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Field grand challenge with emerging superbugs and the novel coronavirus (SARS-CoV-2) on plastics and in water

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ABSTRACT

This opinion paper reports field grand challenges associated with plastic and water contaminated with the novel coronavirus (severe acute respiratory syndrome coronavirus 2, SARS-CoV-2) and superbugs, given the emergency of public health and environmental protection from the presence of lethal viruses and bacteria. Two primary focuses of detection and treatment methods for superbugs and the novel coronavirus (SARS-CoV-2) are investigated, and the future outlook is provided based on grand challenges identified in the water field. Applying conventional treatment technologies to treat superbugs or the novel coronavirus (SARS-CoV-2) has brought negative results, including ineffective treatment, formation of toxic byproducts, and limitation of long-term performance. Existing detection methods are not feasible to apply in terms of sensitivity, difficulty of applications in field samples, speed, and accuracy at the time of sample collection. Few studies are found on superbugs or adsorption of the novel coronavirus (SARS-CoV-2) on plastic, as well as effects of superbugs or the novel coronavirus (SARS-CoV-2) on treatment of plastic waste and wastewater. With the need for and directions of further research and challenges discussed in this paper, we believe that this opinion paper offers information useful to a wide audience, including scientists, policy makers, consultants, public health workers, and field engineers in the water sector.

1. Introduction

The recent COVID-19 pandemic has raised considerable concern not only due to the transmission of infectious diseases linked with serious illness or even death but also due to environmental pollution. In view of the environmental concern, severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2, the novel coronavirus) is likely to contaminate water from various sources (hospitals, quarantine zones, biomedical waste dumping sites, infected patients’ urine/faeces, etc.), and a residual trace level of the virus may survive for several days even after the virus-contaminated water has been disinfected.

Besides, the COVID-19 pandemic has escalated plastic wastes, even from people’s dining habitat (e.g., using plastic containers for meals delivered to their home/business office, instead of eating in restaurants), leading to the significant burden of waste disposal. Because of the hydrophobic nature of plastics, extensive treatment efficacy may be required for toxic contaminants adsorbed on plastics. In addition to the virus-laden plastics (e.g., medical gloves and masks), there is increasing presence of antibiotic-resistant bacteria from drugs accompanied by the growing number of patients (e.g., antibiotics for pneumonia treatment of patients) during the COVID-19 pandemic. This also raises a considerable health risk or potential infection due to the exposure of field operators at wastewater/waste treatment plants to the deadly virus and superbugs through aerosols or contact with contaminated water.

Superbugs, which are viral infections caused by bacteria and are enormously hard to treat, have been spread by plastics and detected in aquatic environments including oceans, rivers, and lakes, exhibiting heavy contamination with various types of antibiotic-resistant bacteria (ARB) and potential transformation from all bacterial species into superbugs [1]. In fact, the decreasing density of E. coli collected from sewage treatment plants turned out to be due to evolving high antibiotic resistance through genetic mutation from disinfection of bacteria using chlorination or ultraviolet radiation treatment [2]. Five common antibiotic-resistant superbugs are known as carbapenem-resistant enterobacteriaceae (CRE), multidrug-resistant acinetobacter (detected in water and soil), neisseria gonorrhoeae, methicillin-resistant Staphylococcus aureus (MRSA), and clostridium difficile (C. diff) [3]. Viral pathogens transmitted through water are considered to be emerging
contaminants because of their increasing threat to water quality and persistence to degradation in water environments. Major human viruses that are potentially transmitted via a waterborne route include norovirus, enterovirus, hepatitis virus, adenovirus, influenza virus, and coronavirus [4]. Similarly to viral pathogens transmitted through water, antibiotics are detected in surface water through various routes including wastewater treatment plants (WWTPs), medical waste, agricultural and stormwater runoff, and industrial waste [5]. At WWTPs, detection of different types of residual antibiotics (e.g., pharmaceuticals including clindamycin antibiotics, sulfamethoxazole, and tetracycline) were reported from various treatment processes [5]. The treatment efficiencies ranged from 72% removal of clarithromycin with a combined activated sludge and UV, to 99% removal of ciprofloxacin with a combined activated sludge and chlorination, and to 100% removal of tetracycline with a carbon membrane coated with nano-TiO₂.

