sci. Total Environ.* 2016, 568, 810–818. [CrossRef] [PubMed]
- 11. Heijnen, L.; Medema, G. Surveillance of Influenza A and the Pandemic Influenza A (H1N1) 2009 in Sewage and Surface Water in the Netherlands. *J. Water Health* **2011**, *9*, 434–442. [CrossRef] [PubMed]

Water 2021, 13, 2897 11 of 14

12. Hellmér, M.; Paxéus, N.; Magnius, L.; Enache, L.; Arnholm, B.; Johansson, A.; Bergström, T.; Norder, H. Detection of Pathogenic Viruses in Sewage Provided Early Warnings of Hepatitis A Virus and Norovirus Outbreaks. *Appl. Environ. Microbiol.* **2014**, *80*, 6771–6781. [CrossRef] [PubMed]

- 13. Sano, D.; Fukushi, K.; Yoshida, Y.; Omura, T. Detection of Enteric Viruses in Municipal Sewage Sludge by a Combination of the Enzymatic Virus Elution Metho Dan d RT-PCR. *Water Res.* **2003**, *37*, 3490–3498. [CrossRef]
- 14. Sano, D.; Watanabe, T.; Matsuo, T.; Omura, T. Detection of Infectious Pathogenic Viruses in Water and Wastewater Samples from Urbanised Areas. *Water Sci. Technol.* **2004**, *50*, 247–251. [CrossRef] [PubMed]
- 15. Asghar, H.; Diop, O.M.; Weldegebriel, G.; Malik, F.; Shetty, S.; El Bassioni, L.; Akande, A.O.; Al Maamoun, E.; Zaidi, S.; Adeniji, A.J.; et al. Environmental Surveillance for Polioviruses in the Global Polio Eradication Initiative. *J. Infect. Dis.* **2014**, 210, S294–S303. [CrossRef] [PubMed]
- 16. Ahmed, W.; Angel, N.; Edson, J.; Bibby, K.; Bivins, A.; O'Brien, J.W.; Choi, P.M.; Kitajima, M.; Simpson, S.L.; Li, J.; et al. First Confirmed Detection of SARS-CoV-2 in Untreated Wastewater in Australia: A Proof of Concept for the Wastewater Surveillance of COVID-19 in the Community. Sci. Total Environ. 2020, 728, 138764. [CrossRef] [PubMed]
- 17. Haramoto, E.; Malla, B.; Thakali, O.; Kitajima, M. First Environmental Surveillance for the Presence of SARS-CoV-2 RNA in Wastewater and River Water in Japan. *Sci. Total Environ.* **2020**, *737*, 140405. [CrossRef]
- La Rosa, G.; Iaconelli, M.; Mancini, P.; Bonanno Ferraro, G.; Veneri, C.; Bonadonna, L.; Lucentini, L.; Suffredini, E. First Detection of SARS-CoV-2 in Untreated Wastewaters in Italy. Sci. Total Environ. 2020, 736, 139652. [CrossRef] [PubMed]
- 19. Medema, G.; Heijnen, L.; Elsinga, G.; Italiaander, R.; Brouwer, A. Presence of SARS-Coronavirus-2 RNA in Sewage and Correlation with Reported COVID-19 Prevalence in the Early Stage of the Epidemic in The Netherlands. *Environ. Sci. Technol. Lett.* **2020**, 7, 511–516. [CrossRef]
- 20. Bar-Or, I.; Yaniv, K.; Shagan, M.; Ozer, E.; Erster, O.; Mendelson, E.; Mannasse, B.; Shirazi, R.; Kramarsky-Winter, E.; Nir, O.; et al. Regressing SARS-CoV-2 Sewage Measurements onto COVID-19 Burden in the Population: A Proof-of-Concept for Quantitative Environmental Surveillance. *medRxiv* 2020. [CrossRef]
- Peccia, J.; Zulli, A.; Brackney, D.E.; Grubaugh, N.D.; Kaplan, E.H.; Casanovas-Massana, A.; Ko, A.I.; Malik, A.A.; Wang, D.; Wang, M.; et al. SARS-CoV-2 RNA Concentrations in Primary Municipal Sewage Sludge as a Leading Indicator of COVID-19 Outbreak Dynamics. medRxiv 2020. [CrossRef]
