

Literature Review:

SARS—CoV-2 Drinking Water Risks

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Introduction

The purpose of this document is to provide evidence on the risks to drinking water and subsequential virus transmission from SARS—CoV-2. In 2020 England and Wales had national measures initiated to prevent the spread of the SARS—CoV-2 virus. Surveillance monitoring has been completed across a number of areas including wastewater treatment plants, surface waters, key infrastructure and communities to track the transmission of the virus and identify areas of high risk where exposure to the virus may be greater. There is now a substantial number of scientific and grey literature publications on this topic. The information presented below provides a literature review summary on 5 key areas;

- 1. Amount of SARS-CoV-2 RNA excreted in urine and faeces
- 2. SARS-CoV-2 removal in wastewater treatment
- 3. Survival of SARS-CoV-2 RNA in surface waters
- 4. Transmission in the wider environment
- 5. DWI evidence of virus removal in drinking water treatment

1. Amount of SARS—CoV-2 RNA excreted in urine and faeces

Table 1: Excretion value data from studies. Reference, findings and risks summarised in table.

Reference:	Findings:	Risk Assessment:
Jones et al. 2020	Virus genetic material is more frequently reported in faeces than urine.	Likelihood of the virus being
Shedding of SARS-CoV- 2 in feces and urine and its potential role	Presence of the virus in faeces is similar in patients with and without gastrointestinal (GI) symptoms.	transmitted via faeces or urine is low due to the low virus genetic material via faeces/urine.
in person-to-person	Infectious virus has occasionally been recovered from urine/stool samples.	
transmission and the environment-based	Virus genetic material (measured in genome copies (gc) / ml) is much lower in urine $(10^2 - 10^5 \text{ gc/ml})$ and faeces $(10^2 - 10^7 \text{ gc/ml})$ compared to nasopharyngeal fluids $(10^5 - 10^{11} \text{ gc/ml})$.	
spread of COVID-19	Evidence suggests that asymptomatic, pre/post-symptomatic individuals may still shed the virus at appreciable levels.	
	Evidence suggests viral shedding pattern in faeces indicate rapid accumulation and slow decline.	
Kutti-Sridharan et al. 2020	Faecal shedding may occur for prolonged period beyond when patients test negative based on nasopharyngeal swabs.	Strong possibility for faecal-oral transmission although without
SARS-CoV-2 in different body fluids, risks of transmission,	Live viral Ribonucleic Acid (RNA) was isolated from stool samples of 2 patients who were not displaying diarrhoea as a symptom. There is a possibility for feco-oral transmission and was suggested to explain higher rates of transmission in familial clustering.	identification of active virus RNA, uncertainty remains.
and preventing COVID-19	Unknown whether positive faecal test results are due to inactive RNA or active virion particles.	

Ling et al. 2020 Persistence and clearance of viral RNA in 2019 novel Coronavirus disease rehabilitation patients	43/55 patients recovering after treatment had a longer duration until stool specimens were negative for viral RNA compared to throat swabs, with a median delay of 2 days. 4/58 urine samples tested positive for viral RNA, but viral RNA was still present in 3 patients urine samples after throat swabs were clear.	
Santos et al. 2020 Prolonged faecal shedding of SARS-CoV- 2 in paediatric patients	Greater proportion of children had viral shedding in stools after 14 days of symptoms compared to respiratory samples Viral RNA shedding was longer in faecal samples (mean diff: 9 days) compared to respiratory samples.	Presence does not confirm risk, but increases likelihood.
Gupta et al. 2020 Persistent viral shedding of SARS-CoV- 2 in faeces – a rapid review	53.9% (of combined study results) of those tested for faecal RNA were positive. Faecal shedding duration ranged between 1 – 33 days after negative nasopharyngeal swab.	Insufficient evidence to suggest COVID-19 is transmitted via faecal shed virus.
Wurtzer et al. 2020 Evaluation of lockdown impact on SARS-CoV-2 dynamics through viral genome quantification in Paris Wastewater	Monitoring viral load in wastewater (measured as genome units/ L) showed exponential increase (5.10^4 GU/L on 5 March 2020 to 3.10^6 GU/L on 9 April 2020 – a 2-log increase). SARS-CoV-2 RNA detected in treated wastewater showed 2-log removal of virus compared to wastewater treatment works influent.	