Despite increasing concerns on superbugs, which are detected even in drinking water and plastic waste, there has been a paucity of research on emerging contaminants (e.g., superbugs and the novel coronavirus (SARS-CoV-2)) in water and adsorbed on plastics. During the COVID-19 pandemic, the potential risk of superbugs is growing, and superbugs on microplastics (MPs) enrich antibiotic-resistant bacteria on the MP surfaces compared to those in water. While there has been lack of information regarding potential risks of superbugs on plastics, one recent study by Zhang et al. (2020) [6] revealed increasing ARB on the surface of MPs consisting of 75% polyethylene terephthalate among the collected MPs (e.g., 100–5000 times higher counts of cultivable ARB than those in water samples), implying that MPs serve as vectors to spread ARB, especially superbugs. Similarly, the novel coronavirus (SARS-CoV-2) on plastics lasts at least 3 days [7–9]; this increases the potential risks to the environment and public health, especially because the public could be reinfected by touching the plastic surface contaminated with the deadly virus.

Although incineration of plastics contaminated with the novel coronavirus (SARS-CoV-2) (e.g., medical gloves and masks) could prevent the spread of virus, the virus adsorbed on plastics is a grand challenge, because of a number of reasons. First, incineration has substantial costs of building the infrastructure and running such incineration plants; these are drawbacks of recycling and waste reduction. Second, for developing and underdeveloped countries, incineration of plastic contaminated with the virus is not affordable to operate. Furthermore, incinerators release a toxic smoke by burning hazardous waste, thus increasing air pollution considerably. For instance, India, a country with the second highest number of the novel coronavirus cases (according to the COVID-19 Dashboard by the Center for Systems Science and Engineering (CSSE) at Johns Hopkins University), has dealt with significant air pollution issues. Incineration of significant amounts of plastic waste contaminated with the novel coronavirus (SARS-CoV-2) is not an ideal solution for the country, as it deteriorates the air pollution issue.

Field grand challenges exist in dealing with recalcitrant contaminants adsorbed on plastics, in monitoring and detecting such contaminants in complex aquatic systems, and in developing new treatment technologies because of their resonance to existing (conventional) treatment methods. In this opinion paper, field grand challenges, the novel coronavirus (SARS-CoV-2) and superbugs, are examined in detection and monitoring their risk to public health, as well as in treatment processes of wastewater containing superbugs or the novel coronavirus (SARS-CoV-2). Then, the future outlook for addressing water and plastic contaminated with emerging superbugs and the novel coronavirus (SARS-CoV-2) is provided.

2. Detecting and monitoring superbugs and the novel coronavirus (SARS-CoV-2)

The detection of residual contaminants, particularly bacteria and viruses, is critical yet difficult to measure because of their presence at low concentrations in aquatic environments. Among several detection methods, a phylogenetic microarray was reported to detect dozens of pathogens including human enteric viruses at a fast speed and without cultivation for environmental samples [10]. This technology appears to be practical to detect emerging pathogens present in floodwaters and could be a possible candidate for detection of SARS-CoV-2 in water samples. Given the transmission from symptomatic and asymptomatic infected patients and rapid spread of SARS-CoV-2, wastewater-based epidemiology (WBE) could serve as a worthy tool for informing the public of potential infectious disease outbreaks.

Recent developments in various sensors could focus on not only applying the sensors in field measurements but also tracing possible sources of the novel coronavirus (SARS-CoV-2), which is critical for infection prevention by the deadly virus [11]. Although WBE offers benefits including early warning of potential infectious disease outbreaks, one of its challenges is extraction of the target contaminants in a complex wastewater matrix. Ideally, analytical methods for simple, accurate, rapid, affordable, field-applicable, and adequately sensitive detection even at a trace level are suited for the detection of novel coronavirus (SARS-CoV-2) and superbugs, public health monitoring and prevention of infectious diseases.