- 22. Wu, F.; Zhang, J.; Xiao, A.; Gu, X.; Lee, W.L.; Armas, F.; Kauffman, K.; Hanage, W.; Matus, M.; Ghaeli, N.; et al. SARS-CoV-2 Titers in Wastewater Are Higher than Expected from Clinically Confirmed Cases. *mySystems* **2020**, *5*, e00614-20. [CrossRef] [PubMed]
- 23. Weidhaas, J.; Aanderud, Z.; Roper, D.; Van Derslice, J.; Gaddis, E.; Ostermiller, J.; Hoffman, K.; Jamal, R.; Heck, P.; Zhang, Y.; et al. Correlation of SARS-CoV-2 RNA in wastewater with COVID-19 disease burden in sewersheds. *Sci. Total Environ.* **2021**, 775, 145790. [CrossRef] [PubMed]
- 24. Hemalatha, M.; Kiran, U.; Kumar, K.S.; Kopperi, H.; Gokulan, C.G.; Mohan, S.V.; Mishra, R.K. Comprehensive surveil- lance of SARS-CoV-2 spread using wastewater-based epidemiology studies. *medRxiv* **2020**. [CrossRef]
- 25. Saththasivam, J.; El-Malah, S.S.; Gomez, T.A.; Jabbar, K.A.; Remanan, R.; Krishnankutty, A.K.; Ogunbiyi, O.; Rasool, K.; Ashhab, S.; Rashkeev, S.; et al. COVID-19 (SARS-CoV-2) outbreak monitoring using wastewater-based epidemiology in Qatar. *Sci. Total Environ.* 2021, 774, 145608. [CrossRef] [PubMed]
- 26. Medema, G.; Been, F.; Heijnen, L.; Petterson, S. Implementation of environmental surveillance for SARS-CoV-2 virus to support public health decisions: Opportunities and challenges. *Curr. Opin. Environ. Sci. Health* **2020**, *17*, 49–71. [CrossRef]
- 27. World's Most Vulnerable Countries Lack the Capacity to Respond to a Global Pandemic. Available online: https://www.un.org/ohrlls/news/world\T1\textquoterights-most-vulnerable-countries-lack-capacity-respond-global-pandemic-credit-mfdelyas-alwazir. (accessed on 18 April 2021).
- 28. Josephson, A.; Kilic, T.; Michler, J.D. Socioeconomic Impacts of COVID-19 in Low-Income Countries. *Nat. Hum. Behav.* **2021**, *5*, 557–565. [CrossRef]
- 29. Tracking Covid-19 Vaccinations Worldwide. Available online: https://edition.cnn.com/interactive/2021/health/global-covid-vaccinations/ (accessed on 16 August 2021).
- 30. Global Lineage Reports. Available online: https://cov-lineages.org/index.html#global_reports (accessed on 16 August 2021).
- 31. Gómez, C.E.; Perdiguero, B.; Esteban, M. Emerging SARS-CoV-2 Variants and Impact in Global Vaccination Programs against SARS-CoV-2/COVID-19. *Vaccines* **2021**, *9*, 243. [CrossRef]
- 32. Harvey, W.T.; Carabelli, A.M.; Jackson, B.; Gupta, R.K.; Thomson, E.C.; Harrison, E.M.; Ludden, C.; Reeve, R.; Rambaut, A.; COVID-19 Genomics UK (COG-UK) Consortium; et al. SARS-CoV-2 Variants, Spike Mutations and Immune Escape. *Nat. Rev. Microbiol.* 2021, 19, 409–424. [CrossRef] [PubMed]
- 33. European Commission. Commission Recommendation on a Common Approach to Establish a Systematic Surveillance of SARS-CoV-2 and Its Variants in Wastewaters in the EU. *European Commission*. 2021. Available online: https://ec.europa.eu/environment/pdf/water/recommendation_covid19_monitoring_wastewaters.pdf (accessed on 20 March 2020).
- 34. Dharmadhikari, T.; Yadav, R.; Dastager, S.; Dharne, M. Translating SARS-CoV-2 Wastewater-Based Epidemiology for Prioritizing Mass Vaccination: A Strategic Overview. *Environ. Sci. Pollut. Res.* **2021**, *28*, 42975–42980. [CrossRef]
- 35. Larsen, D.A.; Wigginton, K.R. Tracking COVID-19 with Wastewater. Nat. Biotechnol. 2020, 38, 1151–1153. [CrossRef] [PubMed]