Quick overview of Table 1:

A brief snapshot of the studies listed above suggests the presence of virus RNA in faeces is likely, albeit in lower concentrations than nasopharyngeal fluids. It remains unclear whether the virus RNA in faeces is viable. Results have demonstrated that there may be a likelihood of active/viable viral RNA in faeces.

2 . SARS–CoV-2 removal in wastewater treatment

Table 2: Study evaluation of removal in wastewater treatment. Reference, findings and risks summarised in table.

Reference:	Findings:	Risk Assessment:
Rimoldi et al. 2020 Presence and	Raw and treated water samples from 3 WWTW were compared (Milan, Italy). Results found that all raw water samples tested positive (PCR amplification) while treated water samples were always negative.	Suggests low risk of infective virus in wastewater treatment effluent
vitality of SARS- CoV-2 virus in wastewaters and rivers	Virus infectiousness was not significant and was attributed to a combination of effective removal via treatment and decay of viral vitality. Estimated time from stool emission to the arrival at WWTW was 6-8 hours.	
Westhaus et al. 2021 Detection of SARS- CoV-2 in raw and treated wastewater in Germany	Detections at the inflow at all 9 WWTW studied were at similar concentrations. 3 to 20 gene equivalents/ml were found in the raw wastewater. The infectivity of the raw wastewater samples was assessed and were not found to be infectious. In wastewater treatment comparison of the aqueous and the solid phase of the effluent samples was undertaken. Findings showed 8.8 gene equivalents (ge)/ml compared to 13 ge/ml, respectively. Observed poor removal of SARS-CoV-2 RNA in all 3 of the studied conventional activated-sludge WWTP. Full-scale ozonation at one plant seemed to reduce SARS-CoV-2 fragments in the effluent. Membrane-based WWTW were not studied.	Evidence WWTWs do not fully remove viral RNA through treatment. Although results demonstrated that it was not infectious. Paper concludes that further research is needed to properly evaluate the risks.

Zahedi et al. 2021 Wastewater- based epidemiology – surveillance and early detection of waterborne pathogens	Table of studies considering removal efficiency of wastewater treatment processes.	Demonstrates mixed results.
Hart and Halden 2020 Computational analysis of SARS- CoV-2 surveillance by wastewater- based epidemiology	Wastewater temperatures varies (seasonally) and it has been estimated that at 20°C ~25% of SARS-CoV-2 virus RNA in wastewater should persist even with a transit time to the WWTW of 10 hrs and low virus stability.	Temperature and transit times can impact virus detectability.

Quick overview of Table 2:

The literature gives mixed conclusions with regard to removal of SARS–CoV-2 at wastewater treatment works. Further research into effectiveness of different types of treatment is needed to better understand WWTW effectiveness of removal. The current literature does suggest that RNA found in WWTW effluent is unlikely to be infectious.

3. Survival of SARS—CoV-2 in surface waters

Table 3: Survival in surface waters. Reference, findings and risks summarised in table.

