

AperTO - Archivio Istituzionale Open Access dell'Università di Torino

## Occurrence of human pathogenic viruses in drinking water and in its sources: A review

**This is a pre print version of the following article:**

*Original Citation:*

*Availability:*

This version is available <http://hdl.handle.net/2318/1884391> since 2023-01-02T10:32:25Z

*Published version:*

DOI:10.1016/j.jes.2022.07.035

*Terms of use:*

Open Access

Anyone can freely access the full text of works made available as "Open Access". Works made available under a Creative Commons license can be used according to the terms and conditions of said license. Use of all other works requires consent of the right holder (author or publisher) if not exempted from copyright protection by the applicable law.

(Article begins on next page)

# Journal of Environmental Sciences

## Virus occurrence in sources for drinking water production and in drinking water: a review --Manuscript Draft--

<b>Manuscript Number:</b>			
<b>Article Type:</b>	Review Article		
<b>Section/Category:</b>	Biology and microbiology		
<b>Keywords:</b>	drinking water; enteric virus; human health; microbial water quality; molecular methods; surface water		
<b>Corresponding Author:</b>	Marta Gea Università degli Studi di Torino: Università degli Studi di Torino Torino, (TO) Italy ITALY		
<b>First Author:</b>	Marco Panizzolo		
<b>Order of Authors:</b>	Marco Panizzolo Marta Gea Elisabetta Carraro Giorgio Gilli Silvia Bonetta Cristina Pignata		
<b>Abstract:</b>	<p>Microbiological quality of drinking water (DW) is crucial for Public Health. Many diseases linked to DW consumption are due to viruses. The aim of this review was to describe virus presence detected using molecular methods in sources for DW production and in DW. Four water types were considered: surface water used for DW production (SW-D), groundwater used for DW production (GW-D), water used for human consumption (DW) and bottled water (BW). The considered viruses were human pathogens; moreover plant pathogens proposed as novel viral indicators were presented. Studies published in the last 10 years were analysed and 79 articles were included in the review.</p> <p>Regarding virus occurrence in SW-D, GW-D, DW, high percentages of positive samples were reported for adenovirus, polyomavirus and pepper mild mottle virus. The most searched viruses were adenovirus, enterovirus, norovirus GI/GII and rotavirus. These viruses were frequently detected in SW-D, while they were rarely found in GW-D, suggesting that GW may be safer as a DW source. These viruses were detected also in DW, posing a possible threat for human health. Considering global occurrence, the lowest percentages of positive samples were found in Europe, while the highest percentages in Asia and South America. Only three articles assessed viruses in BW. Considering detection methods, filtration was the most applied concentration method, while nucleic acid extraction and molecular detection were generally performed using spin columns with silica membrane and quantitative PCR respectively.</p> <p>This review highlighted some critical issues such as method standardization lack and need for legislation updates.</p>		
<b>Suggested Reviewers:</b>	Marcello Iaconelli ISS: Istituto Superiore Di Sanita marcello.iaconelli@iss.it Research activity focused on environmental monitoring and human pathogenic viruses.	Mark A. Borchardt Marshfield Clinic Research Institute mark.borchardt@ars.usda.gov Expert in viruses in drinking water	Fernando R. Spilki FEEVALE University: Universidade FEEVALE fernandors@feevale.br

	expert in removal of viral agents in water and sewage and contamination of water, soil and foods by enteric viruses.
	Tahir Ahmad COMSATS University Islamabad Baig42@gmail.com Expert in environmental virology
	Walter Randazzo University of Valencia: Universitat de Valencia walter.randazzo@uv.es Expert in analytical and detection methods for virology, environmental contamination and public health
	Annalaura Carducci University of Pisa: Universita degli Studi di Pisa annalaura.carducci@unipi.it Expert in environmental virology, water quality and molecular biology.



UNIVERSITÀ DEGLI STUDI DI TORINO  
**DIPARTIMENTO DI SCIENZE DELLA SANITA' PUBBLICA E PEDIATRICHE**  
**DEPARTMENT OF PUBLIC HEALTH AND PEDIATRICS**

*DIRETTORE: PROF. SSA CARLA MARIA ZOTTI*  
Piazza Polonia, 94 – 10126 Torino (Italia)  
Codice Fiscale 80088230018 – P.IVA IT02099550010

February 22<sup>nd</sup>, 2022

Dear Editor,

We send the review “*Virus occurrence in sources for drinking water production and in drinking water: a review*” by Marco Panizzolo, Marta Gea, Elisabetta Carraro, Giorgio Gilli, Silvia Bonetta, Cristina Pignata on JOURNAL OF ENVIRONMENTAL SCIENCES.

Many studies showed that microbial water quality used for human consumption is critical for Public Health. Outbreaks linked to the consumption of contaminated or improperly treated water have been reported all over the world. Among all the waterborne pathogens, viruses are of major concern. Indeed, they can induce gastroenteritis through the faecal-oral route and, due to their peculiar characteristics compared to other pathogens, they are not efficiently removed by drinking water treatments. Virus presence in drinking water is among the main causes of death in developing countries and it induces a consistent percentage of drinking water outbreaks in high-income countries. Therefore, virus detection at all phases of the integrated water cycle, from wastewater to drinking water, has a key role for human health. However, a complete overview of viral occurrence in sources for drinking water production and in drinking water from all over the world is still lacking. The aim of the submitted review was to describe available data about virus occurrence in sources for drinking water production and in drinking water using molecular methods. Water types considered were: surface water used for drinking production, groundwater used for drinking water production, water used for human consumption (drinking water) and bottled water. Two virus types were considered: human pathogens and plant pathogens proposed as novel viral indicators. Scientific studies published in the last 10 years from all over the world were analyzed and 79 articles were finally included in the review.

We believe that the paper fits the aims and scope of the Journal, specifically, fits the following subjects:

- Aquatic environments
- Environmental microbiology

The study does not involve human subjects. All of the authors have read and approved the paper and it has not been published previously nor is it being considered by any other peer-reviewed journal. All authors are aware of and accept responsibility for the manuscript. All figures and tables were produced by the authors. Lastly, all authors declare no conflicting interests.

Hoping that the manuscript may fulfil the scientific standards of JOURNAL OF ENVIRONMENTAL SCIENCES, our best regards.

*Marta Gea and Co-authors*



UNIVERSITÀ DEGLI STUDI DI TORINO  
DIPARTIMENTO DI SCIENZE DELLA SANITA' PUBBLICA E PEDIATRICHE  
DEPARTMENT OF PUBLIC HEALTH AND PEDIATRICS

DIRETTORE: PROF. SSA CARLA MARIA ZOTTI  
Piazza Polonia, 94 – 10126 Torino (Italia)  
Codice Fiscale 80088230018 – P.IVA IT02099550010

**Virus occurrence in sources for drinking water production and in drinking water: a  
review**

Marco Panizzolo<sup>1#</sup>, Marta Gea<sup>1#\*</sup>, Elisabetta Carraro<sup>1</sup>, Giorgio Gilli<sup>1</sup>, Silvia Bonetta<sup>2</sup>, Cristina Pignata<sup>1</sup>

<sup>1</sup>Department of Public Health and Pediatrics, University of Torino, Piazza Polonia 94, 10126 Torino, Italy

<sup>2</sup>Department of Life Sciences and Systems Biology, University of Torino, via Accademia Albertina 13, 10123 Torino, Italy

#Marco Panizzolo and Marta Gea contributed equally to this work.

**\*Corresponding author:**

Marta Gea

marta.gea@unito.it

Department of Public Health and Pediatrics,

University of Torino,

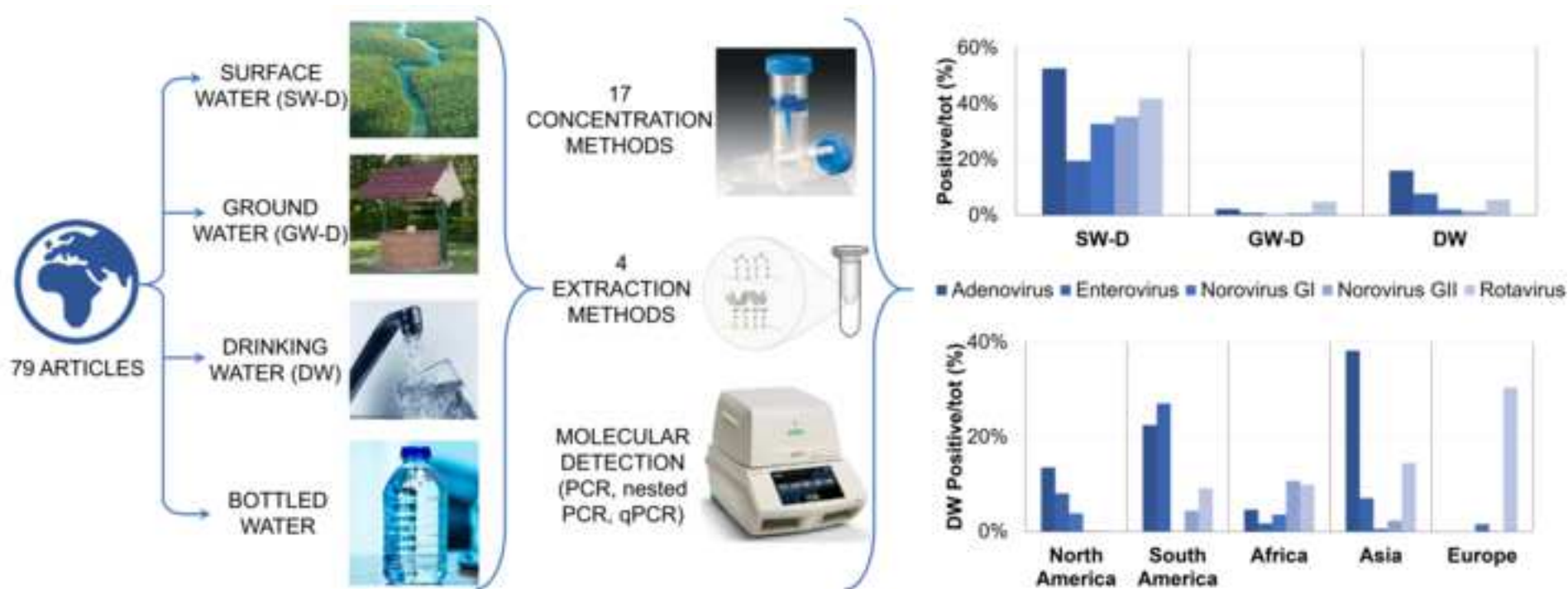
Piazza Polonia 94, 10126 Torino, Italy.

Phone: +39 011 6703190

Submitted to JOURNAL OF ENVIRONMENTAL SCIENCES

**Highlights (max 5, max 85 characters, including spaces, per bullet point)**

1. Adenovirus, enterovirus, norovirus, rotavirus were the most searched viruses
2. High % of positive samples for adenovirus, polyomavirus, pepper mild mottle virus
3. Viruses frequently detected in surface water, rarely in ground and drinking water
4. Viruses more detected in drinking water from Asia/South America than from Europe
5. Detection usually performed with filtration (negative filter) and quantitative PCR





UNIVERSITÀ DEGLI STUDI DI TORINO  
DIPARTIMENTO DI SCIENZE DELLA SANITA' PUBBLICA E PEDIATRICHE  
DEPARTMENT OF PUBLIC HEALTH AND PEDIATRICS

*DIRETTORE: PROF. SSA CARLA MARIA ZOTTI*  
Piazza Polonia, 94 – 10126 Torino (Italia)  
Codice Fiscale 80088230018 – P.IVA IT02099550010

**Virus occurrence in sources for drinking water production and in drinking water: a review**

Marco Panizzolo<sup>1#</sup>, Marta Gea<sup>1#\*</sup>, Elisabetta Carraro<sup>1</sup>, Giorgio Gilli<sup>1</sup>, Silvia Bonetta<sup>2</sup>, Cristina Pignata<sup>1</sup>

<sup>1</sup>Department of Public Health and Pediatrics, University of Torino, Piazza Polonia 94, 10126 Torino, Italy

<sup>2</sup>Department of Life Sciences and Systems Biology, University of Torino, via Accademia Albertina 13, 10123 Torino, Italy

#Marco Panizzolo and Marta Gea contributed equally to this work.

**\*Corresponding author:**

Marta Gea  
marta.gea@unito.it  
Department of Public Health and Pediatrics,  
University of Torino,  
Piazza Polonia 94, 10126 Torino, Italy.  
Phone: +39 011 6703190

**Other e-mail addresses:**

marco.panizzolo@unito.it

elisabetta.carraro@unito.it

giorgio.gilli@unito.it

silvia.bonetta@unito.it

cristina.pignata@unito.it



1 **Virus occurrence in sources for drinking water production and in drinking**  
2 **water: a review**

3 Marco Panizzolo<sup>1#</sup>, Marta Gea<sup>1#\*</sup>, Elisabetta Carraro<sup>1</sup>, Giorgio Gilli<sup>1</sup>, Silvia Bonetta<sup>2</sup>, Cristina  
4 Pignata<sup>1</sup>

5 <sup>1</sup>Department of Public Health and Pediatrics, University of Torino, Piazza Polonia 94, 10126  
6 Torino, Italy

7 <sup>2</sup>Department of Life Sciences and Systems Biology, University of Torino, via Accademia  
8 Albertina 13, 10123 Torino, Italy

9 #Marco Panizzolo and Marta Gea contributed equally to this work.

10

11 \*Corresponding author:

12 Marta Gea

13 marta.gea@unito.it

14 Department of Public Health and Pediatrics,

15 University of Torino,

16 Piazza Polonia 94, 10126 Torino, Italy.

17 Phone: +39 011 6703190

18

19 **ABSTRACT (max 250 words)**

20 Microbiological quality of drinking water (DW) is crucial for Public Health. Many diseases  
21 linked to DW consumption are due to viruses. The aim of this review was to describe virus  
22 presence detected using molecular methods in sources for DW production and in DW. Four  
23 water types were considered: surface water used for DW production (SW-D), groundwater  
24 used for DW production (GW-D), water used for human consumption (DW) and bottled water  
25 (BW). The considered viruses were human pathogens; moreover plant pathogens proposed  
26 as novel viral indicators were presented. Studies published in the last 10 years were  
27 analysed and 79 articles were included in the review.

28 Regarding virus occurrence in SW-D, GW-D, DW, high percentages of positive samples  
29 were reported for adenovirus, polyomavirus and pepper mild mottle virus. The most searched  
30 viruses were adenovirus, enterovirus, norovirus GI/GII and rotavirus. These viruses were  
31 frequently detected in SW-D, while they were rarely found in GW-D, suggesting that GW may  
32 be safer as a DW source. These viruses were detected also in DW, posing a possible threat  
33 for human health. Considering global occurrence, the lowest percentages of positive samples  
34 were found in Europe, while the highest percentages in Asia and South America. Only three  
35 articles assessed viruses in BW.  
36 Considering detection methods, filtration was the most applied concentration method, while  
37 nucleic acid extraction and molecular detection were generally performed using spin columns  
38 with silica membrane and quantitative PCR respectively.  
39 This review highlighted some critical issues such as method standardization lack and need  
40 for legislation updates.

41

42 **Keywords (max 6):** drinking water, enteric virus, human health, microbial water quality,  
43 molecular methods, surface water.

44

45 **Abbreviations:**

46 AdV = adenovirus

47 AiV = aichivirus

48 AstV = astrovirus

49 BW = bottled water (water used for human consumption)

50 DW = drinking water (water used for human consumption, not bottled)

51 EPA = Environmental Protection Agency

52 EV = enterovirus

53 GW = groundwater

54 GW-D = groundwater used as a source for DW production

55 HAV = hepatitis A virus  
56 HEV = hepatitis E virus  
57 NoV = norovirus  
58 PMMoV = pepper mild mottle virus  
59 PyV = polyomavirus  
60 RoV = rotavirus  
61 SW = surface water  
62 SW-D = surface water used as a source for DW production  
63 SaV = sapovirus  
64 TMV = tobacco mosaic virus  
65 TTV = torque teno virus  
66 PCR = polymerase chain reaction  
67 qPCR = quantitative PCR  
68 QMRA = Quantitative Microbial Risk Assessment  
69 WSP = Water Safety Plan

70

71 **Contents:**

72 1. INTRODUCTION  
73 2. SEARCH CRITERIA  
74 3. VIRUS TYPES AND CHARACTERISTICS IN SOURCES FOR DW PRODUCTION  
75 AND IN DW  
76 3.1 Human pathogens  
77 3.2 Plant pathogens proposed as novel viral indicators  
78 4. VIRUS OCCURRENCE IN SOURCES FOR DW PRODUCTION AND IN DW  
79 4.1 Virus occurrence in SW-D, GW-D, DW  
80 4.2 Comparison of virus occurrence among the water types  
81 4.3 Comparison of virus occurrence in DW among continents  
82 4.4 Virus occurrence in BW

83 5. METHODS FOR VIRUS CONCENTRATION AND DETECTION IN SOURCES FOR  
84 DW PRODUCTION AND IN DW  
85 5.1 Virus concentration methods  
86 5.2 Virus detection methods  
87 6. CONCLUSIONS  
88 7. FUNDING  
89 8. ACKNOWLEDGEMENTS  
90 9. DECLARATION OF INTERESTS  
91 10. REFERENCES

92

## 93 **1. INTRODUCTION**

94 Fresh water is an essential resource for life on our planet and just a small part is accessible  
95 because the most one is present in aquifers or in form of ice. Water scarcity increases and  
96 so its reuse is essential (Cocoran et al., 2010). Moreover, water quality as well as water  
97 quantity is important. Nowadays, 20% of world's population have no access to drinking water  
98 (DW) and 40% suffer the consequences of improperly treated water (Cocoran et al., 2010;  
99 Seelen et al., 2019).