Among cell-based, nucleic acid-based, and biosensor-based detection technologies, miniaturized polymerase chain reaction (PCR) instruments were found to be most suitable for easy field deployment for sensitive pathogenic bacterial detection [12]. An optical online bacteria sensor based on 3D image recognition, which identifies bacteria and abiotic particles, was applied to monitor drinking water quality [13]. Although online-monitoring sensors are useful for early warning with rapid detection (with a 10 min time resolution) of microbes at the point of water sample collection and have capacity to distinguish between bacteria and abiotic particles, the method has a detection limit for smaller particles or bacteria. Compared to culture-based methods, which may yield false negative results because of the non-culturable state of pathogens, molecular techniques could acquire accurate quantification and characterization of pathogens, even if there are issues with the lack of standard protocols and sample processing [14].

Several detection methods for waterborne pathogens are compared, along with challenges identified for each method (Table 1). Antibiotic-resistant Staphylococcus bacteria were detected using a biosensor-based method [15]; however, issues associated with sensitivity and application of the biosensor to field environmental samples are challenging to overcome. Recently, nanopores for fluorescence in situ hybridization (FISH) were applied to detect ampicillin-resistant E. coli, exhibiting strong fluorescent signals with pH stability and demonstrating possible application of light stable FISH for antibiotic-resistant bacteria [16]. However, FISH, which is a molecular cytogenetic technique that exposes chromosomes to a small DNA sequence with an attached fluorescent molecule, has issues with low sensitivity and requires pre-enrichment and concentration steps [17].

Surface-enhanced Raman spectroscopy (SERS), a type of Raman spectroscopy using nanomaterials, is another technique that has a capacity to detect antibiotic-resistant bacteria (e.g., methicillin-resistant S. aureus (MRSA) and pseudomonas aeruginosa) [18]. However, major challenges with this detection method include the high cost of a SERS instrument, difficulty to deploy the SERS instrument in field monitoring settings, and matrix interference and spectra changes during measurement [19].

Polymerase chain reaction (PCR), one of molecule-based methods, is based on denaturalization of duplex DNA, annealing of the primers, and elongation of the primers to amplify a specific target DNA sequence [20]. The novel coronavirus (SARS-CoV-2) in secondary-treated wastewater has recently been detected using an N_Sarbeco qPCR assay following the electronegative membrane-vortex (ENV) method [21]. The ENV method used for the extraction of water samples exhibited superior performance compared to the membrane adsorption-direct RNA extraction method [21]. However, because of a higher limit of
Overall, primary consideration for development of detection technologies includes detection of a low concentration of pathogens, sensitivity, speed (close-to-real-time detection of viruses and bacteria), low cost, automation, reproducibility, specificity, sensitivity, alleviating inhibitors and interactions among pathogens.

### 3. Field wastewater treatment

With the world’s increasing number of the emerging disease outbreaks and the resultant high risk to the environment of a community, mitigation and control of viruses and bacteria are essential following monitoring and detection of emerging contaminants. Treated effluent is critical for water reuse as reclaimed water could be applied in various ways, including irrigation.

In a recent case study, high level (500–18,700 copies L⁻¹) of the novel coronavirus (SARS-CoV-2) was found in a septic tank even after disinfection with 800 g m⁻³ sodium hypochlorite, indicating release of the virus from stool particles as a secondary source of the virus and potential transmission through drainage pipelines [26]. The study indicated that the novel coronavirus (SARS-CoV-2) is not completely removed by following the WHO disinfection guidelines (e.g., free chlorine more than 0.5 mg L⁻¹ after at least 30 min). As such, development and application of new treatment methods to the novel coronavirus (SARS-CoV-2) are encouraged, because of the formation of disinfection byproducts at an excessive dosage of sodium hypochlorite. Furthermore, chlorination resulted in increasing antibiotic-resistant bacteria including potentially pathogenic organisms, as also found in the occurrence of antibiotic-resistant bacteria in both drinking water and wastewater [27].