Reference:	Findings:	Risk Assessment:
Rimoldi et al. 2020 Presence and vitality of SARS- CoV-2 virus in wastewaters and rivers	Samples collected from 2 river sampling points downstream of WWTWs in Milano, Italy. Receiving rivers showed some positive PCR amplification for SARS-CoV-2. In this study the presence of SARS-CoV-2 RNA in river samples, in spite of the absence in treated WWTW effluent samples, was thought to indicate the presence of non-treated wastewater present in the surface waters possibly from non-collected domestic discharges of combined sewer overflows. In positive PCR amplification samples the vitality of the SARS-CoV-2 was negligible, indicating absence of sanitary risk and environmental risk of infection from the river water.	The presence of SARS-CoV-2 genome in rivers indicated the partial efficiency of the current sewerage system. Test for vitality indicated that pathogenicity of virus in wastewaters and surficial waters is negligible, and risk for public health should not be significant.
Casanova et al. 2009 Survival of surrogate coronaviruses in water	Study evaluated the survival of 2 surrogate coronaviruses, transmissible gastroenteritis (TGEV) and mouse hepatitis (MHV). Viral stocks were propagated before being added to test water aliquots (viable virus used). These viruses remained infectious in water and sewage for days to weeks suggesting contaminated water is a potential vehicle for human exposure if aerosols are generated. There was a progressive decline in the infectivity of both viruses over 49 days at 25°C. Time required for 99% reduction in infectious titer in reagent-grade water at 25°C was 22 days for TGEV and 17 days for MHV. No significant decline in infectious titer of either virus over 49 days at 4°C. Time required for 99% reduction in infectious titer in lake water at 25°C was 13 days for TGEV and 10 days for MHV. At 4°C, TGEV infectivity declined by approximately 1 log10 by day 14; in contrast, MHV infectivity persisted with no decline in titer after 14 days at 4°C. Time required for 99% reduction in infectious titer in pasteurised settled sewage were 9 days for TGEV and 7 days for MHV at 25oC	Results of this study suggest that coronaviruses can survive and remain infectious in different water types remaining infectious at low (4ºC) and ambient (25ºC) temperatures. Important to note that study samples were inoculated with viable virus. Studies on SARS-Cov-2 completed in 2020 have had negligible success in cultivating the virus from WWTW or surface waters.

Gundy et al. 2009 & 2019 *Survival of coronaviruses in water and wastewater*

The survival of representative coronaviruses, was determined in filtered and unfiltered tap water (4 and 23°C) and wastewater (23°C).

The time required for the virus titer to decrease 99.9% (T99.9) shows that in tap water, coronaviruses are inactivated faster in water at 23°C (10 days) than in water at 4°C (100 days). Coronaviruses die off rapidly in wastewater, with T99.9 values of between 2 and 4 days.

The presence of organic matter and suspended solids in water can provide protection for viruses that adsorb to these particles but at the same time can be a mechanism for removal of viruses if the solids settle out. The level of organic matter and suspended solids in the test waters increased from tap water to secondary effluent to primary effluent. Coronavirus inactivation was greater in filtered tap water than unfiltered tap water.

The hydrophobicity of the viral envelope makes coronaviruses less soluble in water and could therefore increase the tendency of these viruses to adhere to the solids.

Coronaviruses die off very rapidly in wastewater, with a 99.9% reduction in 2–3 days, which is comparable to the data on SARS-CoV survival.

This study demonstrates that the transmission of coronaviruses would be less than enteroviruses in the aqueous environment due to the fact that coronaviruses are more rapidly inactivated in water and wastewater at ambient temperatures.

Coagulation and settlement for particle removal are likely to be effective virus treatment barriers.

La Rosa et al 2020

Coronavirus in water environments: Occurrence, Persistence and concentration methods- A scoping review

This scoping review was conducted to summarize research data on CoV in water environments.

The review investigated three main areas: 1) CoV persistence/ survival in waters; 2) CoV occurrence in water environments; 3) methods for recovery of CoV from waters.

The evidence-based knowledge reported in this paper is useful to support risk analysis processes within the drinking and wastewater chain (i.e., water and sanitation safety planning) to protect human health from exposure to coronavirus through water.

Transmission of COVID-19 through the fecal-oral route has not been demonstrated, nor occurrence of viable SARS-CoV-2 in water environments been proved to date.

The evidence of the presence of CoV in waters is currently very scarce and there is no evidence that human CoV are present in surface or groundwater sources or transmitted through contaminated drinking-water.

Although different studies showed different viral inactivation rates for CoV in water, based on the type of virus and the type of water, generally, there is evidence that CoV is considered unstable in the environment.