Water 2021, 13, 2897 12 of 14

36. Wurtzer, S.; Marechal, V.; Mouchel, J.M.; Maday, Y.; Teyssou, R.; Richard, E.; Almayrac, J.L.; Moulin, L. Evaluation of lockdown effect on SARS-CoV-2 dynamics through viral genome quantification in wastewater, Greater Paris, France, 5 March to 23 April 2020. Euro Surveill. 2020, 25, 50. [CrossRef]

- 37. World Health Organization. *Critical Preparedness, Readiness and Response Actions for COVID-19: Interim Guidance*; World Health Organization: Geneva, Switzerland, 2021; WHO reference number: WHO/2019-nCoV/Community_Actions/2021.1.
- 38. World Health Organization. *Laboratory Biosafety Guidance Related to Coronavirus Disease (COVID-19): Interim Guidance;* World Health Organization: Geneva, Switzerland, 2021; WHO reference number: WHO/WPE/GIH/2020.3.
- 39. World Health Organization. *Laboratory Testing for Coronavirus Disease* 2019 (COVID-19) in Suspected Human Cases; World Health Organization: Geneva, Switzerland, 2020; WHO reference number: WHO/COVID-19/laboratory/2020.4.
- 40. World Health Organization. *Regional Office for South-East Asia Guidelines on Establishment of Virology Laboratory in Developing Countries*; World Health Organization, Regional Office for South-East Asia: New Delhi, India, 2008; ISBN 978-92-9022-335-1.
- 41. Giri, A.K.; Rana, D.R. Charting the Challenges behind the Testing of COVID-19 in Developing Countries: Nepal as a Case Study. *Biosaf. Health* **2020**, *2*, 53–56. [CrossRef]
- 42. Center of Disease Control and Prevention. Developing A Wastewater Surveillance Sampling Strategy. 2020. Available online: https://www.cdc.gov/coronavirus/2019-ncov/cases-updates/wastewater-surveillance/developing-a-wastewater-surveillance-sampling-strategy.html (accessed on 20 March 2020).
- 43. Hart, O.E.; Halden, R.U. Computational Analysis of SARS-CoV-2/COVID-19 Surveillance by Wastewater-Based Epidemiology Locally and Globally: Feasibility, Economy, Opportunities and Challenges. *Sci. Total Environ.* **2020**, *730*, 138875. [CrossRef] [PubMed]
- 44. Napit, R.; Manandhar, P.; Chaudhary, A.; Shrestha, B.; Poudel, A.; Raut, R.; Pradhan, S.; Raut, S.; Mathema, S.; Rajbhandari, R.; et al. Rapid Genomic Surveillance of SARS-CoV-2 in a Dense Urban Community Using Environmental (Sewage) Samples. *medRxiv* 2021. [CrossRef]
- 45. Gwinn, M.; MacCannell, D.; Armstrong, G.L. Next-Generation Sequencing of Infectious Pathogens. *JAMA* **2019**, *321*, 893. [CrossRef] [PubMed]
- 46. UNICEF/WHO/IFRC. Social Stigma Associated with COVID-19-A Guide to Preventing and Addressing Social Stigma. 2020. Available online: https://www.who.int/publications/m/item/a-guide-to-preventing-and-addressing-social-stigma-associated-with-covid-19 (accessed on 10 April 2020).
- 47. Bagcchi, S. Stigma during the COVID-19 Pandemic. Lancet Infect. Dis. 2020, 20, 782. [CrossRef]
- 48. Taylor, L. Covid-19 Misinformation Sparks Threats and Violence against Doctors in Latin America. *BMJ* **2020**, *370*, m3088. [CrossRef]
- 49. UN. COVID-19 and Human Rights We Are All in This Together. 2020. Available online: https://data2.unhcr.org/en/documents/details/75828 (accessed on 10 April 2020).
- 50. Roelen, K.; Ackley, C.; Boyce, P.; Farina, N.; Ripoll, S. COVID-19 in LMICs: The Need to Place Stigma Front and Centre to Its Response. *Eur. J. Dev. Res.* **2020**, *32*, 1592–1612. [CrossRef] [PubMed]
- 51. Rohwerder, B. *Disability Stigma in Developing Countries*; K4D Helpdesk Report; Institute of Development Studies: Brighton, UK, 2018; Volume 26.
- 52. Stangl, A.L.; Earnshaw, V.A.; Logie, C.H.; van Brakel, W.; Simbayi, L.C.; Barré, I.; Dovidio, J.F. The Health Stigma and Discrimination Framework: A Global, Crosscutting Framework to Inform Research, Intervention Development, and Policy on Health-Related Stigmas. *BMC Med.* **2019**, *17*, 31. [CrossRef]
- 53. Roelen, K. Receiving Social Assistance in Low- and Middle-Income Countries: Negating Shame or Producing Stigma? *J. Soc. Policy* **2019**, *49*, 705–723. [CrossRef]
- 54. Mizumoto, K.; Kagaya, K.; Zarebski, A.; Chowell, G. Estimating the Asymptomatic Proportion of Coronavirus Disease 2019 (COVID-19) Cases on Board the Diamond Princess Cruise Ship, Yokohama, Japan, 2020. Euro. Surveill. 2020, 25, 2000180. [CrossRef]
- 55. Nishiura, H.; Kobayashi, T.; Miyama, T.; Suzuki, A.; Jung, S.; Hayashi, K.; Kinoshita, R.; Yang, Y.; Yuan, B.; Akhmetzhanov, A.R.; et al. Estimation of the Asymptomatic Ratio of Novel Coronavirus Infections (COVID-19). *Int. J. Infect. Dis.* **2020**, *94*, 154–155. [CrossRef] [PubMed]
- 56. Treibel, T.A.; Manisty, C.; Burton, M.; McKnight, Á.; Lambourne, J.; Augusto, J.B.; Couto-Parada, X.; Cutino-Moguel, T.; Noursadeghi, M.; Moon, J.C. COVID-19: PCR screening of asymptomatic health-care workers at London hospital. *Lancet* 2020, 395, 1608–1610. [CrossRef]
- 57. Schmitz, B.W.; Innes, G.K.; Prasek, S.M.; Betancourt, W.Q.; Stark, E.R.; Foster, A.R.; Abraham, A.G.; Gerba, C.P.; Pepper, I.L. Enumerating Asymptomatic COVID-19 Cases and Estimating SARS-CoV-2 Fecal Shedding Rates via Wastewater-Based Epidemiology. *Sci. Total Environ.* 2021, 801, 149794. [CrossRef] [PubMed]
- 58. Devault, D.A.; Maguet, H.; Merle, S.; Péné-Annette, A.; Lévi, Y. Wastewater-Based Epidemiology in Low Human Development Index States: Bias in Consumption Monitoring of Illicit Drugs. *Environ. Sci. Pollut. Res.* **2018**, 25, 27819–27838. [CrossRef] [PubMed]
- World Health Organization. UNICEF Progress on Sanitation and Drinking-Water: 2015 Update and MDG Assessment; World Health Organization: Geneva, Switzerland; UNICEF: New York, NY, USA, 2015; ISBN 978-92-4-150914-5.