100 Microbiological water quality used for human consumption is considered by many studies to  
101 be critical for Public Health. Outbreaks linked to the DW consumption have been reported  
102 worldwide and the main causes for contamination were identified as intrusion of animal  
103 faeces or wastewaters due to heavy rain in groundwaters (GWs), wastewaters discharge into  
104 the DW source, malfunctioning of the disinfection equipment at DW treatment plants and  
105 cross-connections, pipe breaks and wastewater intrusion into the distribution system (Ligon  
106 and Bartram, 2016; Moreira and Bondelind, 2017). Waterborne outbreaks were reported, for  
107 examples, in China (Shang et al., 2017; Xue et al., 2014), Denmark (Van Alphen et al.,  
108 2014), Albania (Donia et al., 2011), Spain (Blanco et al., 2017), Switzerland (Breitenmoser et  
109 al., 2011), Italy (Giammanco et al., 2014), Philippines (Rebato et al., 2019), India (Tripathy et  
110 al., 2019) and United States (Beer et al., 2015). In these studies, through retrospective

111 investigations and environmental analyses, pathogenic viruses were hypothesised as  
112 possible causative agents.

113 Viruses can induce viral gastroenteritis through the faecal-oral route. In developing countries  
114 diarrhoeal diseases due to virus presence in DW are one of the main causes of death  
115 (Fayomi et al., 2019; WWAP, 2017). Similarly, in high income countries data concerning DW  
116 outbreaks show that in 2013-2014 7% of outbreaks in the United States were caused by  
117 viruses (CDC, 2021), while in the European Union Member States in 2019 most of DW  
118 outbreaks with strong-evidence were related to norovirus (NoV) and other calicivirus (ECDC,  
119 2021).

120 Viruses are naturally present in environmental matrices such as in water where their  
121 presence can be promoted by the discharge of not properly treated wastewater (Gibson et  
122 al., 2011; Masciopinto et al., 2019; Okoh et al., 2010; Upfold et al., 2021). Moreover, several  
123 studies have shown that DW treatments do not always succeed in removing viruses (Kato et  
124 al., 2018; Salvador et al., 2020; Ye et al., 2012); therefore, detection of viruses at all phases  
125 of the integrated water cycle (from wastewater to DW) has a key role for human health.

126 In literature, virus presence within wastewaters have been investigated by many reviews  
127 (Bhatt et al., 2020; Corpuz et al., 2020; Foladori et al., 2020; Sano et al., 2016), whereas  
128 virus occurrence in DW has been considered by just few reviews which were focused on DW  
129 treatment systems and DW related outbreaks (Chen et al., 2021; Moreira and Bondelind,  
130 2017). An overview of viral presence in water used as a source for DW production and in DW  
131 is still lacking. Consequently, the aim of this review is to report the recent available  
132 knowledge about virus occurrence in sources for DW production and in DW. Water types  
133 considered were surface water used for DW production (SW-D), GW used for DW production  
134 (GW-D), DW and bottled water (BW). Moreover, two virus types were considered: human  
135 pathogens and plant pathogens proposed as novel viral indicators. Scientific studies  
136 published in the last 10 years from all over the world were analysed and data of virus  
137 presence assessed using molecular methods were summarized and discussed. In addition,  
138 virus characteristics, concentration methods, nucleic acid extraction and molecular detection

139 techniques reported in these studies were detailed. To the best of our knowledge, this is the  
140 first review that describes the most recent data on the worldwide virus occurrence in water  
141 used as sources for DW production and in water used for human consumption (DW).

142

## 143 **2. SEARCH CRITERIA**

144 In order to find information about the virus presence in sources for DW production and in  
145 DW, a literature search was performed in PubMed, Scopus and Web of Science. These  
146 databases were selected to be the most relevant and used for research on environmental  
147 topics. The search terms “virus” and “presence” or “detection” were combined with “drinking  
148 water” or “bottled water” or “mineral water”. Article search was set in the last 10 years and  
149 were chosen only articles published between 2011 and 2021. The search gave 798 results in  
150 PubMed, 379 results in Scopus and 367 results in Web of Science (total = 1,544 results).

151 Two authors of the review independently screened the 1,544 publications. Using PRISMA  
152 approach, 79 articles were finally included in this review (Fig. 1).

153 The search was limited to environmental monitoring articles that analysed human pathogens  
154 or plant pathogens proposed as novel viral indicators. The articles were included when they  
155 were written in English and met the following criteria: i) the analysed water type was water  
156 used as a source for DW production or water used as DW, ii) origin of sources for DW  
157 production was reported (surface water-SW or GW), iii) the detection of viruses was  
158 performed using molecular methods, iv) viruses were not spiked intentionally into the  
159 samples, v) data could be extrapolated for each viral agent and for each water type.

160

## 161 **3. VIRUS TYPES AND CHARACTERISTICS IN SOURCES FOR DW**

### 162 **PRODUCTION AND IN DW**

163 Viruses are obligatory intracellular parasites able to spread and be environmentally  
164 transmitted through air, inert surfaces or waters. One of the main vehicles of viral  
165 transmission is water through faecal-oral route. Inevitably, all water types can be subject to  
166 contamination starting with SW (rivers, lakes), GW (wells, springs) and finally the seas and

167 oceans. Therefore, it is essential to study virus's resistance within these matrices (Pinon and  
168 Vialette, 2019; Shoham et al., 2012).

169 Without host cells, viruses may decrease in number or remain stable. Their reduction can  
170 occur depending on the water type (SW, GW, DW) and on various environmental conditions  
171 such as temperature, sunlight (UV) and disinfection products (chlorine and derivatives).

172 Water type influences the persistence of viral agents. In fact, GW, unlike SW, is a more  
173 stable environment with few changes in chemical and physical parameters over time, and  
174 thus constitutes a favourable matrix (Espinosa et al., 2008). Temperature is one of the most  
175 studied environmental condition and has been recognised as the most influential factor that  
176 affect viral persistence (Espinosa et al., 2008; Pinon and Vialette, 2019).

177 In general, it has been shown that most viral agents can survive for years at low  
178 temperatures, whereas at higher temperatures viruses are reduced within a few days. In  
179 some studies, resistance to different temperature ranges was tested for certain viral agents  
180 in different water matrices. A reduction of 5 log units of polioviruses and echoviruses was  
181 found after one month in ocean water at around 21-26°C, while the same reduction was seen  
182 after over 2 months when ocean water had temperatures between 4 and 16°C. Moreover, in  
183 mineral waters 1 log unit decrease of poliovirus and hepatitis A virus (HAV) was  
184 demonstrated in about 11 months at 4°C, in contrast, the same reduction at 23°C took about  
185 1 month (Pinon and Vialette, 2019). Finally, Ogorzaly et al. (2010) study, carried out on DW  
186 and GW, showed a decrease of 2-1 log units of astrovirus (AstV) and adenovirus (AdV) with  
187 increasing water temperature from 4 to 20°C in half to a third of the time, respectively.

188 Another important factor that significantly influences viral viability is sunlight (UV). It is well  
189 known that UV promotes a significant reduction in viral particles, indeed the study by  
190 Flannery et al. (2013) showed a reduction of 1 log unit after 4 hours exposure to light  
191 simulating winter conditions (10°C), while the same reduction was induced by 15 minutes  
192 exposure to light simulating summer conditions (17°C). UV effectiveness is also confirmed by  
193 Garver et al. (2013) study that showed a reduction of 2-3 logs in 3 hours in deep water (less  
194 UV) compared to 3-4 logs in 1.5 hours in superficial water (more UV).

195 Other factors responsible for viral reduction may be the presence of disinfectants, pH  
196 extremes, or heavy metals (Pinon and Vialette, 2019).

197 In parallel to the environmental factors, some viral characteristics can affect viral survival  
198 such as aggregation tendency, genome type and capsid composition. Actually, under  
199 adverse conditions, viruses tend to aggregate with each other and with organic matter to  
200 form aggregate suspension material, making their elimination difficult. Indeed, disinfectants  
201 are unable to penetrate the viral capsid so merely bind to the biofilm of organic matter in the  
202 aggregates (Espinosa et al., 2008). Moreover, viruses are characterized by a different  
203 persistence according to their genome type. For example, due to its double-stranded DNA  
204 genome, AdV showed greater stability in water than enterovirus (EV) which has RNA  
205 genome (Mena and Gerba, 2008). Due to the higher persistence in the environment, AdV  
206 has been suggested as a possible indicator of the viral contamination water (Verani et al.,  
207 2019). Finally, a comparison of rotavirus (RoV) and AstV, both of which are RNA viruses,  
208 showed that RoVs are more persistent in GW, as they have a triple-layer capsid and a  
209 double-stranded RNA genome, whereas AstV have a single-layer capsid and a single-  
210 stranded RNA genome (Espinosa et al., 2008).

211 In the 79 included articles, virus analysis in sources for DW production and in DW was  
212 mainly focused on the detection of two virus types namely human pathogens and indicators.  
213 The first viruses are able to infect hosts causing diseases, while the second ones are used  
214 as indicators of faecal contamination. The articles included in this review mainly investigated  
215 the following viruses: AdV, HAV, EV, aichivirus (AiV), hepatitis E virus (HEV), sapovirus  
216 (SaV), NoV, torque teno virus (TTV), RoV, AstV, polyomavirus (PyV), pepper mild mottle  
217 virus (PMMoV) and tobacco mosaic virus (TMV). The characteristics of these viruses are  
218 described in Table 1.

219

### 220 **3.1 Human pathogens**

221 The integrated water cycle plays a key role in reduction and elimination of pathogenic  
222 viruses, decreasing their spread in environment. Wastewater treatments are often unable to



223 eliminate viruses and fail to prevent their release in the environment; moreover, pathogenic  
224 viruses could be also resistant to DW treatments threatening Public Health. Among the  
225 human pathogens, enteric viruses are obligate parasites that infect and replicate within the  
226 human gastrointestinal tract (Upfold et al., 2021). Depending on the type of viral agent  
227 colonising the gastrointestinal tract, different possible diseases may occur. Enteric viruses  
228 are one of the main causes of waterborne diseases transmissible via the faecal-oral route so  
229 their detection in water used as DW source or in DW is of crucial importance in order to  
230 assess the risk for human health.

231

### 232 **3.2 Plant pathogens proposed as novel viral indicators**

233 Currently the monitoring of microbial water quality is generally performed quantifying  
234 bacterial indicators of faecal contamination. Since they are easy to identify, more present and  
235 resistant than pathogens, bacterial indicators are used to assess the faecal contamination  
236 and to estimate the presence of microbial pathogens. However, numerous studies showed  
237 that the concentration of indicator bacteria is not related to the concentration of pathogenic  
238 viruses, suggesting that these indicators are unsuitable to define the presence of human viral  
239 pathogens in water (Liang et al., 2015). Indeed, environmental conditions affect differently  
240 bacteria and viruses (Kitajima et al., 2018).

241 Bacteriophages (e.g. coliphages) have been proposed as alternative indicators instead of  
242 bacteria. Coliphages are viruses that infect *Escherichia coli* and other coliforms (Leclerc et  
243 al., 2000). Environmental transport and survival of coliphages is similar to enteric viruses.  
244 However, coliphages show a greater persistence than human enteric viruses in environment  
245 since their replication in bacterial hosts can continue after being shed in faeces. In addition,  
246 only a small percentage of human or animal faecal samples test positive for coliphages so  
247 these viruses may be too sparse to be detected in some environmental waters (Griffin et al.,  
248 2008). Therefore, other viruses were suggested by the scientific community as possible viral  
249 indicators of faecal contamination. In particular, two plant pathogens were proposed as

250 alternative viral faecal indicators: PMMoV and TMV (Kitajima et al., 2018; Tandukar et al.,  
251 2020a). These two viruses were analysed by some research articles included in this review.  
252 PMMoV is a plant pathogen globally distributed that causes significant economic and crop  
253 losses worldwide (e.g. in the United States, Japan and China). Its presence may be  
254 indicative of faecal contamination because is the most abundant virus type in human faecal  
255 samples (Kitajima et al., 2018). However, its application as a viral indicator has limitations,  
256 since studies show conflicting results on the correlation between concentrations of this virus  
257 and concentrations of human enteric viruses (Kitajima et al., 2018; Tandukar et al., 2020a).  
258 As PMMoV, TMV is a plant pathogen. TMV was discovered in the 19<sup>th</sup> century when a new  
259 infection was affecting tobacco plants causing characteristic patterns, such as mosaic-like  
260 mottling and discoloration on the leaves (Tandukar et al., 2020a). Similarly to PMMoV, this  
261 virus is excreted by a large proportion of healthy people.  
262 PMMoV and TMV are widely distributed in SW, in GW and even in DW. They are used as  
263 indicators of faecal contamination in wastewater, SW and also in DW because their presence  
264 is high in human faeces and in sewage. In the analysed articles the presence of these  
265 viruses was studied in sources for DW production and in DW (Haramoto et al., 2013; Kuroda  
266 et al., 2015; Tandukar et al., 2020a, 2018).

267

#### 268 **4. VIRUS OCCURRENCE IN SOURCES FOR DW PRODUCTION AND IN DW**

##### 269 **4.1 Virus occurrence in SW-D, GW-D, DW**

270 In the 79 included articles, water samples coming from all over the world were analysed (Fig.  
271 2). In particular, 24 articles analysed samples from Asia, 17 from South America, 13 from  
272 Africa, 13 from Europe, 11 from North America and 1 from Oceania (79 total articles). The  
273 articles assessed virus occurrence in three different water types.

- 274 ● SW-D was analysed by 43 articles, SW-D was collected from rivers, estuarine bays,  
275 dams, lagoons, ponds, lakes and other reservoirs.
- 276 ● GW-D was analysed by 17 articles. GW-D was collected from wells and springs.

277 • DW was analysed by 45 articles. These articles analysed different water types (e.g.  
278 tap water, DW treatment plant effluents, SW used as DW without any treatment).

279 The mean of sample volume analysed was significantly different according to the water types  
280 (Kruskal-Wallis test followed by pairwise comparisons, SW-D vs GW-D, SW-D vs DW, GW-D  
281 vs DW,  $p < 0.05$ ). Mean values were  $43.58 \pm 114.83$  L ranging from 0.050 L to 2340 L for SW-  
282 D,  $321.91 \pm 407.99$  L ranging from 0.250 L to 1783 L for GW-D and  $242.05 \pm 467.26$  L  
283 ranging from 0.050 L to 3400 L for DW. In particular, volumes were higher for GW-D/DW  
284 samples than SW-D probably because a lower viral presence was expected.

285 Table 2 presents the cumulative percentages of positive samples for each viral agent (total  
286 positive samples/total samples). For NoV the cumulative percentages were calculated  
287 dividing data according to the viral subtype (cumulative percentages were calculated  
288 independently for NoV GI, NoV GII, NoV GIII, NoV GIV).

289 The percentages of positive samples were compared among virus types. For some viral  
290 types a small number of samples was analysed, therefore the percentages could not reflect  
291 the real occurrence of these viruses in sources for DW production and in DW. For this  
292 reason, comparison was performed considering viruses that were analysed in at least 100  
293 samples. The highest percentages of positive samples were found for PMMoV (85.31%),  
294 AdV (52.61%), PyV (44.25%), AiV (43.86%), RoV (41.81%) in SW-D, for PMMoV (5.94%),  
295 RoV (4.88%), AdV (2.21%), PyV (0.97%), EV (0.78%) in GW-D, for PMMoV (28.33%), AdV  
296 (15.96%), PyV (12.94%), AiV (11.27%), EV (7.73%) in DW.

297 AdV and PyV were among the human pathogenic viruses that showed the highest  
298 percentages of positive samples. This result can be explained considering that AdV and PyV  
299 are characterized by a DNA genome which is generally more stable in the environment and  
300 less affected by the physico-chemical treatments applied to obtain DW with respect to RNA  
301 genome (Ye et al., 2012).

302 For the three water types, the percentages of PMMoV positive samples were the highest  
303 compared to the percentages of the other viruses. This finding is interesting since PMMoV  
304 has been proposed as a possible viral indicator of human faecal contamination in several

305 studies (Kitajima et al., 2018; Zhang et al., 2006). Indeed, the high percentages of positive  
306 samples found in SW-D, GW-D and DW confirm that this virus is more persistent in water  
307 than other enteric viruses, including AdV and PyV (Hamza et al., 2011; Haramoto et al.,  
308 2013), suggesting that it could be an excellent candidate as an indicator and it could be used  
309 as a possible process control to measure the removal of enteric viruses during water  
310 treatments (Symonds et al., 2018). The higher presence of this virus with respect to human  
311 pathogenic viruses could also be due to the fact that, while other enteric viruses are more  
312 abundant in water when there is an increase of infected individuals, PMMoV presence seems  
313 not to be characterized by seasonal variations (Haramoto et al., 2013).  
314 Virus detection in SW-D, GW-D, DW is reported divided by reference in Table S.1, S.2, S.3,  
315 respectively.

316

#### 317 **4.2 Comparison of virus occurrence among the water types**

318 The percentages of positive samples were compared among the water types. The  
319 comparison was performed considering viruses that were searched in the highest number of  
320 samples and that were analysed by most articles (i.e. the most searched viruses for number  
321 of total samples and number of total articles). This choice was adopted to compare data that  
322 could reflect the real virus occurrence and could be considered representative of the global  
323 situation. As can be seen in Table 2, AdV, EV, NoV GI, NoV GII and RoV were most  
324 searched viruses in all water types. These viruses were the most searched probably because  
325 are important foodborne pathogens (Koopmans and Duizer, 2004).

326 In Fig. 3 are reported the percentages of positive samples in SW-D, GW-D and DW of these  
327 viruses. As can be seen, all the five viruses were frequently detected in SW-D. This result  
328 could be explained considering that these enteric viruses are excreted in large quantities in  
329 the faeces of infected individuals (symptomatic and asymptomatic), which are conveyed to  
330 sewage treatment plants. Since the water treatments of these plants can be not efficient to  
331 remove all viruses, they may be released into SW (Bhatt et al., 2020). Moreover, the high  
332 presence of enteric viruses in SW is not only due to municipal wastewaters but may also

333 result from livestock slurry from livestock farms, which are sometimes not conveyed to the  
334 wastewater treatment plants but directly discharged in SW (Haramoto et al., 2018).  
335 Among the water types, the percentages of positive samples in GW-D samples were the  
336 lowest. This result suggests that GWs are more protected from possible sources of  
337 contamination, making them safer when they are used to produce DW. Nevertheless, GW, if  
338 not properly protected, are susceptible and can easily be polluted from some contamination  
339 sources. After a period of heavy rainfall, GW located in proximity to livestock farms can be  
340 contaminated by livestock slurry leaching into the ground or due to damage or deficiency of  
341 pipes conveying wastewater effluents to the plants (Gibson and Schwab, 2011a; Gotkowitz  
342 et al., 2016). Percentages of positive samples in GW-D were also lower than in DW. This  
343 result is not surprising considering that the DWs include not only treated GWs but also  
344 treated SWs.