CoV has a low stability in the environment;

CoV appears to be inactivated significantly faster in water than non-enveloped human enteric viruses with known waterborne transmission;

Temperature is an important factor influencing viral survival

There is no current evidence that human coronaviruses are present in surface or ground waters or are transmitted through contaminated drinking-water;

Ouick overview of Table 3:

Viability of SARS—CoV-2 in surface waters is negligible presenting a low risk to public health. However, where sources of viable virus entering surface waters are presented e.g. sewerage overflow outfalls/ untreated domestic waste leaks there is evidence to show coronaviruses can survive for long periods at ambient temperatures. Transmission of coronaviruses are thought to be less than enteroviruses in the aqueous environment due to the fact that coronaviruses are more rapidly inactivated in water and wastewater at ambient temperatures. SARS—CoV-2 has low stability in the environment. The hydrophobicity of the viral envelope makes coronaviruses less soluble in water and could therefore increase the tendency of these viruses to adhere to the solids emphasising the removal efficiency of treatments using solids removal via settlement/ sedimentation.

4. Transmission in the wider environment Group (TWEG) Report

Table 4: Evaluation of transmission in the wider environment. Reference, findings and risks summarised in table.

Reference:	Findings:	Risk Assessment:
TWEG report to SAGE 2020	 There is no evidence that SARS-CoV-2 is present in drinking water from either a chlorinated mains or non-mains supply. 	Transmission risk from water is low.
Monitoring the presence and	 SARS-CoV-2 RNA is shed in faeces from many infected individuals and therefore enters building wastewater systems and the sewage network. 	
infections risk of SARS-CoV-2 in the	 Theoretical potential exists for viral material to subsequently enter freshwater or marine water bodies, though inactivation rates and dilution are likely to be high. 	
environment	 Ambient conditions in sewers and natural waterbodies mean that any infectious SARS- CoV-2 is likely to degrade rapidly, whereas RNA is relatively persistent. 	
	 The use of wastewater analysis for detection of SARS-CoV-2 in a population is potentially a sensitive technique capable of providing a leading indicator in advance of clinical testing, particularly where significant numbers of otherwise asymptomatic infections occur. The approach can support public health decision making. 	
	 Current studies have demonstrated little success in recovering infectious SARS-CoV-2 virus from faecal samples in clinical cases. Therefore, although there is a genetic signal for the presence of RNA, there may not be infectious virus present in wastewater 	
	 Quantifying transmission risk is a challenge, however models such as Quantitative Microbial Risk Assessment (QMRA) offers one possible approach45. This is a widely used approach in environmental engineering to assess risks from pathogens, particularly in water and food. 	
	 The challenges with evaluating risk are also compounded by the lack of dose-response data for the virus. Data are available for a number of other coronaviruses48, however most of these are derived from animal studies. 	
	 To assess whether there is an infection risk, environmental samples ideally need to demonstrate that the virus is infectious (viable) and present in adequate dose response concentrations. 	
	RNA is more stable in the environment than infectious virus, and there is no clear correlation between RNA abundance and infectiousness. Emerging data show that 90% of	
	infectious SARS-CoV-2 in filtered fresh water is lost after approximately 2 days at 20°C whereas SARS-CoV-2 RNA is effectively stable under similar (laboratory) conditions.	

Quick overview of Table 4:

Conditions in wastewater networks and WWTW in addition to natural waterbodies mean that any infectious SARS-CoV-2 is likely to degrade rapidly. RNA is more stable in the environment than the infectious virus, and there is no clear correlation between RNA abundance and infectiousness. Theoretical potential exists for viral material to subsequently enter freshwater or marine water bodies, though inactivation rates and dilution are likely to be high.

5. DWI evidence of virus removal in drinking water treatment

Table 5: Evidence of virus removal in drinking water treatment processes (DWI Research Project Reference: 70/2/234). Reference, findings and risks summarised in table.

La Rosa et al 2020
Coronavirus in
water
environments:

Reference:

Occurrence, Persistence and concentration methods- A scoping review

Findings:

The viruses considered of concern for water in WHO Guidelines are principally enteric viruses (familes Adenoviridae, Astroviridae, Caliciviridae, Hepeviridae, Picornaviridae, and Reoviridae) which are non-enveloped viruses. It is well known that these viruses are more resistant to environmental conditions, water treatments and disinfectants than enveloped viruses like coronavirus, as lysis of the viral envelope leads to the loss of functional receptors required for infection of susceptible cells (Wigginton et al., 2015).