Chlorine disinfection is widely applied for the inactivation of waterborne pathogens because of its easy deployment, broad sterilization, cost-effectiveness, and high efficiency; however, harmful and carcinogenic disinfection byproduct formation is a major challenge [26]. Other disinfection methods with solar/UV irradiation, boiling, nanofiltration, or addition of free chlorine could be suited for remote local communities, where centralized water treatment facilities are not available [28]. Unlike non-enveloped viruses, coronaviruses proved to be more sensitive to UV irradiation through damaging the nucleic acid by pyrimidine [29].

UV irradiation and chlorine disinfection are commonly applied for inactivation of viruses through oxidation processes, but with sand-filtered pretreatment, better removal of coliphage and E. coli was achieved [30]. Ozone is widely applied because of its high efficacy in treating various viruses and recalcitrant contaminants. Nonetheless, there have been no reports about treatment efficacy of the novel coronavirus (SARS-CoV-2) with ozone. Given that the SARS-CoV-2 is an enveloped virus with 0.1 μm in diameter [31], ultrafiltration, nanofiltration, and reverse osmosis membrane filtration could serve as an alternative to chlorine disinfection or advanced oxidation technologies of treating the novel coronavirus (SARS-CoV-2) in water.

Microalgae-based biological method as an alternative to disinfection or other physicochemical methods could be applied for virus removal under sunlight with optimum pH and temperature [32]. Wastewater treatment ponds for virus-containing wastewater exhibited treatment efficacy of one log₁₀ reduction of virus content between around 15 and 21 days, indicating further treatment requirements [33]. However, no data/results are reported about the removal of the novel coronavirus (SARS-CoV-2) from wastewater treatment ponds.

Among treatment developments, a multifunctional fluorescence-magnetic biochar exhibited highly efficient removal of methicillin-resistant Staphylococcus aureus (MRSA) superbugs in water, suggesting the capability of melittin, an antimicrobial peptide-attached multifunctional biochar, to disinfect the superbugs followed by magnetic separation [34]. Using a biochar is beneficial in capturing pathogens because of a carbon-rich affordable byproduct from biomass and a strong adsorption capability [34]. This method has exhibited to be detection (LOD) with a lower filter volume (i.e., 200 mL) from the influent compared to that for secondary-treated wastewater with a higher filter volume of 5000 mL, no coronavirus (SARS-CoV-2) was detected from influent samples, whereas a concentration of 2400 copies higher filter volume of 5000 mL, no coronavirus (SARS-CoV-2) was detected in influent compared to that for secondary-treated wastewater with a high risk of losing functionality of the agents (e.g., nanosilver and bro-nopol) applied for electrosyn membrane during leaching [23]. Potential interference from inhibitors present in field samples during nanofiber membrane filtration should be overcome.

### Table 1: Challenges and applications of detection technologies.

| Detection method                           | Challenges                                      | Application of detection methods to superbugs or the novel coronavirus (SARS-CoV-2) |
|-------------------------------------------|-------------------------------------------------|-------------------------------------------------------------------------------------|
| Biosensor-based method                    | • Sensitivity to pH, change of mass, and temperature [24]  
• Difficult for biosensors to apply to real-world environmental samples (e.g., interferring microbial species, particulate matter, and humic substances) [25] | Staphylococcus Bacteria [15]                                                      |
| Fluorescence in situ hybridization (FISH) | • Low sensitivity  
• Pre-enrichment and concentration steps [17] | Ampicillin-resistant Escherichia coli [16]                                           |
| Surface-enhanced Raman spectroscopy (SERS) | • Matrix interference and spectra changes during measurement [19]  
• High cost of the confocal micro-Raman instrument  
• Need for user-friendly software  
• Not applicable for field monitoring settings | Novel coronavirus (SARS-CoV-2), with an N_Sarbeco qPCR assay following the electronegative membrane vortex (ENV) method [21] |
| Polymerase chain reaction (PCR)           | • Accurate primers and optimal reaction mixture are required to avoid faulty results [17,22]. | Novel coronavirus (SARS-CoV-2) [23]                                                 |
| Nanofiber filters (application of a nanofiber membrane at a pretreatment process stage) | • High risk of losing functionality of agents (e.g., nanosilver and bronopol) applied for electrosyn membranes during leaching [23]  
• Interference from inhibitors in field samples during nanofiber membrane filtration | Novel coronavirus (SARS-CoV-2) [23]                                                 |