345 The percentages of positive samples were higher in SW-D than in DW. This may be  
346 attributable to the fact that DW are generally treated with physico-chemical processes which  
347 can reduce viral presence in this water type (Asami et al., 2016; Atabakhsh et al., 2019;  
348 Jacob et al., 2015; Kato et al., 2018; Tandukar et al., 2020b; Ye et al., 2012).

349 Even if at lower percentages compared to SW-D, the five enteric viruses were detected also  
350 in DW. Since high percentages of positive samples in water used for human consumption  
351 may be a source of risk to the population, the presence of these viruses in DW might pose a  
352 possible threat to human health. Indeed, the ingestion of water contaminated by enteric  
353 viruses can lead to sporadic episodes of viral gastroenteritis, which, if not treated with  
354 appropriate care, could lead to death in children (Wang et al., 2016). It is important to  
355 highlight that in this review were presented only data of virus presence analysed using  
356 molecular methods; therefore, the percentages of positive samples do not necessarily mean  
357 that these samples contain active and pathogenic viruses but only that in these samples the  
358 viral genomic material was detected (Rachmadi et al., 2016). Indeed, many studies  
359 compared virus infectivity and virus detection using molecular methods in water samples and

360 found that samples in which viral genomes were detected did not always contain infectious  
361 viral particles (Iaconelli et al., 2017; Salvador et al., 2020).

362

### 363 **4.3 Comparison of virus occurrence in DW among the continents**

364 Considering virus detection in DW samples, the percentages of positive samples were  
365 compared among the continents. As for the comparison among the water types, the  
366 comparison was performed considering the most searched viruses for number of total  
367 samples and number of total articles (see paragraph 4.2). In Fig. 4 are reported the  
368 percentages of positive sample in DW samples divided according to continents. It should be  
369 noted that the number of studies is not the same across continents. Indeed, there are fewer  
370 studies in Europe than in the other continents. The global distribution of the samples is  
371 probably not homogenous because in some continents such as Europe the risk associated  
372 with water consumption is not considered a major health concern, so the research articles  
373 focused on this topic are limited. On the contrary, in developing countries diseases  
374 associated with water consumption are a major issue, thus this research topic is more  
375 investigated. Comparing the percentages among the continents, except for RoV, the lowest  
376 percentages of positive samples were found in Europe. In contrast, the highest percentages  
377 of positive samples were found in Asia and South America.

378 The different virus occurrence in Europe with respect to Asia and South America could be  
379 due to several factors. Indeed, in developing countries the quality of sources for DW  
380 production could be lower due to a higher discharge of not properly treated wastewaters;  
381 moreover, technologies used for DW treatment could be less efficient in virus removal.  
382 Finally, water distribution networks could be less monitored and more prone to breakdowns  
383 that may cause the intrusion of contaminated water in DW distribution systems. Regarding  
384 RoV, the unexpected percentage of positive samples in Europe could be explained  
385 considering that only one study carried out in Slovenia assessed RoV occurrence in  
386 European DW (Steyer et al., 2011), so this percentage could be not representative of the  
387 whole European occurrence of this virus.

388 The results obtained analysing African samples are interesting. Indeed, although the number  
389 of articles is higher than in Europe and the articles analysed samples coming from different  
390 African countries (giving a complete picture of virus occurrence throughout the continent), the  
391 percentages of positive samples were less than 11%. These percentages seem to be too low  
392 if compared with the incidence of viral gastrointestinal diseases transmitted by water  
393 consumption in this continent, so further studies are needed to clarify this discrepancy.  
394 Finally, considering the occurrence of viruses in North America, the percentages of positive  
395 samples were quite low suggesting that the DW quality is quite good in this continent.

396

#### 397 **4.4 Virus occurrence in BW**

398 Three articles assessed the virus presence in BW (Da Silva Luz et al., 2020; Dos Santos et  
399 al., 2015; Kuroda et al., 2015). The analysed BW was produced using GW as source (water  
400 from wells/springs) and samples were collected in Brazil and Vietnam from bottles containing  
401 different water volumes (0.5, 1.5, 19, 20 L). The analysed water volume ranged from 0.5 L to  
402 100 L, while the percentages of positive samples ranged from 81.69% (AdV) to 0% (PMMoV,  
403 NoV GII, AiV). It's important to highlight that these percentages of positive samples were  
404 calculated considering only three articles which analysed few samples collected in Brazil (2  
405 articles) and in Vietnam (1 article). The limited number of articles on virus occurrence in BW  
406 is probably due to the low frequency of outbreaks linked to the consumption of BW. However,  
407 since also in this water type genomes of some viruses were found (e.g. AdV, EV, NoV, RoV),  
408 more studies to assess the real virus occurrence in BW are needed. Virus detection in BW is  
409 reported in Table S.4.

410

## 411 **5. METHODS FOR VIRUS CONCENTRATION AND DETECTION IN SOURCES**

### 412 **FOR DW PRODUCTION AND IN DW**

#### 413 **5.1. Virus concentration methods**

414 In the analysed studies, different methods/methodologies of concentration, extraction and  
415 identification of viral particles were reported. Many of these concentration methods were

416 established in the 1980s and have not been changed. They include the use of negatively and  
417 positively charged membranes, glass wool filters and ultrafiltration. These methods can be  
418 applied alone or can be followed by a secondary concentration that allows for a higher  
419 concentration of the treated water sample (Ikner et al., 2012).

420 Concentration methods used in the analysed articles are described below.

- 421 • Adsorption-elution method, also known as filtration method, is based on the  
422 absorption of organisms on a solid membrane utilising the ionic properties of the  
423 micro-organisms (bacteria/viruses) to be concentrated. The filters mainly used in this  
424 technique can be membrane filters (cellulose) or glass filters; moreover, filters could  
425 be with neutral charge or could have electropositive/electronegative charge using  
426 electrostatic forces to concentrate viruses (Cashdollar and Wymer, 2013; Ikner et al.,  
427 2012). The adsorption phase (with filters) is followed by an elution phase using a  
428 specific fluid which is variable according to the analysed virus type (Cai et al., 2015;  
429 Ruhanya, 2016). For instance, the Environmental Protection Agency (EPA) has  
430 proposed a procedure to detect human enteric viruses in water whose first step is  
431 based on adsorption-elution method (i.e. filtration through electropositive filters,  
432 followed by elution using a solution of glycine and beef extract) (Fout et al., 2015).
- 433 • Tangential flow filtration system consists in flowing the liquid parallel to the filtering  
434 medium to reduce the probability of clogging of the latter and thus enhance its filtering  
435 capacity. This method is still used today to concentrate micro-organisms present in a  
436 matrix (e.g. water). It is essential to adopt an appropriate membrane according to the  
437 type of microbial agent researched (Cai et al., 2015). In the analysed studies, 30 kDa  
438 and 100 kDa filter membranes were used.
- 439 • Ultrafiltration is commonly used as water treatment technology for the removal of  
440 human pathogens and can be considered as a special form of filtration that uses  
441 positive pressure to promote the flow of water through a membrane (Reeve et al.,  
442 2016). This method allows to retain not only particles and macromolecules but also



443 micro-organisms such as viruses and bacteria. The membranes used in ultrafiltration  
444 process have pores with diameters ranging from 1 to  $10^{-3}$   $\mu\text{m}$  (Shao et al., 2011).

- 445 • Polyethylene glycol is a biocompatible polymer used for protein precipitation. Its  
446 properties promote virus precipitation sequestering water molecules from the outer  
447 layer of their pericapsids/capsids to promote virus-virus interactions and thus virus  
448 concentration (Corpuz et al., 2020).
- 449 • Skimmed milk flocculation is based on three physical processes, i.e. adsorption,  
450 sedimentation and dissolution. The first two steps consist in the adsorption of viruses  
451 on pre-flocculated skimmed milk proteins and precipitation of flakes with adsorbed  
452 viruses. After sedimentation, sediment is dissolved using a buffer solution. This  
453 methodology does not require the use of special equipment and long processing  
454 steps, making its use advantageous (Corpuz et al., 2020).

455 In the 79 articles analysed, 7 different primary concentration methods were used, which were  
456 or were not followed by other 4 types of secondary concentration methods for a total of 17  
457 different combinations of primary-secondary concentration methods (Table 3). The most  
458 frequently used combinations of primary-secondary methods were filtration with negatively  
459 charged membranes (applied in 17 articles), filtration with negatively charged membranes  
460 followed by a secondary concentration using ultrafiltration (applied in 11 articles), filtration  
461 with positively charged membranes (applied in 10 articles), filtration with positively charged  
462 membranes followed by a secondary concentration with polyethylene glycol (applied in 7  
463 articles). The other methods were reported in less than 6 articles. The filtration with  
464 negatively charged membranes was the most applied method probably because it has  
465 numerous advantages. Indeed, this method is cheap and it allows high recoveries for  
466 viruses. Moreover, since electronegative filters are less influenced by clogging, this method  
467 is suitable also for turbid waters (Cashdollar and Wymer, 2013).

468

469

470

## 471 **5.2 Virus detection methods**

472 Virus detection can be performed using both cell culture and molecular methods. Cell culture  
473 methods are based on virus ability to grow in cell cultures causing morphological changes.  
474 Viruses are inoculated on cell monolayers and the number of infectious viruses is quantified  
475 counting the plaque forming units (cell destruction areas caused by viruses) (Gerba et al.,  
476 2017). Cell culture methods are able to quantify viruses that potentially can replicate in  
477 humans causing the disease. Before the development of molecular methods, virus detection  
478 in environmental matrices was exclusively performed using these methods (Gerba and  
479 Betancourt, 2019). However, the main disadvantage of cell culture methods is that they can  
480 not be applied to analyse all virus types because some of them, such as NoV, can not  
481 replicate in cell culture (Fout et al., 2015); therefore, in order to assess the presence of these  
482 viruses in environmental matrices, only molecular methods can be applied. Moreover, these  
483 detection methods seem to underestimate virus concentration at least by 2-3 orders of  
484 magnitude (Chen et al., 2021). The underestimation could be due to viral aggregation;  
485 indeed, many aggregated viruses can form a single plaque forming unit so they can be  
486 counted as one infectious viral particle. Moreover, one group of viruses may grow faster than  
487 another or interfere with the replication of another group of viruses, causing an  
488 underestimation of the viral particle number (Gerba and Betancourt, 2019).  
489 Therefore, nowadays, virus detection in waters is generally performed using molecular  
490 methods, which are based on the detection of viral genomes. Due to the low environmental  
491 stability of genomes, especially for RNA viruses a positive molecular result indicates that viral  
492 particles are intact; however, this result can be obtained also for viruses that have been  
493 inactivated by chemical disinfection, heat or proteases (Kopecka et al., 1993). As a  
494 consequence, one of the most important disadvantages of molecular methods is that they  
495 can not distinguish between infectious and inactivated viruses. On the contrary, these  
496 methods have numerous advantages. They are able to detect low virus concentrations so  
497 they are more sensitive than cell culture methods. In addition, they are characterized by high  
498 specificity and rapidity. Finally, in contrast to cell culture methods, they potentially allow the

499 detection of all virus types, detecting also viruses that are hardly propagated using cell  
500 cultures such as RoV and NoV (Carducci et al., 2003; Corpuz et al., 2020). Detection of viral  
501 genomes is performed through extraction of nucleic acids followed by amplification of specific  
502 nucleic acid fragments using polymerase chain reaction (PCR).

503 In the 79 analysed articles, different methods for nucleic acid extraction were reported. In  
504 some articles more than one extraction type was used. The different methods are described  
505 below.

- 506 ● Nucleic acids can be purified through the binding with silica membrane. The principle  
507 of this method is the following. DNA binds specifically to the silica-gel membrane,  
508 while contaminants pass through. Then unwanted materials are generally removed  
509 with washing steps and finally the remaining nucleic acids are eluted in either water  
510 or a buffer. This extraction type can be performed using both spin columns or vacuum  
511 columns. This method was the most applied for nucleic acid extraction. Indeed 71  
512 articles applied it using spin columns, while one using vacuum columns.
- 513 ● Magnetic beads separation is a method based on specific interaction between nucleic  
514 acids and magnetizable particles. Briefly, after a lysis step to release the nucleic  
515 acids, viral genomes bind to magnetizable particles in the presence of a binding  
516 buffer. The other molecules are washed with a water-based wash buffer and finally  
517 the nucleic acids are eluted in an elution buffer (Nargessi and Ou, 2010). This  
518 extraction type was applied by 13 articles.
- 519 ● Nucleic acids can be purified through the binding with glass fibre or glass powder. For  
520 example, nucleic acids can be immobilized through the binding to the surface of the  
521 glass fibre fleece in the presence of a chaotropic salt. Sample is mixed with a  
522 chaotropic salt and applied to the glass fibre fleece. Nucleic acids bind to the glass  
523 fleece, while contaminating substances are removed through washing steps. Nucleic  
524 acids are finally eluted in a small volume of low-salt buffer or water. Among the  
525 analysed articles, 4 applied this extraction method.

526       • Organic extraction is a method that uses organic solvents. According to this method,  
527       samples are mixed with a reagent composed by a monophasic solution of guanidine  
528       thiocyanate and phenol. Then chloroform is added and the homogenate is allowed to  
529       separate into different phases containing RNA, DNA and proteins. The phases are  
530       separated and finally the nucleic acids are isolated through precipitation with organic  
531       solvents (e.g. isopropanol, ethanol). 4 included articles reported this extraction  
532       method.

533       After extraction of nucleic acids, the molecular detection of viruses is performed through the  
534       amplification of specific nucleic acid fragments using PCR. For RNA viruses, viral genome is  
535       reverse transcribed through a reverse transcriptase-PCR before PCR to obtain the cDNA.

536       Molecular detection methods can provide both qualitative and quantitative data depending on  
537       the PCR type. Qualitative data can be obtained using conventional PCR or nested/semi-  
538       nested PCR (performed by two successive conventional PCR), whose products are  
539       subjected to agarose gel electrophoresis (Corpuz et al., 2020). On the contrary, quantitative  
540       data can be obtained using quantitative PCR (qPCR). Virus quantification can be affected by  
541       some factors that can cause data variability. For example the quantification can be  
542       influenced by recovery efficiency of the applied extraction method, by PCR inhibitory  
543       substances within the samples or by PCR conditions (number of replicates, primer/probe  
544       design, thermal cycling conditions) (Gerba et al., 2018). In addition to providing quantitative  
545       data, another qPCR advantage is that it has a high sensitivity, therefore it can detect even  
546       small amounts of nucleic acids (Corpuz et al., 2020).

547       Regarding molecular detection in the 79 analysed articles, conventional PCR, qPCR, nested  
548       PCR or semi-nested PCR were applied for the identification of viral particles. Overall, the  
549       most applied detection method was qPCR (60/79 articles, 75.9%), followed by nested/semi-  
550       nested PCR and conventional PCR (17/79 articles, 21.5% and 10/79 articles, 12.7%,  
551       respectively). The use of qPCR was frequent, probably because it has a higher sensitivity  
552       than the other molecular methods. The higher sensitivity was confirmed by Assis et al. (2015)  
553       and Dos Santos et al. (2015) studies. These studies applied conventional PCR and qPCR to

554 detect the same virus type; the results showed that a higher number of positive samples was  
555 found using qPCR than using conventional PCR. The qPCR was frequently applied also  
556 because it can provide quantitative data. However, it is important to highlight that even if  
557 using qPCR, the number of genomic copies/L can be quantified, many of the included  
558 articles did not report virus concentrations. In some articles virus concentrations were not  
559 reported because data were under the quantification limit.

560 Methods applied for virus concentration, nucleic acid extraction and molecular analyses are  
561 reported in Table S.5 divided by reference, while data on detection limit/quantification limit  
562 are shown in Tables S1-S4. One of the main problems related to the monitoring of viruses in  
563 water is linked to the fact that different methods can be applied to detect them. Furthermore,  
564 even if quality assurance/quality control is important to assure data quality, in environmental  
565 monitoring studies, this information is often not reported.

566

## 567 **6. CONCLUSIONS**

568 The microbiological water quality for human consumption is crucial for Public Health. As long  
569 as viruses are one of the most important causative agents of waterborne diseases, their  
570 detection in sources for DW production and in DW has a key role in healthcare. In this  
571 review, scientific studies of the last 10 years from all over the world were analysed in order to  
572 summarize data of virus presence assessed using molecular methods in sources for DW  
573 production and in DW. Water types considered were SW-D, GW-D, DW and BW.

574 In the 79 articles finally included in the review different virus types were searched. However,  
575 only some of them are important for Public Health because they may cause waterborne  
576 outbreaks. Therefore, in order to collect more data in short times, the authors of this review  
577 believe that it could be more appropriate to focus research on these viruses (i.e. AdV, EV,  
578 NoV GI, NoV GII, RoV).

579 As highlighted by this review, data on virus presence in water for human consumption are  
580 very heterogeneous. This finding could be related to the methods used for virus detection;  
581 indeed, different combinations of primary-secondary concentration methods and different

582 nucleic acid extraction methods were carried out. This evidence raises an important question  
583 about a lack of standardization of methodologies for virus detection. It is not easy to compare  
584 data collected using different methodologies and it would be desirable to standardise  
585 methodologies in order to make data more comparable.

586 The comparison of virus detection among the water types showed that in SW-D viruses were  
587 frequently detected, while the percentages of positive samples in GW-D were the lowest. It is  
588 crucial to investigate viral presence in sources for DW production (SW-D and GW-D),  
589 because a higher presence in SW and GW could lead to a higher presence in DW. In  
590 particular, the assessment of virus occurrence in SW is important because the use of this  
591 water as DW source will probably increase in the next years. Indeed, climate change and  
592 global population growth will lead to more DW demand and less water availability.  
593 Consequently, to produce DW it will be necessary to increase the use of sources most  
594 vulnerable to contamination, such as SW.