Water 2021, 13, 2897 13 of 14

60. UNESCO. *Wastewater: The Untapped Resource*; The United Nations World Water Development Report; UNESCO: Paris, France, 2017; ISBN 978-92-3-100201-4.

- 61. Shrestha, S.; Shrestha, S.; Shindo, J.; Sherchand, J.B.; Haramoto, E. Virological Quality of Irrigation Water Sources and Pepper Mild Mottle Virus and Tobacco Mosaic Virus as Index of Pathogenic Virus Contamination Level. *Food Environ. Virol.* **2018**, *10*, 107–120. [CrossRef] [PubMed]
- 62. WHO; UNICEF. *Progress on Drinking Water, Sanitation and Hygiene-2017 Update and SDG Baseline*; World Health Organization (WHO); United Nations Children's Fund (UNICEF): Geneva, Switzerland, 2017; Available online: https://apps.who.int/iris/bitstream/handle/10665/258617/9789241512893-eng.pdf?sequence=1 (accessed on 23 March 2020).
- 63. Eales, K.; Blackett, I.; Siregar, R.; Febriani, E. Review of Community-Managed Decentralized Wastewater Treatment Systems in Indonesia; World Bank: Washington, DC, USA, 2013; Available online: http://hdl.handle.net/10986/17751 (accessed on 16 August 2021).
- 64. WHO; UNICEF JMP. Estimates on the Use of Water, Sanitation, and Hygiene. 2019. Available online: https://data.unicef.org/topic/water-and-sanitation/covid-19/ (accessed on 25 March 2020).
- 65. Massoud, M.A.; Tarhini, A.; Nasr, J.A. Decentralized Approaches to Wastewater Treatment and Management: Applicability in Developing Countries. *J. Environ. Manag.* **2009**, *90*, 652–659. [CrossRef]
- 66. Mara, D.D.; Evans, B.E. Sanitation and Water Supply in Low-Income Countries; Ventus Publishing: Erie, CO, USA, 2011; p. 149.
- 67. Kennedy-Walker, R.; Mehta, N.; Thomas, S.; Gambrill, M. Connecting the Unconnected: Approaches for Getting Households to Connect to Sewerage Networks; World Bank Group: Washington, DC, USA, 2011; Available online: https://openknowledge.worldbank.org/bitstream/handle/10986/34791/154444.pdf?sequence=7&isAllowed=y (accessed on 23 March 2020).
- 68. Hyun, C.; Burt, Z.; Crider, Y.; Nelson, K.L.; Prasad, C.S.S.; Rayasam, S.D.G.; Tarpeh, W.; Ray, I. Sanitation for Low-Income Regions: A Cross-Disciplinary Review. *Annu. Rev. Environ. Resour.* **2019**, *44*, 287–318. [CrossRef]
- 69. Calabria de Araujo, J.; Gavazza, S.; Leao, T.L.; Florencio, L.; da Silva, H.P.; Albuquerque, J.D.O.; de Lira Borges, M.A.; de Oliveira Alves, R.B.; Rodrigues, R.H.A.; dos Santos, E.B. SARS-CoV-2 Sewage Surveillance in Low-Income Countries: Potential and Challenges. J. Water Health 2021, 19, 1–19. [CrossRef]
- 70. Pandey, D.; Verma, S.; Verma, P.; Mahanty, B.; Dutta, K.; Daverey, A.; Arunachalam, K. SARS-CoV-2 in Wastewater: Challenges for Developing Countries. *Int. J. Hyg. Environ. Health* **2021**, 231, 113634. [CrossRef]
- 71. Kumar, M.; Patel, A.K.; Shah, A.V.; Raval, J.; Rajpara, N.; Joshi, M.; Joshi, C.G. First Proof of the Capability of Wastewater Surveillance for COVID-19 in India through Detection of Genetic Material of SARS-CoV-2. *Sci. Total Environ.* **2020**, 746, 141326. [CrossRef]