**Factors for development of detection methods**

• Detecting low concentration of pathogens  
• Sensitivity, automation, reproducibility, and specificity  
• Speed (close-to-real-time detection of viruses and bacteria)  
• Low cost; alleviating interactions among pathogens  
• Elimination of inhibitors
useful for selective inactivation of MRSA superbugs in water (containing 430,000 colony-forming unit (cfu) mL⁻¹ of MRSA superbugs) through an anti-MRSA antibody-attached multifunctional biochar, achieving complete removal of MRSA superbugs through pore formation on the MRSA membrane [34].

Recently, novel ZnO-coated nanobeads were employed to treat antibiotic-resistant bacteria (i.e. E. coli DH-5α and pseudomonas aeruginosa) at the initial concentration of 10⁶ CFU mL⁻¹, achieving the removal efficiency of 98 % and 88 %, respectively, through the generation of reactive oxygen species (ROS) [35]. Nanohybrids such as ZnO- and TiO₂-conjugated carbon nanotube (CNT) or graphene oxide (GO) exhibited high treatment efficiency of a multidrug-resistant colliform bacterium (i.e. E. coli DH5α) [36]. Among the four tested nanohybrids, ZnO-GO had the highest removal efficiency, followed by ZnO-CNT, TiO₂-GO, and TiO₂-CNT through the dominant mechanism of ROS formation. The increasing dispersion of TiO₂ and ZnO by GO attributed to higher treatment efficiency than that from ZnO- and TiO₂-conjugated CNTs [36]. Various types of nanomaterials (e.g., fullerenes, zero-valent iron, and TiO₂) could be incorporated into other treatment methods (e.g., photocatalysis and membranes) for inactivation of the novel coronavirus (SARS-CoV-2) and superbugs.

In the study investigating treatment efficacy of the novel coronavirus (SARS-CoV-2), physical and chemical disinfection technologies and incineration were compared under various conditions, including cost, pH, degree of maintenance on chemical reagents, and amount of wastes [37]. According to the study, several disinfection technologies for hospital wastewater are proposed under the conditions such as hospital size, costs, and maintenance. For instance, ozone or UV or sodium hypochlorite disinfection is suggested at small-scale hospitals, whereas liquid chlorine or chloramine dioxide disinfection is recommended for large-scale hospitals.

The occurrence of antibiotic-resistant bacteria at a WWTP was studied, and it exhibited the highest concentrations of ampicillin-resistant E. coli (63 % among antibiotic-resistant E. coli) in the effluent treated with post-UV and chlorination disinfection [38]. The fate of the novel coronavirus (SARS-CoV-2) in wastewater is unknown, but it may be affected by environmental conditions (e.g., pH, temperature, dissolved matter, wastewater characteristics and compositions) and by different wastewater treatment processes [39]. Several factors, which may affect the survival of coronavirus in wastewater, include viral structure, wastewater characteristics/composition, temperature, and pH [39].