595 Even if at lower percentages compared to SW-D, viruses were detected also in DW, where  
596 they might pose a possible threat to human health. Although these percentages do not  
597 necessarily mean that these samples contain viable pathogenic viruses (because they were  
598 found through molecular detection methods), this evidence suggests the need for regulatory  
599 updates. Indeed, the monitoring of enteric viruses together with coliphages and phages, is  
600 considered important for the assessment of DW treatments effectiveness. However, the only  
601 parameter proposed by WHO guidelines for verification of microbial quality of DW is the  
602 monitoring of *Escherichia coli* or thermotolerant coliform bacteria, whereas for viruses no  
603 guidelines values have been proposed yet (WHO, 2017). Even the new European legislation  
604 (The European Parliament and the Council of the European Union, 2020) requires only the  
605 search for *E. coli* and fecal enterococci to establish the DW requirement for water intended  
606 for human consumption. *Clostridium perfringens* and *Legionella* spp. must only be analysed  
607 on the basis of the risk assessment. Finally, the legislation provides for the search of somatic  
608 coliphages in untreated waters if specifically indicated in the risk assessment.

609 As had already been proposed by the WHO (2004), an excellent way to take into account the  
610 risk associated with the presence of viruses in DW could be the application of the Water  
611 Safety Plans (WSP) which have also been introduced by the new European legislation (The  
612 European Parliament and the Council of the European Union, 2020) and will be mandatory  
613 for all Member States since 2029. It is an approach based on the risk assessment and  
614 management throughout all the water supply chain, from catchment to consumer. The main  
615 limitation derives from the fact that very often water companies do not have data on the  
616 presence of enteric viruses from source to tap (Masciopinto et al., 2019; van den Berg et al.,  
617 2019) and therefore the risk assessment and management are based on the presence of the  
618 microbiological indicators required by the regulations, even if the correlation between *E. coli*,  
619 faecal enterococci and bacteriophages and the presence of viruses is often absent or very  
620 low (Edge et al., 2013; Goh et al., 2019; Lee et al., 2014; Payment and Locas, 2011). This  
621 further strengthens the need to include the search for enteric viruses or alternative indicators  
622 in the monitoring programs, in order to obtain objective data for the application of WSPs.  
623 Considering the comparison of virus detection in DW among the continents, this review  
624 showed that the number of studies is not homogeneously distributed across the continents.  
625 Indeed, few studies have assessed DW collected in Europe while, to our knowledge, there is  
626 no study on Oceanian DW. This finding highlighted the need to analyse additional samples  
627 from these geographical regions. Moreover, the results obtained analysing African samples  
628 were unexpected, so the authors believe that they are worth of further studies. Finally, the  
629 bibliographic research performed in this review demonstrated that only three articles  
630 assessed virus presence in BW, underlining another research gap.

631 In order to estimate the potential human health risk due to virus exposure through DW,  
632 reliable data on virus occurrence in this matrix are needed. However, virus concentration  
633 data in DW are still limited, so future studies are needed to fulfil this research gap. Besides  
634 studies focused on viral occurrence in DW, future research should also investigate virus  
635 distribution in other environmental matrices, such as SW and GW. Indeed, these data

636 together with DW data could be used as inputs to perform Quantitative Microbial Risk  
637 Assessment (QMRA), allowing a more precise estimate of human health risk.

638

## 639 **7. FUNDING**

640 This research was funded by the University of Torino and by Piedmont Region.

641

## 642 **8. ACKNOWLEDGEMENTS**

643 The authors express their gratitude to Dr. Paolo Gardois for assistance in the methodological  
644 approach and for suggestions on the bibliographic research.

645

## 646 **9. DECLARATION OF INTERESTS**

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

649

## 650 **10. REFERENCES**

651 Ahmad, T., Adnan, F., Nadeem, M., Kakar, S.J., Anjum, S., Saad, A., Waheed, A., Arshad,  
652 N., 2018. Assessment of the risk for human health of enterovirus and hepatitis a virus in  
653 clinical and water sources from three metropolitan cities of Pakistan. *Ann. Agric. Environ.*  
654 *Med.* 25, 708–713. <https://doi.org/10.26444/aaem/99590>

655 Ahmad, T., Anjum, S., Afzal, M.S., Raza, H., Zaidi, N. us S.S., Arshad, N., 2015. Molecular  
656 confirmation of enterovirus from sewage and drinking water samples from three cities,  
657 Pakistan: A potential risk factor for public health. *Southeast Asian J. Trop. Med. Public*  
658 *Health* 46, 640–649.

659 Asami, T., Katayama, H., Torrey, J.R., Visvanathan, C., Furumai, H., 2016. Evaluation of  
660 virus removal efficiency of coagulation-sedimentation and rapid sand filtration processes in a  
661 drinking water treatment plant in Bangkok, Thailand. *Water Res.* 101, 84–94.  
662 <https://doi.org/10.1016/j.watres.2016.05.012>



663 Assis, A.S.F., Cruz, L.T., Ferreira, A.S., Bessa, M.E., de Oliveira Pinto, M.A., Vieira, C.B.,  
664 Otenio, M.H., Miagostovich, M.P., da Rosa e Silva, M.L., 2015. Relationship between viral  
665 detection and turbidity in a watershed contaminated with group A rotavirus. *Environ. Sci.*  
666 *Pollut. Res.* 22, 6886–6897. <https://doi.org/10.1007/s11356-014-3874-8>  
667 Atabakhsh, P., Kargar, M., Doosti, A., 2019. Molecular surveillance of human rotaviruses in  
668 drinking water and investigation of the efficiency of their removal in Isfahan water treatment  
669 plant. *Environ. Monit. Assess.* 191. <https://doi.org/10.1007/s10661-019-7834-0>  
670 Aw, T.G., Gin, K.Y.H., 2011. Prevalence and genetic diversity of waterborne pathogenic  
671 viruses in surface waters of tropical urban catchments. *J. Appl. Microbiol.* 110, 903–914.  
672 <https://doi.org/10.1111/j.1365-2672.2011.04947.x>  
673 Baggen, J., Thibaut, H.J., Strating, J.R.P.M., Van Kuppeveld, F.J.M., 2018. The life cycle of  
674 non-polio enteroviruses and how to target it. *Nat. Rev. Microbiol.* 16, 368–381.  
675 <https://doi.org/10.1038/s41579-018-0005-4>  
676 Beer, K.D., Gargano, J.W., Roberts, V.A., Hill, V.R., Garrison, L.E., Kutty, P.K., Hilborn, E.D.,  
677 Wade, T.J., Fullerton, K.E., Yoder, J.S., 2015. Surveillance for Waterborne Disease  
678 Outbreaks Associated with Drinking Water — United States, 2011–2012. *MMWR. Morb.*  
679 *Mortal. Wkly. Rep.* 64, 842–848. <https://doi.org/10.15585/mmwr.mm6431a2>  
680 Bhatt, A., Arora, P., Kumar, S., 2020. Since January 2020 Elsevier has created a COVID-19  
681 resource centre with free information in English and Mandarin on the novel coronavirus  
682 COVID- 19 . The COVID-19 resource centre is hosted on Elsevier Connect , the company ' s  
683 public news and information .  
684 Blanco, A., Guix, S., Fuster, N., Fuentes, C., Bartolomé, R., Cornejo, T., Pintó, R.M., Bosch,  
685 A., 2017. Norovirus in bottled water associated with gastroenteritis outbreak, Spain, 2016.  
686 *Emerg. Infect. Dis.* 23, 1531–1534. <https://doi.org/10.3201/eid2309.161489>  
687 Bonanno Ferraro, G., Suffredini, E., Mancini, P., Veneri, C., Iaconelli, M., Bonadonna, L.,  
688 Montagna, M.T., De Giglio, O., La Rosa, G., 2021. Pepper Mild Mottle Virus as Indicator of  
689 Pollution: Assessment of Prevalence and Concentration in Different Water Environments in  
690 Italy. *Food Environ. Virol.* 13, 117–125. <https://doi.org/10.1007/s12560-020-09458-6>

691 Borchardt, M.A., Spencer, S.K., Kieke, B.A., Lambertini, E., Loge, F.J., 2012. Viruses in  
692 nondisinfected drinking water from municipal wells and community incidence of acute  
693 gastrointestinal illness. *Environ. Health Perspect.* 120, 1272–1279.  
694 <https://doi.org/10.1289/ehp.1104499>

695 Bortagaray, V., Girardi, V., Pou, S., Lizasoain, A., Tort, L.F.L., Spilki, F.R., Colina, R.,  
696 Victoria, M., 2020. Detection, Quantification, and Microbial Risk Assessment of Group A  
697 Rotavirus in Rivers from Uruguay. *Food Environ. Virol.* 12, 89–98.  
698 <https://doi.org/10.1007/s12560-019-09416-x>

699 Bosch, A., Pintó, R.M., Guix, S., 2014. Human Astroviruses. *Clin. Microbiol. Rev.* 27, 1048–  
700 1074.

701 Breitenmoser, A., Fretz, R., Schmid, J., Besl, A., Etter, R., 2011. Outbreak of acute  
702 gastroenteritis due to a washwater-contaminated water supply, Switzerland, 2008. *J. Water*  
703 *Health* 9, 569–576. <https://doi.org/10.2166/wh.2011.158>

704 Broderick, M., Myers, C., Balansay, M., Vo, S., Osuna, A., Russell, K., 2017. Adenovirus 4/7  
705 vaccine's effect on disease rates is associated with disappearance of adenovirus on building  
706 surfaces at a military recruit base. *Mil. Med.* 182, e2069–e2072.  
707 <https://doi.org/10.7205/MILMED-D-17-00109>

708 Cai, L., Yang, Y., Jiao, N., Zhang, R., 2015. Evaluation of tangential flow filtration for the  
709 concentration and separation of bacteria and viruses in contrasting marine environments.  
710 *PLoS One* 10, 1–12. <https://doi.org/10.1371/journal.pone.0136741>

711 Calgua, B., Fumian, T., Rusiñol, M., Rodriguez-Manzano, J., Mbayed, V.A., Bofill-Mas, S.,  
712 Miagostovich, M., Girones, R., 2013. Detection and quantification of classic and emerging  
713 viruses by skimmed-milk flocculation and PCR in river water from two geographical areas.  
714 *Water Res.* 47, 2797–2810. <https://doi.org/10.1016/j.watres.2013.02.043>

715 Canh, V.D., Torii, S., Furumai, H., Katayama, H., 2021. Application of Capsid Integrity (RT-  
716 )qPCR to Assessing Occurrence of Intact Viruses in Surface Water and Tap Water in Japan.  
717 *Water Res.* 189, 116674. <https://doi.org/10.1016/j.watres.2020.116674>

718 Carducci, A., Casini, B., Bani, A., Rovini, E., Verani, M., Mazzoni, F., Giuntini, A., 2003.  
719 Virological control of groundwater quality using biomolecular tests. *Water Sci. Technol.* 47,  
720 261–266.

721 Carter, M.J., 2005. Enterically infecting viruses: Pathogenicity, transmission and significance  
722 for food and waterborne infection. *J. Appl. Microbiol.* 98, 1354–1380.  
723 <https://doi.org/10.1111/j.1365-2672.2005.02635.x>

724 Cashdollar, J.L., Wymer, L., 2013. Methods for primary concentration of viruses from water  
725 samples: A review and meta-analysis of recent studies. *J. Appl. Microbiol.* 115, 1–11.  
726 <https://doi.org/10.1111/jam.12143>

727 CDC, 2021. *Waterborne Disease & Outbreak Surveillance Reporting 2013-2014.*

728 Charest, A.J., Plummer, J.D., Long, S.C., Carducci, A., Verani, M., Sidhu, J.P.S., 2015.  
729 Global occurrence of Torque teno virus in water systems. *J. Water Health* 13, 777–789.  
730 <https://doi.org/10.2166/wh.2015.254>

731 Chen, L., Deng, Y., Dong, S., Wang, H., Li, P., Zhang, H., Chu, W., 2021. The occurrence  
732 and control of waterborne viruses in drinking water treatment: A review. *Chemosphere* 281,  
733 130728. <https://doi.org/10.1016/j.chemosphere.2021.130728>

734 Chigor, V.N., Okoh, A.I., 2012a. Quantitative RT-PCR detection of hepatitis A virus,  
735 rotaviruses and enteroviruses in the Buffalo River and source water dams in the Eastern  
736 Cape Province of South Africa. *Int. J. Environ. Res. Public Health* 9, 4017–4032.  
737 <https://doi.org/10.3390/ijerph9114017>

738 Chigor, V.N., Okoh, A.I., 2012b. Quantitative Detection and Characterization of Human  
739 Adenoviruses in the Buffalo River in the Eastern Cape Province of South Africa. *Food*  
740 *Environ. Virol.* 4, 198–208. <https://doi.org/10.1007/s12560-012-9090-0>

741 Cocoran, E., Nellesmann, C., Baker, E., Bos, R., Osborn, D., Savelli, H., 2010. SICK  
742 WATER? The central role of wastewater management in sustainable development-a rapid  
743 response assesment. UNEP (United Nations Environment Programme), UN-HABITAT,  
744 Nairobi, Kenia.

745 Corpuz, M.V.A., Buonerba, A., Vigliotta, G., Zarra, T., Ballesteros, F., Campiglia, P.,  
746 Belgiorno, V., Korshin, G., Naddeo, V., 2020. Viruses in wastewater: occurrence, abundance  
747 and detection methods. *Sci. Total Environ.* 745, 140910.  
748 <https://doi.org/10.1016/j.scitotenv.2020.140910>

749 Crawford, S.E., Ramani, S., Tate, J.E., Parashar, U.D., Svensson, L., Hagbom, M., Franco,  
750 M.A., Greenberg, H.B., O’Ryan, M., Kang, G., Desselberger, U., Estes, M.K., 2017.  
751 Rotavirus infection. *Nat. Rev. Dis. Prim.* 3. <https://doi.org/10.1038/nrdp.2017.83>

752 Cuevas-Ferrando, E., Randazzo, W., Pérez-Cataluña, A., Sánchez, G., 2020. HEV  
753 Occurrence in Waste and Drinking Water Treatment Plants. *Front. Microbiol.* 10, 1–10.  
754 <https://doi.org/10.3389/fmicb.2019.02937>

755 Da Silva Luz, I., Vasconcellos, L., De Mello Medeiros, V., Miranda, C.A.C., De Oliveira  
756 Rosas, C., Pimenta, M.M.A., Ferreira, F.C., Romaão, C.M.C.P.A., Brandaão, M.L.L.,  
757 Miagostovich, M.P., 2020. Assessment of the microbiological quality of natural mineral  
758 waters according to the manufacturing time of 20 L returnable packs in Brazil. *FEMS*  
759 *Microbiol. Lett.* 367, 1–9. <https://doi.org/10.1093/femsle/fnaa120>

760 de Souza, F.G., da Silva, F.P., Staggemeier, R., Rigotto, C., Spilki, F.R., 2018. Low  
761 occurrence of Hepatitis A virus in water samples from an urban area of Southern Brazil. *J.*  
762 *Sao Paulo Inst. Trop. Med.* 60, 1–6. [https://doi.org/http://dx.doi.org/10.1590/S1678-](https://doi.org/http://dx.doi.org/10.1590/S1678-9946201860069)  
763 [9946201860069](https://doi.org/http://dx.doi.org/10.1590/S1678-9946201860069)

764 Dienus, O., Sokolova, E., Nyström, F., Matussek, A., Löfgren, S., Blom, L., Pettersson,  
765 T.J.R., Lindgren, P.E., 2016. Norovirus Dynamics in Wastewater Discharges and in the  
766 Recipient Drinking Water Source: Long-Term Monitoring and Hydrodynamic Modeling.  
767 *Environ. Sci. Technol.* 50, 10851–10858. <https://doi.org/10.1021/acs.est.6b02110>

768 Diston, D., Sinreich, M., Zimmermann, S., Baumgartner, A., Felleisen, R., 2015. Evaluation  
769 of Molecular- and Culture-Dependent MST Markers to Detect Fecal Contamination and  
770 Indicate Viral Presence in Good Quality Groundwater. *Environ. Sci. Technol.* 49, 7142–7151.  
771 <https://doi.org/10.1021/acs.est.5b00515>

772 Donia, D., Kota, M., Leno, L., Ylli, A., Cenko, F., Divizia, M., 2011. First outbreak of norovirus  
773 in Albania. *Lett. Appl. Microbiol.* 53, 283–287. [https://doi.org/10.1111/j.1472-](https://doi.org/10.1111/j.1472-765X.2011.03104.x)  
774 [765X.2011.03104.x](https://doi.org/10.1111/j.1472-765X.2011.03104.x)

775 Dos Santos, V.R., Rigotto, C., Staggemeier, R., Vecchia, A.D., Henzel, A., Spilki, F.R., 2015.  
776 Preliminary evaluation of enteric viruses in bottled mineral water commercialized in Brazil.  
777 *Beverages* 1, 140–148. <https://doi.org/10.3390/beverages1030140>

778 ECDC, 2021. The European Union One Health 2019 Zoonoses Report. *EFSA J.* 19.  
779 <https://doi.org/10.2903/j.efsa.2021.6406>

780 Edge, T.A., Khan, I.U.H., Bouchard, R., Guo, J., Hill, S., Locas, A., Moore, L., Neumann, N.,  
781 Nowak, E., Payment, P., Yang, R., Yerubandi, R., Watson, S., 2013. Occurrence of  
782 waterborne pathogens and escherichia coli at offshore drinking water intakes in lake Ontario.  
783 *Appl. Environ. Microbiol.* 79, 5799–5813. <https://doi.org/10.1128/AEM.00870-13>

784 Espinosa, A.C., Mazari-Hiriart, M., Espinosa, R., Maruri-Avidal, L., Méndez, E., Arias, C.F.,  
785 2008. Infectivity and genome persistence of rotavirus and astrovirus in groundwater and  
786 surface water. *Water Res.* 42, 2618–2628. <https://doi.org/10.1016/j.watres.2008.01.018>