According to the results of Wang (2005), SARS-CoV resistance to chlorine is lower than for bacteria. It follows that the current water disinfection practices (drinking water, wastewater, water from swimming pool), effective against non-enveloped viruses and bacteria, are expected to be effective also towards enveloped viruses such as coronaviruses.

Risk Assessment:

Although different studies showed different viral inactivation rates for Coronavirus in water, based on the type of virus and the type of water, generally, there is evidence that Coronavirus is generally considered unstable in the environment and is more susceptible to oxidants, such as chlorine than non-enveloped viruses.

Hyder Consulting 2013 *Project Report 70_2_234*

Study assessed Adenovirus and Norovirus 74% of raw water samples were Adenovirus positive indicating the persistence and ubiquitous nature of this virus in environmental/abstraction waters. Adenovirus was present in raw waters throughout the year and, whilst the water treatment process reduced the level of Adenovirus by between 2 and 4 orders of magnitude, the virus was able to persist through to the pre-chlorination stages. Around 20% of all pre-chlorination (final stage) samples were Adenovirus positive although none of the isolates proved to be infective when assessed by ICC-PCR.

Norovirus was generally not detected in raw waters except from December-March, when 94% of the samples were positive. This winter 'peak' reflects a high prevalence of Norovirus in the community which is consistent with epidemiological evidence. In contrast to Adenovirus, there was apparently no significant effect of treatment on the level of Norovirus. The levels of Norovirus in raw waters were often so low that the demonstration of a significant reduction in numbers was impossible.

For Norovirus, there appeared to be no significant effect of treatment. However, the levels of Norovirus in raw waters were often below the Limit of Detection (LOD), and the demonstration of a significant reduction in numbers was impossible. The proportion of Norovirus positive samples for raw waters (54%) was greater than at later treatment stages (33%) however the difference between treatment stages was not statistically significant.

Removal of Adenovirus in treatment occurred mainly at the first stage (post clarification), thereafter removal was negligible. The coagulation/flocculation stage will facilitate the adsorption of charged viruses on to suspended matter in the water which will then be removed during clarification. Most of the treated water samples were negative (below LOD) (20.3%) positive in pre-disinfection waters. The proportion of Adenovirus positive assays in raw waters (74%) was statistically significantly greater than at later stages (18%). For each works studies levels of Adenovirus were generally reduced from between 2 Log_{10} and 4 Log_{10} (raw water) to below LOD (pre disinfection treatment sample point). The Adenovirus isolated and quantified by q-PCR in both raw and pre-chlorination samples subsequently proved to be non-infective when assessed by semi-quantitative ICC-PCR.

Study demonstrated that Adenovirus is consistently removed from raw water through the water treatment process. Whilst the virus was sometimes detected in low numbers by qPCR in pre-chlorinated waters, none of the isolates proved to be infective when transferred to tissue culture.

Drinking water treatment showed reduction of Norovirus and significant reduction of Adenovirus at study sites.

No isolates proved to be infective when transferred to tissue culture suggesting infectivity risk low.

Quick overview of Table 5:

Drinking Water treatment provides barriers that have been shown to reduce viral concentrations for Adenovirus and Norovirus. Coronavirus is generally considered unstable in the environment and is more susceptible to oxidants, such as chlorine than non-enveloped viruses. Studies have shown viability from water samples to be negligible.

Therefore, the combined effects of, removal from WWTW settlement/ sedimentation processes, exposure to environmental stressors in surface waters, settlement/ sedimentation treatment at drinking water works and the susceptibility to oxidants such as chlorine used for drinking water disinfection, are likely to substantially reduce the risk of SARS—CoV-2 transmission through drinking water. There is no evidence that SARS—CoV-2 is present in drinking water from either a chlorinated mains or non-mains supply. Public health risk from drinking water is low.