- 72. Daughton, C.G. Real-Time Estimation of Small-Area Populations with Human Biomarkers in Sewage. *Sci. Total Environ.* **2012**, 414, 6–21. [CrossRef]
- 73. Xagoraraki, I.; O'Brien, E. Wastewater-Based Epidemiology for Early Detection of Viral Outbreaks. In *Women in Water Quality*; O'Bannon, D.J., Ed.; Springer International Publishing: New York, NY, USA, 2020; pp. 75–97.
- 74. Chiaia, A.C.; Banta-Green, C.; Field, J. Eliminating Solid Phase Extraction with Large-Volume Injection LC/MS/MS: Analysis of Illicit and Legal Drugs and Human Urine Indicators in US Wastewaters. *Environ. Sci. Technol.* **2008**, 42, 8841–8848. [CrossRef] [PubMed]
- 75. Baker, D.R.; Kasprzyk-Hordern, B. Multi-Residue Analysis of Drugs of Abuse in Wastewater and Surface Water by Solid-Phase Extraction and Liquid Chromatography–Positive Electrospray Ionisation Tandem Mass Spectrometry. *J. Chromatogr. A* **2011**, *1218*, 1620–1631. [CrossRef]
- 76. Chen, C.; Kostakis, C.; Gerber, J.P.; Tscharke, B.J.; Irvine, R.J.; White, J.M. Towards Finding a Population Biomarker for Wastewater Epidemiology Studies. *Sci. Total Environ.* **2014**, 487, 621–628. [CrossRef] [PubMed]
- 77. Bright-Davies, L.; Lüthi, C.; Jachnow, A. DEWATS for Urban Nepal: A Comparative Assessment for Community Wastewater Management. *Waterlines* **2015**, *34*, 119–138. [CrossRef]
- 78. Singh, A.; Sawant, M.; Kamble, S.J.; Herlekar, M.; Starkl, M.; Aymerich, E.; Kazmi, A. Performance evaluation of a decentralized wastewater treatment system in India. *Environ. Sci. Pollut. Res.* **2019**, *26*, 21172–21188. [CrossRef] [PubMed]
- 79. Shuma, R.S.; Alamgir, M. Wastewater Quality at Different Stages of Decentralised Wastewater Treatment Process in the Peoples Panchtala Colony at Kalishpur, Khulna. International Conference on Mechanical, Industrial and Materials Engineering 2013 (ICMIME2013), Rajshahi, Bangladesh, 1–3 September 2013; RUET, 814 Paper ID: RT-10. Available online: http://icmime-ruet.ac.bd/2013/Contents/Technical%20Papers/Related%20Technology/RT-10.pdf (accessed on 17 September 2021).
- 80. Water Information Network–South Africa. Dewats Processor for Decentralized Wastewater Treatment–Technical Lessons from eThekwini Municipality. 2014. Available online: https://www.susana.org/_resources/documents/default/2-2039-dewats-process-for-decentralised-wastewater-treatmentlessonv9.pdf (accessed on 17 September 2021).
- 81. Rice, E.W.; Baird, R.B.; Eaton, A.D. (Eds.) *Standard Methods for the Examination of Water and Wastewater*, 23rd ed.; American Public Health Association: Washington, DC, USA; American Water Works Association: Denver, CO, USA; Water Environment Federation: Alexandria, VA, USA, 2017; ISBN 9780875532875.
- 82. Koottatep, T.; Ferre, A.; Chapagain, S.; Fakkaew, K.; Strande, L. *Methods for Faecal Sludge Analysis*; Velkushanova, K., Strande, L., Ronteltap, M., Koottatep, T., Brdjanovic, D., Buckley, C., Eds.; IWA Publishing: London, UK, 2021; pp. 55–84.
- 83. Larsen, T.A.; Hoffman, S.; Luthi, C.; Truffer, B.; Maurer, M. Emerging Solutions to the Water Challenges of an Urbanizing World. *Science* **2016**, 352. [CrossRef] [PubMed]