787 Fayomi, G.U., Mini, S.E., Fayomi, O.S.I., Owodolu, T., Ayoola, A.A., Wusu, O., 2019. A Mini  
788 Review on the Impact of Sewage Disposal on Environment and Ecosystem. *IOP Conf. Ser.*  
789 *Earth Environ. Sci.* 331. <https://doi.org/10.1088/1755-1315/331/1/012040>

790 Ferguson, A.S., Layton, A.C., Mailloux, B.J., Culligan, P.J., Williams, D.E., Smartt, A.E.,  
791 Saylor, G.S., Feighery, J., McKay, L.D., Knappett, P.S.K., Alexandrova, E., Arbit, T., Emch,  
792 M., Escamilla, V., Ahmed, K.M., Alam, M.J., Streatfield, P.K., Yunus, M., van Geen, A., 2012.  
793 Comparison of fecal indicators with pathogenic bacteria and rotavirus in groundwater. *Sci.*  
794 *Total Environ.* 431, 314–322. <https://doi.org/10.1016/j.scitotenv.2012.05.060>

795 Ferrer, O., Casas, S., Galvañ, C., Lucena, F., Bosch, A., Galofré, B., Mesa, J., Jofre, J.,  
796 Bernat, X., 2015. Direct ultrafiltration performance and membrane integrity monitoring by  
797 microbiological analysis. *Water Res.* 83, 121–131.  
798 <https://doi.org/10.1016/j.watres.2015.06.039>

799 Flannery, J., Rajko-Nenow, P., Keaveney, S., O'Flaherty, V., Dorè, W., 2013. Simulated  
800 sunlight inactivation of norovirus and FRNA bacteriophage in seawater. *J. Appl. Microbiol.*  
801 115, 915–922. <https://doi.org/10.1111/jam.12279>

802 Foladori, P., Cutrupi, F., Segata, N., Manara, S., Pinto, F., Malpei, F., Bruni, L., La Rosa, G.,  
803 2020. SARS-CoV-2 from faeces to wastewater treatment: What do we know? A review. *Sci.*  
804 *Total Environ.* 743, 140444. <https://doi.org/10.1016/j.scitotenv.2020.140444>

805 Fongaro, G., Nascimento, M.A.D., Rigotto, C., Ritterbusch, G., Da Silva, A.D.A., Esteves,  
806 P.A., Barardi, C.R.M., 2013. Evaluation and molecular characterization of human adenovirus  
807 in drinking water supplies: Viral integrity and viability assays. *Viol. J.* 10, 1–9.  
808 <https://doi.org/10.1186/1743-422X-10-166>

809 Fongaro, G., Padilha, J., Schissi, C.D., Nascimento, M.A., Bampi, G.B., Viancelli, A., Barardi,  
810 C.R.M., 2015. Human and animal enteric virus in groundwater from deep wells, and  
811 recreational and network water. *Environ. Sci. Pollut. Res.* 22, 20060–20066.  
812 <https://doi.org/10.1007/s11356-015-5196-x>

813 Fout, G.S., Cashdollar, J.L., Varughese, E.A., Parshionkar, S.U., Grimm, A.C., 2015. EPA  
814 method 1615. Measurement of enterovirus and norovirus occurrence in water by culture and  
815 RT-qPCR. I. collection of virus samples. *J. Vis. Exp.* 2015, 1–7.  
816 <https://doi.org/10.3791/52067>

817 Gad, M.A., Allayeh, A.K., Elmahdy, E.M., Shaheen, M.N.F., Rizk, N.M., Al-Herrawy, A.Z.,  
818 Saleh, F.E.Z.R., Marouf, M.A., 2019. Genotyping and interaction-reality of acanthamoeba,  
819 enteric adenovirus and rotavirus in drinking water, Egypt. *Egypt. J. Aquat. Biol. Fish.* 23, 65–  
820 79. <https://doi.org/10.21608/ejabf.2019.29299>

821 Gamazo, P., Victoria, M., Schijven, J.F., Alvareda, E., Tort, L.F.L., Ramos, J., Burutaran, L.,  
822 Olivera, M., Lizasoain, A., Sapriza, G., Castells, M., Colina, R., 2018. Evaluation of Bacterial  
823 Contamination as an Indicator of Viral Contamination in a Sedimentary Aquifer in Uruguay.  
824 *Food Environ. Virol.* 10, 305–315. <https://doi.org/10.1007/s12560-018-9341-9>

825 Garcia, L.A.T., Viancelli, A., Rigotto, C., Pilotto, M.R., Esteves, P.A., Kunz, A., Barardi,  
826 C.R.M., 2012. Surveillance of human and swine adenovirus, human norovirus and swine

827 circovirus in water samples in Santa Catarina, Brazil. *J. Water Health* 10, 445–452.  
828 <https://doi.org/10.2166/wh.2012.190>

829 Garver, K.A., Mahony, A.A.M., Stucchi, D., Richard, J., Van Woensel, C., Foreman, M.,  
830 2013. Estimation of parameters influencing waterborne transmission of infectious  
831 hematopoietic necrosis virus (IHNV) in atlantic salmon (*Salmo salar*). *PLoS One* 8.  
832 <https://doi.org/10.1371/journal.pone.0082296>

833 Gerba, C.P., Betancourt, W.Q., 2019. Assessing the occurrence of waterborne viruses in  
834 reuse systems: Analytical limits and needs. *Pathogens* 8.  
835 <https://doi.org/10.3390/pathogens8030107>

836 Gerba, C.P., Betancourt, W.Q., Kitajima, M., 2017. How much reduction of virus is needed  
837 for recycled water: A continuous changing need for assessment? *Water Res.* 108, 25–31.  
838 <https://doi.org/10.1016/j.watres.2016.11.020>

839 Gerba, C.P., Betancourt, W.Q., Kitajima, M., Rock, C.M., 2018. Reducing uncertainty in  
840 estimating virus reduction by advanced water treatment processes. *Water Res.* 133, 282–  
841 288. <https://doi.org/10.1016/j.watres.2018.01.044>

842 Giammanco, G.M., Di Bartolo, I., Purpari, G., Costantino, C., Rotolo, V., Spoto, V., Geraci,  
843 G., Bosco, G., Petralia, A., Guercio, A., Macaluso, G., Calamusa, G., De Grazia, S., Ruggeri,  
844 F.M., Vitale, F., Maida, C.M., Mammina, C., 2014. Investigation and control of a Norovirus  
845 outbreak of probable waterborne transmission through a municipal groundwater system. *J.*  
846 *Water Health* 12, 452–464. <https://doi.org/10.2166/wh.2014.227>

847 Gibson, K.E., Opryszko, M.C., Schissler, J.T., Guo, Y., Schwab, K.J., 2011. Evaluation of  
848 human enteric viruses in surface water and drinking water resources in southern Ghana. *Am.*  
849 *J. Trop. Med. Hyg.* 84, 20–29. <https://doi.org/10.4269/ajtmh.2011.10-0389>

850 Gibson, K.E., Schwab, K.J., 2011a. Detection of bacterial indicators and human and bovine  
851 enteric viruses in surface water and groundwater sources potentially impacted by animal and  
852 human wastes in Lower Yakima Valley, Washington. *Appl. Environ. Microbiol.* 77, 355–362.  
853 <https://doi.org/10.1128/AEM.01407-10>

854 Gibson, K.E., Schwab, K.J., 2011b. Tangential-flow ultrafiltration with integrated inhibition  
855 detection for recovery of surrogates and human pathogens from large-volume source water  
856 and finished drinking water. *Appl. Environ. Microbiol.* 77, 385–391.  
857 <https://doi.org/10.1128/AEM.01164-10>

858 Goh, S.G., Saeidi, N., Gu, X., Vergara, G.G.R., Liang, L., Fang, H., Kitajima, M., Kushmaro,  
859 A., Gin, K.Y.H., 2019. Occurrence of microbial indicators, pathogenic bacteria and viruses in  
860 tropical surface waters subject to contrasting land use. *Water Res.* 150, 200–215.  
861 <https://doi.org/10.1016/j.watres.2018.11.058>

862 Gotkowitz, M.B., Bradbury, K.R., Borchardt, M.A., Zhu, J., Spencer, S.K., 2016. Effects of  
863 Climate and Sewer Condition on Virus Transport to Groundwater. *Environ. Sci. Technol.* 50,  
864 8497–8504. <https://doi.org/10.1021/acs.est.6b01422>

865 Griffin, J.S., Plummer, J.D., Long, S.C., 2008. Torque teno virus: An improved indicator for  
866 viral pathogens in drinking waters. *Virol. J.* 5, 1–6. <https://doi.org/10.1186/1743-422X-5-112>

867 Grøndahl-Rosado, R.C., Yarovitsyna, E., Trettenes, E., Myrmel, M., Robertson, L.J., 2014. A  
868 One Year Study on the Concentrations of Norovirus and Enteric Adenoviruses in Wastewater  
869 and A Surface Drinking Water Source in Norway. *Food Environ. Virol.* 6, 232–245.  
870 <https://doi.org/10.1007/s12560-014-9161-5>

871 Guerrero-Latorre, L., Carratala, A., Rodriguez-Manzano, J., Calgua, B., Hundesa, A.,  
872 Girones, R., 2011. Occurrence of water-borne enteric viruses in two settlements based in  
873 Eastern Chad: Analysis of hepatitis E virus, hepatitis A virus and human adenovirus in water  
874 sources. *J. Water Health* 9, 515–524. <https://doi.org/10.2166/wh.2011.126>

875 Hamza, I.A., Jurzik, L., Überla, K., Wilhelm, M., 2011. Evaluation of pepper mild mottle virus,  
876 human picobirnavirus and Torque teno virus as indicators of fecal contamination in river  
877 water. *Water Res.* 45, 1358–1368. <https://doi.org/10.1016/j.watres.2010.10.021>

878 Haramoto, E., Kitajima, M., Hata, A., Torrey, J.R., Masago, Y., Sano, D., Katayama, H.,  
879 2018. A review on recent progress in the detection methods and prevalence of human  
880 enteric viruses in water. *Water Res.* 135, 168–186.  
881 <https://doi.org/10.1016/j.watres.2018.02.004>



882 Haramoto, E., Kitajima, M., Kishida, N., Katayama, H., Asami, M., Akiba, M., 2012.  
883 Occurrence of Viruses and Protozoa in Drinking Water Sources of Japan and Their  
884 Relationship to Indicator Microorganisms. *Food Environ. Virol.* 4, 93–101.  
885 <https://doi.org/10.1007/s12560-012-9082-0>

886 Haramoto, E., Kitajima, M., Kishida, N., Konno, Y., Katayama, H., Asami, M., Akiba, M.,  
887 2013. Occurrence of pepper mild mottle virus in drinking water sources in Japan. *Appl.*  
888 *Environ. Microbiol.* 79, 7413–7418. <https://doi.org/10.1128/AEM.02354-13>

889 Hata, A., Shirasaka, Y., Ihara, M., Yamashita, N., Tanaka, H., 2021. Spatial and temporal  
890 distributions of enteric viruses and indicators in a lake receiving municipal wastewater  
891 treatment plant discharge. *Sci. Total Environ.* 780, 146607.  
892 <https://doi.org/10.1016/j.scitotenv.2021.146607>

893 Hssaine, A., Gharbi, J., Harrath, R., Harrak, R., Chait, A., Aouni, M., Hafid, J., *Physiologie,*  
894 *E.I., Aliments, L., Santé, E.*, 2011. In search of enteroviruses in water media in Marrakech.  
895 *African J. Microbiol. Res.* 5, 2380–2384. <https://doi.org/10.5897/AJMR09.462>

896 Iaconelli, M., Muscillo, M., Della Libera, S., Fratini, M., Meucci, L., De Ceglia, M., Giacosa,  
897 D., La Rosa, G., 2017. One-year Surveillance of Human Enteric Viruses in Raw and Treated  
898 Wastewaters, Downstream River Waters, and Drinking Waters. *Food Environ. Virol.* 9, 79–  
899 88. <https://doi.org/10.1007/s12560-016-9263-3>

900 Ikner, L.A., Gerba, C.P., Bright, K.R., 2012. Concentration and Recovery of Viruses from  
901 Water: A Comprehensive Review. *Food Environ. Virol.* 4, 41–67.  
902 <https://doi.org/10.1007/s12560-012-9080-2>

903 Jacob, P., Henry, A., Meheut, G., Charni-Ben-Tabassi, N., Ingr, V., Helmi, K., 2015. Health  
904 risk assessment related to waterborne pathogens from the river to the tap. *Int. J. Environ.*  
905 *Res. Public Health* 12, 2967–2983. <https://doi.org/10.3390/ijerph120302967>

906 Jiménez-Melsió, A., Parés, S., Segalés, J., Kekarainen, T., 2013. Detection of porcine  
907 anelloviruses in pork meat and human faeces. *Virus Res.* 178, 522–524.  
908 <https://doi.org/10.1016/j.virusres.2013.09.035>

909 Joungh, H.K., Han, S.H., Park, S.J., Jheong, W.H., Ahn, T.S., Lee, J.B., Jeong, Y.S., Jang,  
910 K.L., Lee, G.C., Rhee, O.J., Park, J.W., Paik, S.Y., 2013. Nationwide surveillance for  
911 pathogenic microorganisms in groundwater near carcass burials constructed in South Korea  
912 in 2010. *Int. J. Environ. Res. Public Health* 10, 7126–7143.  
913 <https://doi.org/10.3390/ijerph10127126>

914 Jung, J.H., Yoo, C.H., Koo, E.S., Kim, H.M., Na, Y., Jheong, W.H., Jeong, Y.S., 2011.  
915 Occurrence of norovirus and other enteric viruses in untreated groundwaters of Korea. *J.*  
916 *Water Health* 9, 544–555. <https://doi.org/10.2166/wh.2011.142>

917 Kato, R., Asami, T., Utagawa, E., Furumai, H., Katayama, H., 2018. Pepper mild mottle virus  
918 as a process indicator at drinking water treatment plants employing coagulation-  
919 sedimentation, rapid sand filtration, ozonation, and biological activated carbon treatments in  
920 Japan. *Water Res.* 132, 61–70. <https://doi.org/10.1016/j.watres.2017.12.068>

921 Kishida, N., Morita, H., Haramoto, E., Asami, M., Akiba, M., 2012. One-year weekly survey of  
922 noroviruses and enteric adenoviruses in the Tone River water in Tokyo metropolitan area,  
923 Japan. *Water Res.* 46, 2905–2910. <https://doi.org/10.1016/j.watres.2012.03.010>

924 Kitajima, M., Sassi, H.P., Torrey, J.R., 2018. Pepper mild mottle virus as a water quality  
925 indicator. *npj Clean Water* 1. <https://doi.org/10.1038/s41545-018-0019-5>

926 Kittigul, L., Pombubpa, K., 2021. Rotavirus Surveillance in Tap Water, Recycled Water, and  
927 Sewage Sludge in Thailand: A Longitudinal Study, 2007–2018. *Food Environ. Virol.* 13, 53–  
928 63. <https://doi.org/10.1007/s12560-020-09450-0>

929 Kiulia, N.M., Mans, J., Mwenda, J.M., Taylor, M.B., 2014. Norovirus GII.17 Predominates in  
930 Selected Surface Water Sources in Kenya. *Food Environ. Virol.* 6, 221–231.  
931 <https://doi.org/10.1007/s12560-014-9160-6>

932 Kluge, M., Fleck, J.D., Soliman, M.C., Luz, R.B., Fabres, R.B., Comerlato, J., Silva, J.V.S.,  
933 Staggemeier, R., Vecchia, A.D., Capalonga, R., Oliveira, A.B., Henzel, A., Rigotto, C., Spilki,  
934 F.R., 2014. Human adenovirus (HAdV), human enterovirus (hEV), and genogroup A rotavirus  
935 (GARV) in tap water in southern Brazil. *J. Water Health* 12, 526–532.  
936 <https://doi.org/10.2166/wh.2014.202>

937 Knappett, P.S.K., Layton, A., McKay, L.D., Williams, D., Mailloux, B.J., Huq, M.R., Alam,  
938 M.J., Ahmed, K.M., Akita, Y., Serre, M.L., Saylor, G.S., Van Geen, A., 2011. Efficacy of  
939 Hollow-Fiber Ultrafiltration for Microbial Sampling in Groundwater. *Ground Water* 49, 53–65.  
940 <https://doi.org/10.1111/j.1745-6584.2010.00712.x>

941 Koopmans, M., Duizer, E., 2004. Foodborne viruses: An emerging problem. *Int. J. Food*  
942 *Microbiol.* 90, 23–41. [https://doi.org/10.1016/S0168-1605\(03\)00169-7](https://doi.org/10.1016/S0168-1605(03)00169-7)

943 Kopecka, H., Dubrou, S., Prevot, J., Marechal, J., Lopez-Pila, J.M., 1993. Detection of  
944 naturally occurring enteroviruses in waters by reverse transcription, polymerase chain  
945 reaction, and hybridization. *Appl. Environ. Microbiol.* 59, 1213–1219.  
946 <https://doi.org/10.1128/aem.59.4.1213-1219.1993>

947 Kuroda, K., Nakada, N., Hanamoto, S., Inaba, M., Katayama, H., Do, A.T., Nga, T.T.V.,  
948 Oguma, K., Hayashi, T., Takizawa, S., 2015. Pepper mild mottle virus as an indicator and a  
949 tracer of fecal pollution in water environments: Comparative evaluation with wastewater-  
950 tracer pharmaceuticals in Hanoi, Vietnam. *Sci. Total Environ.* 506–507, 287–298.  
951 <https://doi.org/10.1016/j.scitotenv.2014.11.021>

952 Lambertini, E., Spencer, S.K., Kieke, B.A., Loge, F.J., Borchardt, M.A., 2011. Virus  
953 contamination from operation and maintenance events in small drinking water distribution  
954 systems. *J. Water Health* 9, 799–812. <https://doi.org/10.2166/wh.2011.018>

955 Larrue, H., Abravanel, F., Peron, J.M., 2021. Hepatitis E, what is the real issue? *Liver Int.* 41,  
956 68–72. <https://doi.org/10.1111/liv.14880>