Table 2 summarizes challenges identified among conventional and newly developed treatment methods for superbugs or the novel coronavirus (SARS-CoV-2). Chlorine disinfection and UV irradiation removed the novel coronavirus (SARS-CoV-2) to some extent, but issues with the formation of toxic disinfection byproducts at high concentrations of sodium hypochlorite and limitation for large-scale applications should be addressed. Material-based treatment methods (i.e. biochar, nanobeads, and nanohybrids) revealed treatment efficacy of antibiotic-resistant bacteria (e.g., MRSA superbugs, E. coli DH-5α, pseudomonas aeruginosa), but there are challenges to overcome, such as large-scale development, long-term performance, inhibition from other contaminants in wastewater, and potential release of metal ions.

4. Future outlook

Future studies should focus on the validity, reproducible detection methods for emerging superbugs and the novel coronavirus (SARS-CoV-2) on plastic and in water, to determine the extent of contamination and monitor potential infection outbreaks. For public health risk assessment, improvement in quantitative microbial risk assessment (QMRA) analysis is necessary with the target virus-specific properties, water quality and exposure data [4]. As discussed and reviewed in the previous section, no best wastewater treatment method exists to treat the emerging contaminants of concern (e.g., superbugs and the novel coronavirus (SARS-CoV-2)); notably, when using conventional chlorine disinfection, antibiotic resistance was developed when chlorine reacted with drugs in water [40].

The lifespan of the novel coronavirus (SARS-CoV-2) was reported to be from 3 h in aerosols, to 4 h in copper, to 24 h in cardboard, and to 3 days in stainless steel, plastic, sewage, and solid faces [7–9]. Given the long lifespan of the novel coronavirus (SARS-CoV-2) in plastic and in wastewater, further research should be carried out regarding proper control and treatment methods to prevent transmission through contaminated plastic surfaces and aerosols from wastewater treatment facilities. Notably, the mechanisms of the novel coronavirus (SARS-CoV-2)/superbug adsorption on plastic and the resultant effects of the adsorbed virus/superbug on plastic under heterogeneous environmental conditions are not well understood, requiring future research.

There are field grand challenges in dealing with emerging contaminants (i.e. superbugs and the novel coronavirus (SARS-CoV-2)) adsorbed on plastic and in applying conventional treatment methods due to ineffectiveness (e.g., development of antibiotic resistance to chlorine disinfection) and possible transmission of pathogens if not handled or treated properly.

The improperly treated water or mishandled waste return back to the public through drinking of contaminated water, consuming crops from contaminated fertilizer, water reuse for irrigation, or contact with contaminated surfaces or water, threatening public health and the environment, and even transmitting infectious diseases; therefore, this requires further research and new regulations for handling emerging contaminants and applying reclaimed water.

Future perspectives are summarized, based on the identified challenges and a literature analysis:
(1) Given the evolving transmission of the novel virus and superbugs, the efficacy of conventional treatment methods should be re-evaluated for development of new treatment technologies, which should consider the persistence of viruses and bacteria, and environmental factors (e.g., temperature, pH, the presence of NOM and inhibitors, and sunlight).

(2) In detecting the novel coronavirus (SARS-CoV-2) and superbugs in water, several factors such as enrichment and concentration from large volumes, any interference from different types of water, an automated device with online real-time monitoring, optimization of detection methods, and the potential presence of pathogens in biofilm should be counted in new development of detection methods. Furthermore, many ongoing developments of sensors and online detection systems need to improve to test samples in real field conditions, particularly in terms of their sensitivity, accuracy, reproducibility, limit of detection, and software data management.

(3) The spread of the virus through biofilms, the virus-laden biofilm–plastic interactions, and the effect of plastic on biofouling mitigation deserve future research, because of limited research from these perspectives.

(4) There are various routes of virus transmission, such as discharging untreated wastewater into the community, field workers including wastewater operators and workers handling hazardous wastes, faeces and urine of asymptomatic and symptomatic individuals, sewage treatment plants, contaminated crops, inhalation, and reclaimed sludge. Leveraging big data and artificial intelligence (AI) is recommended for mitigating and controlling transmission of the virus during the COVID-19 pandemic, along with wastewater-based epidemiology (WBE).

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.

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