Conclusions

The risk of transmission of SARS—CoV-2 through drinking water is unlikely. A brief overview of the available literature for the 5 different stages outlined above suggests that barriers in wastewater treatment and drinking water treatment are likely to be sufficient to substantially reduce the risk of transmission in drinking water. In addition, due to its instability, infectious SARS—CoV-2 is likely to degrade rapidly in the aqueous environment and rates of inactivation and dilution are estimated to be high, particularly at ambient temperatures. Studies conducted to date have been unable to culture viable virus from wastewater treatment work discharge samples and viability from surface water samples has been negligible. A monitoring study on SARS—CoV-2 at drinking water treatment abstraction points is likely to find RNA in water samples however, literature suggests that detections will have required a substantial level of amplification and that the viral RNA is unlikely to be viable. Therefore, the public health risk of SARS—CoV-2 in drinking water is low and does not pose a risk to public health.

References

Jones et al. 2020. Shedding of SARS-CoV-2 in feces and urine and its potential role in person-to-person transmission and the environment-based spread of COVID-19, *Sci. Total Environ.*, https://doi.org/10.1016/j.scitotenv.2020.141364

Kutti-Sridharan et al. 2020. SARS-CoV-2 in different body fluids, risks of transmission, and preventing COVID-19: A comprehensive evidence-based review, *International Journal of Preventive Medicine*, https://doi.org/10.4103/ijpvm.IJPVM 255 20

Ling et al. 2020. Persistence and clearance of viral RNA in 2019 novel Coronavirus disease rehabilitation patients, *Chin. Med. J.*, https://doi.org/10.1097/CM9.000000000000000774

Santos et al. 2020. Prolonged faecal shedding of SARS-CoV-2 in paediatric patients: A quantitative evidence synthesis, *J. Pediatr. Gastroenerol. Nutr.*,

https://doi.org/10.1097/MPG.000000000002798

Gupta et al. 2020. Persistent viral shedding of SARS-CoV-2 in faeces – a rapid review, *Colorectal Disease*, https://doi.org/10.1111/codi.15138

Wurtzer et al. 2020. Evaluation of lockdown impact on SARS-CoV-2 dynamics through viral genome quantification in Paris Wastewaters, https://doi.org/10.1101/2020.04.12.20062679

Rimoldi et al. 2020. Presence and vitality of SARS-CoV-2 virus in wastewaters and rivers, *Sci. Total Environ.*, https://doi.org/10.1016/j.scitotenv.2020.140911

Westhaus et al. 2021. Detection of SARS-CoV-2 in raw and treated wastewater in Germany – Suitability for COVID-19 surveillance and potential transmission risks, *Sci. Total Environ.*, https://doi.org/10.1016/j.scitotenv.2020.141750

Zahedi et al. 2021. Wastewater-based epidemiology – surveillance and early detection of waterborne pathogens with a focus on SARS-CoV-2, Cryptosporidium and Giardia, *Parasitol Res.*, https://doi.org/10.1007/s00436-020-07023-5

Hart and Halden 2020. Computational analysis of SARS-CoV-2 surveillance by wastewater-based epidemiology locally and globally: Feasibility, economy, opportunities and challenges, *Sci. Total Environ.*, https://doi.org/10.1016/j.scitotenv.2020.138875

Casanova et al. 2009. Survival of surrogate coronaviruses in water, *Water Research*, https://doi.org/10.1016/j.watres.2009.02.002

Gundy et al. 2009 & 2019. Survival of coronaviruses in water and wastewater, *Food and Environmental Virology*, https://doi.org/10.1007/s12560-008-9001-6

La Rosa et al 2020. Coronavirus in water environments: Occurrence, Persistence and concentration methods- A scoping review, *Water Res.*, https://doi.org/10.1016/j.watres.2020.115899

TWEG report to SAGE 2020. Monitoring the presence and infections risk of SARS-CoV-2 in the environment, <u>Evidence of Wider Environmental Transmission of SARS-CoV-2 - 12 June 2020</u> (<u>publishing.service.gov.uk</u>)

Hyder Consulting 2013. Project Report 70_2_234, https://www.dwi.gov.uk/research/completed-research/monitoring-microbiological/viruses-in-raw-and-partially-treated-water-targeted-monitoring-using-latest-methods/