957 Leclerc, H., Edberg, S., Pierzo, V., Delattre, J.M., 2000. Bacteriophages as indicators of  
958 enteric viruses and public health risk in groundwaters. *J. Appl. Microbiol.* 88, 5–21.  
959 <https://doi.org/10.1046/j.1365-2672.2000.00949.x>

960 Lee, C.S., Lee, C., Marion, J., Wang, Q., Saif, L., Lee, J., 2014. Occurrence of human enteric  
961 viruses at freshwater beaches during swimming season and its link to water inflow. *Sci. Total*  
962 *Environ.* 472, 757–766. <https://doi.org/10.1016/j.scitotenv.2013.11.088>

963 Lee, J.S., Joo, I.S., Ju, S.Y., Jeong, M.H., Song, Y.H., Kwak, H.S., 2018. Research on the  
964 contamination levels of norovirus in food facilities using groundwater in South Korea, 2015–  
965 2016. *Int. J. Food Microbiol.* 280, 35–40. <https://doi.org/10.1016/j.ijfoodmicro.2018.05.010>  
966 Liang, L., Goh, S.G., Vergara, G.G.R.V., Fang, H.M., Rezaeinejad, S., Chang, S.Y., Bayen,  
967 S., Lee, W.A., Sobsey, M.D., Rose, J.B., Gin, K.Y.H., 2015. Alternative fecal indicators and  
968 their empirical relationships with enteric viruses, *Salmonella enterica*, and *Pseudomonas*  
969 *aeruginosa* in surface waters of a tropical urban catchment. *Appl. Environ. Microbiol.* 81,  
970 850–860. <https://doi.org/10.1128/AEM.02670-14>  
971 Ligon, G., Bartram, J., 2016. Literature review of associations among attributes of reported  
972 drinking water disease outbreaks. *Int. J. Environ. Res. Public Health* 13.  
973 <https://doi.org/10.3390/ijerph13060527>  
974 Lugo, D., Krogstad, P., 2016. Enteroviruses in the early 21st century: New manifestations  
975 and challenges. *Curr. Opin. Pediatr.* 28, 107–113.  
976 <https://doi.org/10.1097/MOP.0000000000000303>  
977 Mackowiak, M., Leifels, M., Hamza, I.A., Jurzik, L., Wingender, J., 2018. Distribution of  
978 *Escherichia coli*, coliphages and enteric viruses in water, epilithic biofilms and sediments of  
979 an urban river in Germany. *Sci. Total Environ.* 626, 650–659.  
980 <https://doi.org/10.1016/j.scitotenv.2018.01.114>  
981 Malla, B., Shrestha, R.G., Tandukar, S., Bhandari, D., Thakali, O., Sherchand, J.B.,  
982 Haramoto, E., 2019. Detection of pathogenic viruses, pathogen indicators, and fecal-source  
983 markers within tanker water and their sources in the Kathmandu valley, Nepal. *Pathogens* 8,  
984 1–13. <https://doi.org/10.3390/pathogens8020081>  
985 Marie, V., Lin, J., 2017. Viruses in the environment - presence and diversity of bacteriophage  
986 and enteric virus populations in the Umhlangane River, Durban, South Africa. *J. Water*  
987 *Health* 15, 966–981. <https://doi.org/10.2166/wh.2017.066>  
988 Masciopinto, C., De Giglio, O., Scrascia, M., Fortunato, F., La Rosa, G., Suffredini, E.,  
989 Pazzani, C., Prato, R., Montagna, M.T., 2019. Human health risk assessment for the

990 occurrence of enteric viruses in drinking water from wells: Role of flood runoff injections. *Sci.*  
991 *Total Environ.* 666, 559–571. <https://doi.org/10.1016/j.scitotenv.2019.02.107>

992 Mattioli, M.C., Pickering, A.J., Gilsdorf, R.J., Davis, J., Boehm, A.B., 2013. Hands and water  
993 as vectors of diarrheal pathogens in Bagamoyo, Tanzania. *Environ. Sci. Technol.* 47, 355–  
994 363. <https://doi.org/10.1021/es303878d>

995 Mena, K.D., Gerba, C.P., 2008. Mena\_ Waterborne adenovirus.pdf.

996 Miagostovich, M.P., Rocha, M.S., dos Reis, F.B., Sampaio, M.S., de Saldanha da Gama  
997 Gracie Carrijo, R., Malta, F.C., Rodrigues, J., Genuino, A., Ribeiro da Silva Assis, M.,  
998 Fumian, T.M., Barrocas, P.R.G., 2020. Gastroenteric Viruses Detection in a Drinking Water  
999 Distribution-to-Consumption System in a Low-Income Community in Rio de Janeiro. *Food*  
1000 *Environ. Virol.* 12, 130–136. <https://doi.org/10.1007/s12560-020-09423-3>

1001 Miura, T., Gima, A., Akiba, M., 2019. Detection of Norovirus and Rotavirus Present in  
1002 Suspended and Dissolved Forms in Drinking Water Sources. *Food Environ. Virol.* 11, 9–19.  
1003 <https://doi.org/10.1007/s12560-018-9361-5>

1004 Moreira, N.A., Bondelind, M., 2017. Safe drinking water and waterborne outbreaks. *J. Water*  
1005 *Health* 15, 83–96. <https://doi.org/10.2166/wh.2016.103>

1006 Murphy, H.M., McGinnis, S., Blunt, R., Stokdyk, J., Wu, J., Cagle, A., Denno, D.M., Spencer,  
1007 S., Firnstahl, A., Borchardt, M.A., 2020. Septic Systems and Rainfall Influence Human Fecal  
1008 Marker and Indicator Organism Occurrence in Private Wells in Southeastern Pennsylvania.  
1009 *Environ. Sci. Technol.* 54, 3159–3168. <https://doi.org/10.1021/acs.est.9b05405>

1010 Nargessi, D., Ou, C.Y., 2010. MagaZorb: a simple tool for rapid isolation of viral nucleic  
1011 acids. *J. Infect. Dis.* 201 Suppl, 37–41. <https://doi.org/10.1086/650391>

1012 Ogorzaly, L., Bertrand, I., Paris, M., Maul, A., Gantzer, C., 2010. Occurrence, survival, and  
1013 persistence of human adenoviruses and F-specific RNA phages in raw groundwater. *Appl.*  
1014 *Environ. Microbiol.* 76, 8019–8025. <https://doi.org/10.1128/AEM.00917-10>

1015 Okoh, A.I., Sibanda, T., Gusha, S.S., 2010. Inadequately treated wastewater as a source of  
1016 human enteric viruses in the environment. *Int. J. Environ. Res. Public Health* 7, 2620–2637.  
1017 <https://doi.org/10.3390/ijerph7062620>

1018 Opere, W.M., John, M., Ombori, O., 2021. Molecular Detection of Human Enteric  
1019 Adenoviruses in Water Samples Collected from Lake Victoria Waters Along Homa Bay  
1020 Town, Homa Bay County, Kenya. *Food Environ. Virol.* 13, 32–43.  
1021 <https://doi.org/10.1007/s12560-020-09444-y>

1022 Payment, P., Locas, A., 2011. Pathogens in Water: Value and Limits of Correlation with  
1023 Microbial Indicators. *Ground Water* 49, 4–11. [https://doi.org/10.1111/j.1745-](https://doi.org/10.1111/j.1745-6584.2010.00710.x)  
1024 [6584.2010.00710.x](https://doi.org/10.1111/j.1745-6584.2010.00710.x)

1025 Pérez-Sautu, U., Sano, D., Guix, S., Kasimir, G., Pintó, R.M., Bosch, A., 2012. Human  
1026 norovirus occurrence and diversity in the Llobregat river catchment, Spain. *Environ.*  
1027 *Microbiol.* 14, 494–502. <https://doi.org/10.1111/j.1462-2920.2011.02642.x>

1028 Pinon, A., Vialette, M., 2019. Survival of viruses in water. *Intervirol* 61, 214–222.  
1029 <https://doi.org/10.1159/000484899>

1030 Potgieter, N., Karambwe, S., Mudau, L.S., Barnard, T., Traore, A., 2020. Human enteric  
1031 pathogens in eight rivers used as rural household drinking water sources in the northern  
1032 region of South Africa. *Int. J. Environ. Res. Public Health* 17.  
1033 <https://doi.org/10.3390/ijerph17062079>

1034 Rachmadi, A.T., Kitajima, M., Pepper, I.L., Gerba, C.P., 2016. Enteric and indicator virus  
1035 removal by surface flow wetlands. *Sci. Total Environ.* 542, 976–982.  
1036 <https://doi.org/10.1016/j.scitotenv.2015.11.001>

1037 Rashid, M., Khan, M.N., Jalbani, N., 2021. Detection of Human Adenovirus, Rotavirus, and  
1038 Enterovirus in Tap Water and Their Association with the Overall Quality of Water in Karachi,  
1039 Pakistan. *Food Environ. Virol.* 13, 44–52. <https://doi.org/10.1007/s12560-020-09448-8>

1040 Rebato, N.D., de Los Reyes, V.C.D., Sucaldito, M.N.L., Marin, G.R., 2019. Is your drinking-  
1041 water safe? A rotavirus outbreak linked to water refilling stations in the Philippines, 2016.  
1042 *West. Pacific Surveill. response J. WPSAR* 10, 1–5.  
1043 <https://doi.org/10.5365/wpsar.2017.8.1.007>

1044 Reeve, P., Regel, R., Dreyfus, J., Monis, P., Lau, M., King, B., Van Den Akker, B., 2016.  
1045 Virus removal of new and aged UF membranes at full-scale in a wastewater reclamation  
1046 plant. *Environ. Sci. Water Res. Technol.* 2, 1014–1021. <https://doi.org/10.1039/c6ew00197a>  
1047 Rizk, N.M., Allayeh, A.K., 2018. Multiplex Semi-Nested RT-PCR for Genotyping of  
1048 Rotaviruses Group A in Giza Tap Water, Egypt. *Asian J. Water, Environ. Pollut.* 15, 217–221.  
1049 <https://doi.org/10.3233/AJW-180034>  
1050 Ruhanya, V., 2016. Adsorption-Elution Techniques and Molecular Detection of Enteric  
1051 Viruses from Water. *J. Hum. Virol. Retrovirology* 3.  
1052 <https://doi.org/10.15406/jhvr.2016.03.00112>  
1053 Salvador, D., Neto, C., Benoliel, M.J., Filomena Caeiro, M., 2020. Assessment of the  
1054 presence of hepatitis e virus in surface water and drinking water in Portugal. *Microorganisms*  
1055 8, 1–16. <https://doi.org/10.3390/microorganisms8050761>  
1056 Sangsanont, J., The Dan, D., Thi Viet Nga, T., Katayama, H., Furumai, H., 2016. Detection of  
1057 pepper mild mottle virus as an indicator for drinking water quality in Hanoi, Vietnam, in large  
1058 volume of water after household treatment. *J. Environ. Sci. Heal. - Part A Toxic/Hazardous*  
1059 *Subst. Environ. Eng.* 51, 1100–1106. <https://doi.org/10.1080/10934529.2016.1199650>  
1060 Sano, D., Amarasiri, M., Hata, A., Watanabe, T., Katayama, H., 2016. Risk management of  
1061 viral infectious diseases in wastewater reclamation and reuse: Review. *Environ. Int.* 91, 220–  
1062 229. <https://doi.org/10.1016/j.envint.2016.03.001>  
1063 Seelen, L.M.S., Flaim, G., Jennings, E., De Senerpont Domis, L.N., 2019. Saving water for  
1064 the future: Public awareness of water usage and water quality. *J. Environ. Manage.* 242,  
1065 246–257. <https://doi.org/10.1016/j.jenvman.2019.04.047>  
1066 Shang, X., Fu, X., Zhang, P., Sheng, M., Song, J., He, F., Qiu, Y., Wu, H., Lu, Q., Feng, Y.,  
1067 Lin, J., Chen, E., Chai, C., 2017. An outbreak of norovirus-associated acute gastroenteritis  
1068 associated with contaminated barrelled water in many schools in Zhejiang, China. *PLoS One*  
1069 12, 1–11. <https://doi.org/10.1371/journal.pone.0171307>

1070 Shao, J., Hou, J., Song, H., 2011. Comparison of humic acid rejection and flux decline during  
1071 filtration with negatively charged and uncharged ultrafiltration membranes. *Water Res.* 45,  
1072 473–482. <https://doi.org/10.1016/j.watres.2010.09.006>

1073 Shi, D., Ma, H., Miao, J., Liu, W., Yang, D., Qiu, Z., Shen, Z., Yin, J., Yang, Z., Wang, H., Li,  
1074 H., Chen, Z., Li, J., Jin, M., 2021. Levels of human Rotaviruses and Noroviruses GII in urban  
1075 rivers running through the city mirror their infection prevalence in populations. *Sci. Total*  
1076 *Environ.* 754, 142203. <https://doi.org/10.1016/j.scitotenv.2020.142203>

1077 Shoeib, A.R.S., El-Esnawy, N.A., Zarouk, A.W., 2011. Molecular Detection and  
1078 Predominance of Human Torque Teno Virus in Children's with acute hepatitis and  
1079 Environmental Waters. *Life Sci. J.* 8, 165–172.

1080 Shoham, D., Jahangir, A., Ruenphet, S., Takehara, K., 2012. Persistence of Avian Influenza  
1081 Viruses in Various Artificially Frozen Environmental Water Types. *Influenza Res. Treat.* 2012,  
1082 1–11. <https://doi.org/10.1155/2012/912326>

1083 Silva, H.D., Fongaro, G., Garcíazapata, M.T.A., Melo, A.T.O., Silveira-Lacerda, E.P., de  
1084 Faria, K.M.S., Anunciação, C.E., 2015. High Species C Human Adenovirus Genome Copy  
1085 Numbers in the Treated Water Supply of a Neotropical Area of the Central-West Region of  
1086 Brazil. *Food Environ. Virol.* 7, 286–294. <https://doi.org/10.1007/s12560-015-9192-6>

1087 Smith, D.B., Simmonds, P., 2018. Classification and genomic diversity of enterically  
1088 transmitted hepatitis viruses. *Cold Spring Harb. Perspect. Med.* 8, 1–16.  
1089 <https://doi.org/10.1101/cshperspect.a031880>

1090 Spilki, F.R., da Luz, R.B., Fabres, R.B., Soliman, M.C., Kluge, M., Fleck, J.D., Rodrigues,  
1091 M.T., Comerlato, J., Cenci, A., Cerva, C., Dasso, M.G., Roehe, P.M., 2013. Detection of  
1092 human adenovirus, rotavirus and enterovirus in water samples collected on dairy farms from  
1093 Tenente Portela, Northwest Of Rio Grande do Sul, Brazil. *Brazilian J. Microbiol.* 44, 953–957.  
1094 <https://doi.org/10.1590/S1517-83822013000300046>

1095 Staggemeier, R., Bortoluzzi, M., da Silva Heck, T.M., da Luz, R.B., Fabres, R.B., Soliman,  
1096 M.C., Rigotto, C., Baldasso, N.A., Spilki, F.R., de Matos Almeida, S.E., 2015. Animal and



1097 human enteric viruses in water and sediment samples from dairy farms. *Agric. Water Manag.*  
1098 152, 135–141. <https://doi.org/10.1016/j.agwat.2015.01.010>

1099 Steele, A.D., Peenze, I., De Beer, M.C., Pager, C.T., Yeats, J., Potgieter, N., Ramsaroop, U.,  
1100 Page, N.A., Mitchell, J.O., Geyer, A., Bos, P., Alexander, J.J., 2003. Anticipating rotavirus  
1101 vaccines: Epidemiology and surveillance of rotavirus in South Africa. *Vaccine* 21, 354–360.  
1102 [https://doi.org/10.1016/S0264-410X\(02\)00615-1](https://doi.org/10.1016/S0264-410X(02)00615-1)

1103 Steyer, A., Torkar, K.G., Gutiérrez-Aguirre, I., Poljšak-Prijatelj, M., 2011. High prevalence of  
1104 enteric viruses in untreated individual drinking water sources and surface water in Slovenia.  
1105 *Int. J. Hyg. Environ. Health* 214, 392–398. <https://doi.org/10.1016/j.ijheh.2011.05.006>

1106 Stokdyk, J.P., Firnstahl, A.D., Walsh, J.F., Spencer, S.K., de Lambert, J.R., Anderson, A.C.,  
1107 Rezania, L.I.W., Kieke, B.A., Borchardt, M.A., 2020. Viral, bacterial, and protozoan  
1108 pathogens and fecal markers in wells supplying groundwater to public water systems in  
1109 Minnesota, USA. *Water Res.* 178, 115814. <https://doi.org/10.1016/j.watres.2020.115814>

1110 Sylvestre, É., Prévost, M., Burnet, J.B., Pang, X., Qiu, Y., Smeets, P., Medema, G., Hachad,  
1111 M., Dorner, S., 2021. Demonstrating the reduction of enteric viruses by drinking water  
1112 treatment during snowmelt episodes in urban areas. *Water Res.* X 11.  
1113 <https://doi.org/10.1016/j.wroa.2021.100091>

1114 Symonds, E.M., Nguyen, K.H., Harwood, V.J., Breitbart, M., 2018. Pepper mild mottle virus:  
1115 A plant pathogen with a greater purpose in (waste)water treatment development and public  
1116 health management. *Water Res.* 144, 1–12. <https://doi.org/10.1016/j.watres.2018.06.066>

1117 Tandukar, S., Sherchan, S.P., Haramoto, E., 2020a. Applicability of crAssphage, pepper mild  
1118 mottle virus, and tobacco mosaic virus as indicators of reduction of enteric viruses during  
1119 wastewater treatment. *Sci. Rep.* 10, 1–8. <https://doi.org/10.1038/s41598-020-60547-9>

1120 Tandukar, S., Sherchan, S.P., Haramoto, E., 2020b. Reduction of Pathogenic and Indicator  
1121 Viruses at a Drinking Water Treatment Plant in Southern Louisiana, USA. *Food Environ.*  
1122 *Virol.* 12, 269–273. <https://doi.org/10.1007/s12560-020-09436-y>

1123 Tandukar, S., Sherchand, J.B., Bhandari, D., Sherchan, S.P., Malla, B., Shrestha, R.G.,  
1124 Haramoto, E., 2018. Presence of human enteric viruses, protozoa, and indicators of

1125 pathogens in the Bagmati river, Nepal. *Pathogens* 7, 1–11.  
1126 <https://doi.org/10.3390/pathogens7020038>

1127 Teixeira, P., Costa, S., Brown, B., Silva, S., Rodrigues, R., Valério, E., 2020. Quantitative  
1128 PCR detection of enteric viruses in wastewater and environmental water sources by the  
1129 Lisbon municipality: A case study. *Water (Switzerland)* 12, 1–13.  
1130 <https://doi.org/10.3390/w12020544>

1131 The European Parliament and the Council of the European Union, 2020. Directive (EU)  
1132 2020/2184, EU (revised) Drinking Water Directive. Annex 1. Part B. *Off. J. Eur. Communities*  
1133 2019, 35.