Water 2021, 13, 2897 14 of 14

84. Huang, Q.; Jackson, S.; Derakhshan, S.; Lee, L.; Pham, E.; Jackson, A.; Cutter, S.L. Urban-rural differences in COVID-19 exposures and outcomes in the South: A preliminary analysis of South Carolina. *PLoS ONE* **2021**, *16*, e0246548.

- 85. Global Partnership to Make Available 120 Million Affordable, Quality COVID-19 Rapid Tests for Low- and Middle-Income Countries. Available online: https://www.who.int/news/item/28-09-2020-global-partnership-to-make-available-120-million-affordable-quality-covid-19-rapid-tests-for-low--and-middle-income-countries (accessed on 13 September 2021).
- 86. Takeda, T.; Kitajima, M.; Abeynayaka, A.; Huong, N.T.T.; Dinh, N.Q.; Sirikanchana, K.; Navia, M.; Sam, A.A.; Tsudaka, M.; Setiadi, T.; et al. Chapter 11. Governance of wastewater surveillance systems to minimize the impact of COVID-19 and future epidemics: Cases across Asia-Pacific. In *Environmental Resilience and Transformation in Times of COVID-19: Climate Change Effects on Environmental Functionality*; Ramanathan, A., Chidambaram, S., Jonathan, M.P., Prasanna, M.V., Kumar, P., Arriola, F.M., Eds.; Elsevier: Amsterdam, The Netherlands, 2021; pp. 115–125.
- 87. Kitajima, M.; Ahmed, W.; Bibby, K.; Carducci, A.; Gerba, C.P.; Hamilton, K.A.; Haramoto, E.; Rose, J.B. SARS-CoV-2 in Wastewater: State of the Knowledge and Research Needs. *Sci. Total Environ.* **2020**, 739, 139076. [CrossRef]
- 88. Bivins, A.; North, D.; Ahmad, A.; Ahmed, W.; Alm, E.; Been, F.; Bhattacharya, P.; Bijlsma, L.; Boehm, A.B.; Brown, J.; et al. Wastewater-Based Epidemiology: Global Collaborative to Maximize Contributions in the Fight Against COVID-19. *Environ. Sci. Technol.* 2020, 54, 7754–7757. [CrossRef] [PubMed]