1134 Tripathy, A.S., Sharma, M., Deoshatwar, A.R., Babar, P., Bharadwaj, R., Bharti, O.K., 2019.  
1135 Study of a hepatitis e virus outbreak involving drinking water and sewage contamination in  
1136 Shimla, India, 2015-2016. *Trans. R. Soc. Trop. Med. Hyg.* 113, 789–796.  
1137 <https://doi.org/10.1093/trstmh/trz072>

1138 Upfold, N.S., Luke, G.A., Knox, C., 2021. Occurrence of Human Enteric Viruses in Water  
1139 Sources and Shellfish: A Focus on Africa, *Food and Environmental Virology*. Springer US.  
1140 <https://doi.org/10.1007/s12560-020-09456-8>

1141 Van Alphen, L.B., Dorléans, F., Schultz, A.C., Fonager, J., Ethelberg, S., Dalgaard, C.,  
1142 Adelhardt, M., Engberg, J.H., Fischer, T.K., Lassen, S.G., 2014. The application of new  
1143 molecular methods in the investigation of a waterborne outbreak of norovirus in Denmark,  
1144 2012. *PLoS One* 9. <https://doi.org/10.1371/journal.pone.0105053>

1145 van den Berg, H.H.J.L., Friederichs, L., Versteegh, J.F.M., Smeets, P.W.M.H., de Roda  
1146 Husman, A.M., 2019. How current risk assessment and risk management methods for  
1147 drinking water in The Netherlands cover the WHO water safety plan approach. *Int. J. Hyg.*  
1148 *Environ. Health* 222, 1030–1037. <https://doi.org/10.1016/j.ijheh.2019.07.003>

1149 Varughese, E.A., Brinkman, N.E., Anneken, E.M., Cashdollar, J.L., Fout, G.S., Furlong, E.T.,  
1150 Kolpin, D.W., Glassmeyer, S.T., Keely, S.P., 2018. Estimating virus occurrence using  
1151 Bayesian modeling in multiple drinking water systems of the United States. *Sci. Total*  
1152 *Environ.* 619–620, 1330–1339. <https://doi.org/10.1016/j.scitotenv.2017.10.267>

1153 Vecchia, A.D., Kluge, M., da Silva, J.V. do. S., Comerlato, J., Rodrigues, M.T., Fleck, J.D.,  
1154 da Luz, R.B., Teixeira, T.F., Roehe, P.M., Capalonga, R., Oliveira, A.B., Spilki, F.R., 2013.  
1155 Presence of Torque Teno Virus (TTV) in Tap Water in Public Schools from Southern Brazil.  
1156 Food Environ. Virol. 5, 41–45. <https://doi.org/10.1007/s12560-012-9096-7>  
1157 Vellinga, J., Van der Heijdt, S., Hoeben, R.C., 2005. The adenovirus capsid: Major progress  
1158 in minor proteins. J. Gen. Virol. 86, 1581–1588. <https://doi.org/10.1099/vir.0.80877-0>  
1159 Verani, M., Federigi, I., Donzelli, G., Cioni, L., Carducci, A., 2019. Human adenoviruses as  
1160 waterborne index pathogens and their use for Quantitative Microbial Risk Assessment. Sci.  
1161 Total Environ. 651, 1469–1475. <https://doi.org/10.1016/j.scitotenv.2018.09.295>  
1162 Vieira, C.B., de Abreu Corrêa, A., de Jesus, M.S., Luz, S.L.B., Wyn-Jones, P., Kay, D.,  
1163 Vargha, M., Miagostovich, M.P., 2016. Viruses Surveillance Under Different Season  
1164 Scenarios of the Negro River Basin, Amazonia, Brazil. Food Environ. Virol. 8, 57–69.  
1165 <https://doi.org/10.1007/s12560-016-9226-8>  
1166 Vu, D.L., Bosch, A., Pintó, R.M., Guix, S., 2017. Epidemiology of classic and novel human  
1167 astrovirus: Gastroenteritis and beyond. Viruses 9, 1–23. <https://doi.org/10.3390/v9020033>  
1168 Wang, H., Naghavi, M., Allen, C., Barber, R.M., Carter, A., Casey, D.C., Charlson, F.J.,  
1169 Chen, A.Z., Coates, M.M., Coggeshall, M., Dandona, L., Dicker, D.J., Erskine, H.E.,  
1170 Haagsma, J.A., et al., 2016. Global, regional, and national life expectancy, all-cause  
1171 mortality, and cause-specific mortality for 249 causes of death, 1980–2015: a systematic  
1172 analysis for the Global Burden of Disease Study 2015. Lancet 388, 1459–1544.  
1173 [https://doi.org/10.1016/S0140-6736\(16\)31012-1](https://doi.org/10.1016/S0140-6736(16)31012-1)  
1174 WHO, 2004. Guidelines for Drinking-water Quality, 3rd Edition 1, 564.  
1175 WHO 2017, Guidelines for drinking-water quality: fourth edition incorporating the first  
1176 1149 addendum. Geneva: World Health Organization; 2017. Licence: CC BY-NC-SA  
1177 3.0 IGO  
1178 Williamson, W.M., Ball, A., Wolf, S., Hewitt, J., Lin, S., Scholes, P., Ambrose, V., Robson, B.,  
1179 Greening, G.E., 2011. Enteric viruses in New Zealand drinking-water sources. Water Sci.  
1180 Technol. 63, 1744–1751. <https://doi.org/10.2166/wst.2011.117>

1181 WWAP, U.N.W.W.A.P., 2017. The United Nations World Water Development Report 2017.  
1182 Wastewater: The Untapped Resource. Paris, UNESCO, The United Nations World Water  
1183 Development Report 2017. Wastewater: The Untapped Resource. Paris, UNESCO.  
1184 Xue, C., Fu, Y., Zhu, W., Fei, Y., Zhu, L., Zhang, H., Pan, L., Xu, H., Wang, Y., Wang, W.,  
1185 Sun, Q., 2014. An outbreak of acute norovirus gastroenteritis in a boarding school in  
1186 Shanghai: A retrospective cohort study. *BMC Public Health* 14, 1–7.  
1187 <https://doi.org/10.1186/1471-2458-14-1092>  
1188 Ye, X.Y., Ming, X., Zhang, Y.L., Xiao, W.Q., Huang, X.N., Cao, Y.G., Gu, K.D., 2012. Real-  
1189 time PCR detection of enteric viruses in source water and treated drinking water in Wuhan,  
1190 China. *Curr. Microbiol.* 65, 244–253. <https://doi.org/10.1007/s00284-012-0152-1>  
1191 Zhang, T., Breitbart, M., Lee, W.H., Run, J.Q., Wei, C.L., Soh, S.W.L., Hibberd, M.L., Liu,  
1192 E.T., Rohwer, F., Ruan, Y., 2006. RNA viral community in human feces: Prevalence of plant  
1193 pathogenic viruses. *PLoS Biol.* 4, 0108–0118. <https://doi.org/10.1371/journal.pbio.0040003>  
1194  
1195  
1196  
1197  
1198  
1199  
1200  
1201  
1202  
1203  
1204  
1205  
1206  
1207  
1208

1209 **Table captions**

1210 **Table 1.** Virus characteristics. AdV= adenovirus, HAV=hepatitis A virus, EV=enterovirus,  
1211 AiV=aichivirus, HEV=hepatitis E virus, SaV=sapovirus, NoV=norovirus, TTV=torque teno  
1212 virus, RoV=rotavirus, AstV=astrovirus, PyV=polyomavirus, PMMoV=pepper mild mottle virus,  
1213 TMV=tobacco mosaic virus.

1214 **Table 2.** Total number of articles that investigated each viral agent and cumulative  
1215 percentages of positive samples divided by type of water and type of virus. - = not assessed;  
1216 AdV = adenovirus; AiV = aichivirus; ASFLV = asfarvirus-like virus; AstV = astrovirus; CosV =  
1217 cosavirus; EV = enterovirus; HAV = hepatitis A virus; HCV = hepatitis C virus; HEV =  
1218 hepatitis E virus; KV = klassevirus; NoV = norovirus; PMMoV = pepper mild mottle virus; PyV  
1219 = polyomavirus; ReV = reovirus; RoV = rotavirus; SaliV = salivirus; SaV = sapovirus; TMV =  
1220 tobacco mosaic virus; TTV = torque teno virus.

1221 **Table 3.** Primary and secondary concentration methods reported in the analysed articles  
1222 (total articles= 79, two concentration methods were applied by the study of Kuroda et al.,  
1223 2015). TFF = tangential flow filtration; UF = ultrafiltration; F = filtration; F- = filtration with  
1224 electronegative charged membrane; F+ = filtration with electropositive charged membrane;  
1225 PEG = precipitation with polyethylene glycol; SMF = skimmed milk flocculation.

1226

1227 **Figure captions**

1228 **Figure 1.** Flow chart of study selection process.

1229 **Figure 2.** Number of articles that analysed SW-D, GW-D, DW or BW performed in each  
1230 continent (Africa, Asia, Europe, Oceania, North America, South America). Total articles = 79  
1231 (58 analysed one water type, 14 analysed two water types, 6 analysed three water types, 1  
1232 analysed four water types). Satellite image from European Space Agency.

1233 **Figure 3.** Percentages of positive samples in SW-D, GW-D and DW of the most searched  
1234 viruses (AdV, EV, NoV GI, NoV GII, RoV).

1235 **Figure 4.** Percentages of positive samples in DW of the most searched viruses (AdV, EV,  
1236 NoV GI, NoV GII, RoV) divided by continent.

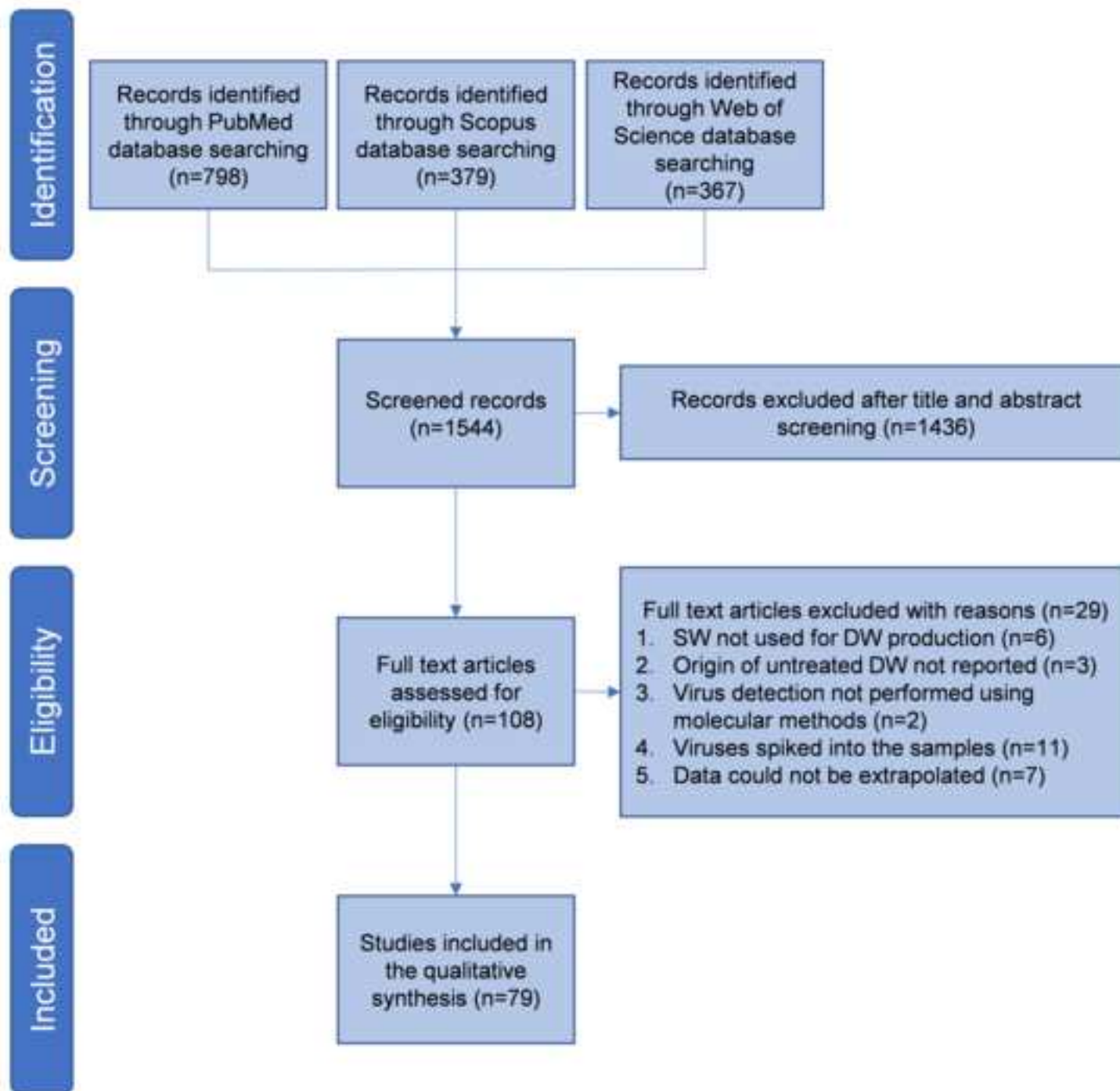


Figure 2

1  
2  
3  
4  
5  
6  
7  
8  
9  
10  
11  
12  
13  
14  
15  
16  
17  
18  
19  
20  
21  
22  
23  
24  
25  
26  
27  
28  
29  
30  
31  
32  
33  
34  
35  
36  
37  
38  
39  
40  
41  
42  
43  
44  
45  
46  
47  
48  
49

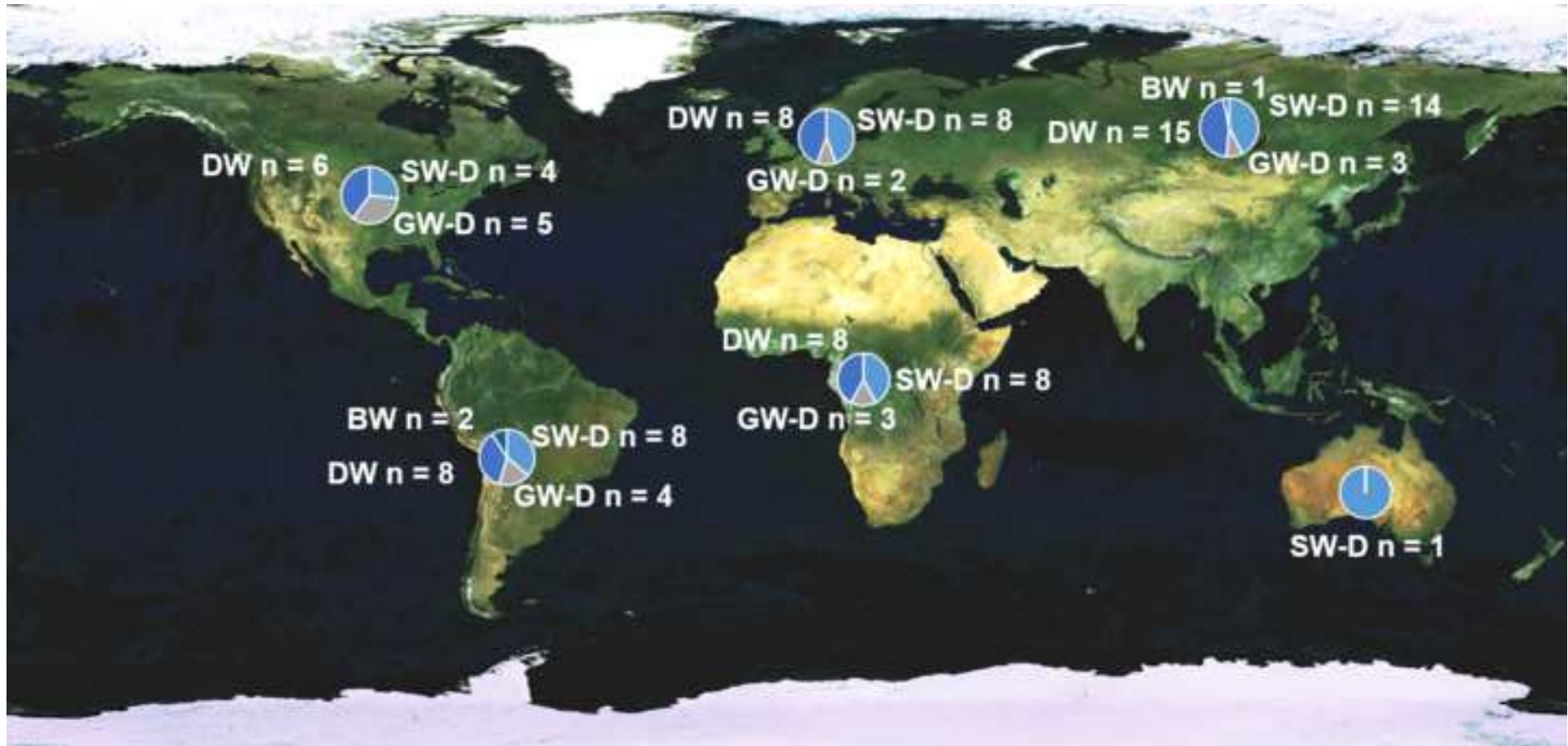


Figure 3

1  
2  
3  
4  
5  
6  
7  
8  
9  
10  
11  
12  
13  
14  
15  
16  
17  
18  
19  
20  
21  
22  
23  
24  
25  
26  
27  
28  
29  
30  
31  
32  
33  
34  
35  
36  
37  
38  
39  
40  
41  
42  
43  
44  
45  
46  
47  
48  
49

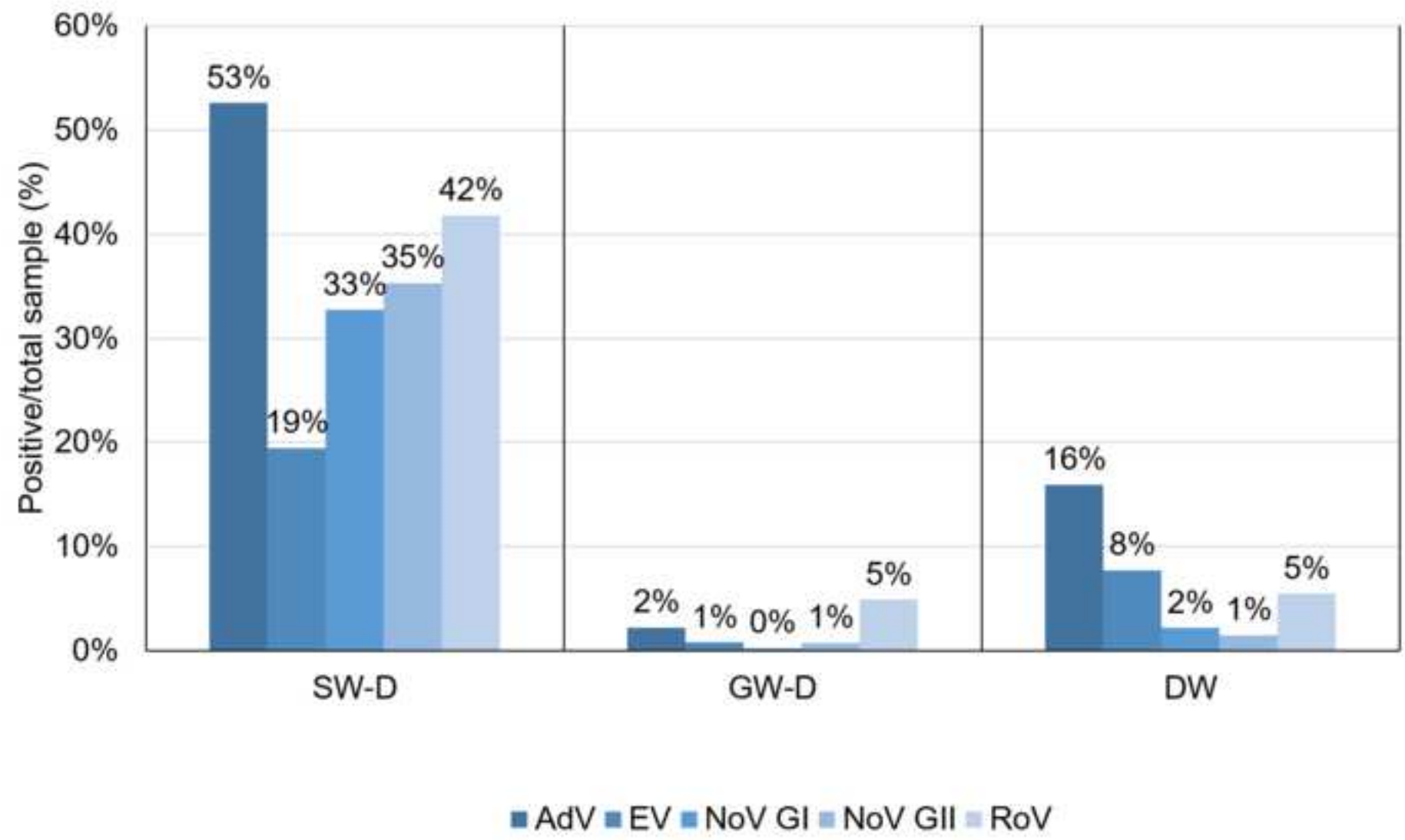
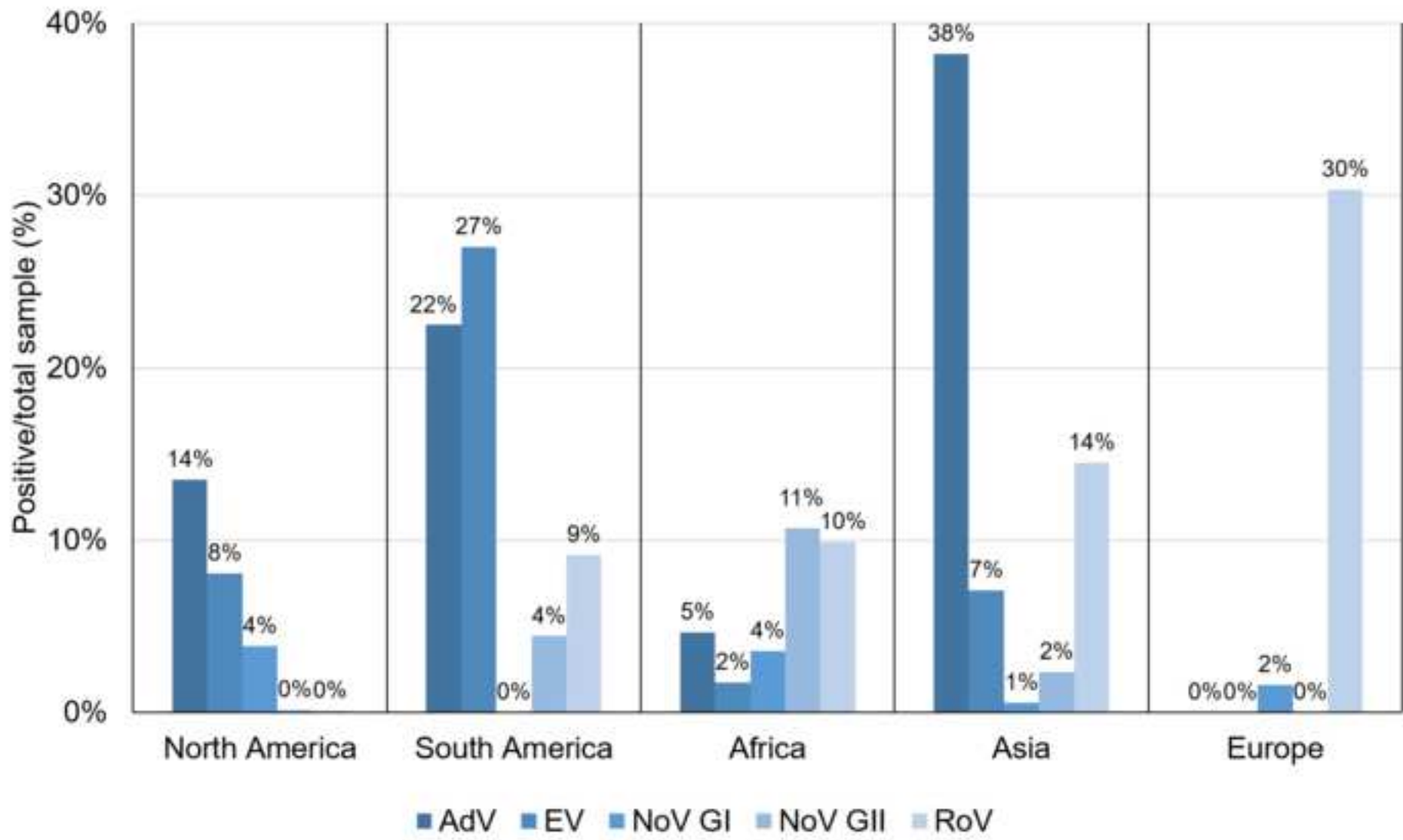




Figure 4

1  
2  
3  
4  
5  
6  
7  
8  
9  
10  
11  
12  
13  
14  
15  
16  
17  
18  
19  
20  
21  
22  
23  
24  
25  
26  
27  
28  
29  
30  
31  
32  
33  
34  
35  
36  
37  
38  
39  
40  
41  
42  
43  
44  
45  
46  
47  
48  
49



**Table 1.** Virus characteristics.

Virus type	Family	Genome	Human pathogens			
			Persistence in water supply <sup>a</sup>	Disease	Vaccine	Reference
AdV	Adenoviridae	double-stranded DNA	Long	pharyngitis, cystitis, gastroenteritis	yes	Broderick et al., 2017; Okoh et al., 2010; Upfold et al., 2021; Vellinga et al., 2005
AiV	Picornaviridae	single-stranded and positive-sense RNA	NA	gastroenteritis	no	Upfold et al., 2021
AstV	Astroviridae	single-stranded and positive-sense RNA	Long	gastroenteritis, respiratory diseases, encephalitis, meningitis, acute flaccid paralysis	no	Bosch et al., 2014; Upfold et al., 2021; Vu et al., 2017
EV	Picornaviridae	single-stranded and positive-sense RNA	Long	gastroenteritis, myocarditis, pericarditis, encephalitis	yes	Baggen et al., 2018; Li et al., 2021; Lugo and Krogstad, 2016
HAV	Picornaviridae	single-stranded and positive-sense RNA	Long	hepatitis	yes	Okoh et al. 2010; Smith and Simmonds, 2018; Upfold et al., 2021
HEV	Hepeviridae	single-stranded and positive-sense RNA	Long	hepatitis	yes	Larrue et al., 2021; Upfold et al., 2021
NoV	Caliciviridae	single-stranded and positive-sense RNA	Long	gastroenteritis	no	Carter et al., 2005; Okoh et al., 2010; Upfold et al., 2021
PyV	Polyomaviridae	double-stranded DNA	NA	progressive multifocal leukoencephalopathy, nephropathy, pulmonary infections, possible oncogenic viruses	no	Calgua et al., 2013
RoV	Reoviridae	double-stranded RNA	Long	gastroenteritis	yes	Crawford et al., 2017; Okoh et al., 2010; Steele et al., 2003; Upfold et al., 2021
SaV	Caliciviridae	single-stranded and positive-sense RNA	Long	diarrhoea, nausea, myalgia	no	Upfold et al., 2021
TTV	Anelloviridae	single-stranded and negative-sense DNA	NA	partially unknown, co-factor in several diseases	no	Charest et al., 2015; Jiménez-Melsiò et al., 2013; Shoeib et al., 2011

---

**Indicators**

---

<b>Virus type</b>	<b>Family</b>	<b>Genome</b>	<b>Host</b>	<b>Reference</b>
PMMoV	Virgoviridae	single-stranded and positive-sense RNA genome	plant	Haramoto et al., 2018; Kitajima et al., 2018
TMV	Virgaviridae	single-stranded and positive-sense RNA	plant	Tandukar et al., 2020a

---

AdV= adenovirus, HAV=hepatitis A virus, EV=enterovirus, AiV=aichivirus, HEV=hepatitis E virus, SaV=sapovirus, NA = not available, NoV=norovirus, TTV=torque teno virus, RoV=rotavirus, AstV=astrovirus, PyV=polyomavirus, PMMoV=pepper mild mottle virus, TMV=tobacco mosaic virus. <sup>a</sup> = detection period for infective stage in water at 20° C: short, up to 1 week; moderate, 1 week to 1 month,; long, over 1 month (WHO, 2017).

**Table 2.** Total number of articles that investigated each viral agent and cumulative percentages of positive samples divided by type of water and type of virus.

virus type	SW-D			GW-D			DW		
	positive/total	%	n° articles	positive/total	%	n° articles	positive/total	%	n° articles
<b>AdV</b>	755/1435	52.61%	29	102/4607	2.21%	13	535/3352	15.96%	29
<b>AiV</b>	50/114	43.86%	6	1/9	11.11%	2	16/142	11.27%	6
<b>ASFLV</b>	1/12	8.33%	1	-	-	-	-	-	-
<b>AstV</b>	48/132	36.36%	3	-	-	-	7/145	4.83%	3
<b>CosV</b>	-	-	-	-	-	-	9/18	50.00%	1
<b>EV</b>	106/544	19.49%	18	13/1667	0.78%	11	266/3439	7.73%	24
<b>HAV</b>	63/268	23.51%	7	0/994	0.00%	3	17/2335	0.73%	10
<b>HCV</b>	3/30	10.00%	1	-	-	-	-	-	-
<b>HEV</b>	26/176	14.77%	5	0/15	0.00%	2	24/80	30.00%	5
<b>KV</b>	2/12	16.67%	1	-	-	-	-	-	-
<b>NoV GI</b>	311/950	32.74%	21	4/1634	0.24%	10	97/4454	2.18%	20
<b>NoV GII</b>	463/1310	35.34%	26	13/1768	0.74%	11	71/4794	1.48%	22
<b>NoV GIII</b>	56/173	32.37%	2	-	-	-	-	-	-
<b>NoV GIV</b>	0/64	0.00%	1	-	-	-	-	-	-
<b>PMMoV</b>	273/320	85.31%	8	64/1078	5.94%	4	34/120	28.33%	7
<b>BK/JC/MC/KI/WU</b>	273/617	44.25%	11	15/1541	0.97%	6	22/170	12.94%	6
<b>PyV</b>									
<b>ReV</b>	0/16	0.00%	1	-	-	-	-	-	-
<b>RoV</b>	347/830	41.81%	14	57/1168	4.88%	4	201/3671	5.48%	18
<b>SaliV</b>	-	-	-	-	-	-	13/18	72.22%	1
<b>SaV</b>	5/144	3.47%	3	-	-	-	1/20	5.00%	1
<b>TMV</b>	8/12	66.67%	1	-	-	-	17/30	56.67%	2
<b>TTV</b>	16/79	20.25%	2	-	-	-	19/69	27.54%	3

- = not assessed; AdV = adenovirus; AiV = aichivirus; ASFLV = asfarvirus-like virus; AstV = astrovirus; CosV = cosavirus; DW = drinking water (water used for human consumption, not bottled); EV = enterovirus; GW-D = groundwater used as a source for DW production; HAV = hepatitis A virus; HCV = hepatitis C virus; HEV = hepatitis E virus; KV = klassevirus; NoV = norovirus; PMMoV = pepper mild mottle virus; PyV = polyomavirus; ReV = reovirus; RoV = rotavirus; SaliV = salivirus; SaV = sapovirus; SW-D = surface water used as a source for DW production; TMV = tobacco mosaic virus; TTV = torque teno virus.

**Table 3.** Primary and secondary concentration methods reported in the analysed articles (total articles= 79, two concentration methods were applied by the study of Kuroda et al., 2015).

Primary concentration	Secondary concentration	Number of articles	Reference
F-	None	17	Ahmad et al., 2018; Bortagaray et al., 2020; Canh et al., 2021; de Souza et al., 2018; Dos Santos et al., 2015; Gad et al., 2019; Kato et al., 2018; Kishida et al., 2012; Kluge et al., 2014; Miagostovich et al., 2020; Miura et al., 2019; Rashid et al., 2021; Rizk and Allayeh, 2018; Spilki et al., 2013; Staggemeier et al., 2015; Tandukar et al., 2020b; Vecchia et al., 2013
	UF	11	Asami et al., 2016; Assis et al., 2015; Da Silva Luz et al., 2020; Diston et al., 2015; Fongaro et al., 2015, 2013; Garcia et al., 2012; Haramoto et al., 2013, 2012; Kuroda et al., 2015; Tandukar et al., 2018
	PEG	1	Mackowiak et al., 2018
F+	None	10	Bonanno Ferraro et al., 2021; Ferrer et al., 2015; Iaconelli et al., 2017; Joung et al., 2013; Jung et al., 2011; Salvador et al., 2020; Silva et al., 2015; Steyer et al., 2011; Sylvestre et al., 2021; Varughese et al., 2018
	PEG	7	Grøndahl-Rosado et al., 2014; Kiulia et al., 2014; Opere et al., 2021; Pérez-Sautu et al., 2012; Potgieter et al., 2020; Shi et al., 2021; Ye et al., 2012
	F	1	Lee et al., 2018
	UF	1	Teixeira et al., 2020
F	UF	6	Chigor and Okoh, 2012a, 2012b; Dienus et al., 2016; Kuroda et al., 2015; Malla et al., 2019; Sangsanont et al., 2016
	None	5	Ferguson et al., 2012; Guerrero-Latorre et al., 2011; Hssaine et al., 2011; Kittigul and Pombubpa, 2021; Mattioli et al., 2013
	PEG	3	Borchardt et al., 2012; Gotkowitz et al., 2016; Lambertini et al., 2011
TFF	UF	1	Aw and Gin, 2011
	PEG	1	Marie and Lin, 2017
UF	PEG	5	Charest et al., 2015; Cuevas-Ferrando et al., 2020; Murphy et al., 2020; Stokdyk et al., 2020; Williamson et al., 2011
	UF	4	Gibson et al., 2011; Gibson and Schwab, 2011a, 2011b; Hata et al., 2021
	None	3	Jacob et al., 2015; Knappett et al., 2011; Shoeib et al., 2011
PEG	F-	1	Ahmad et al., 2015
SMF	None	3	Calgua et al., 2013; Gamazo et al., 2018; Vieira et al., 2016

TFF = tangential flow filtration; UF = ultrafiltration; F = filtration; F- = filtration with electronegative charged membrane; F+ = filtration with electropositive charged membrane; PEG = precipitation with polyethylene glycol; SMF = skimmed milk flocculation.

**Declaration of interests**

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.

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

## JOURNAL OF ENVIRONMENTAL SCIENCES

---

Editorial Department  
P. O. Box 2871  
Beijing 100085  
China  
Tel: 86-10-62920553  
E-mail: jesc@rcees.ac.cn

### Journal Publishing Agreement

Author(s): Marco Panizzolo, Marta Gea, Elisabetta Carraro, Giorgio Gilli,  
Silvia Bonetta, Cristina Pignata

Title: Virus occurrence in sources for drinking water  
production and in drinking water: a review

The undersigned authors, with the consent of all authors, hereby assign to *Journal of Environmental Sciences*, the copyright in the above identified article to be transferred, including supplemental tables, illustrations or other information submitted in all forms and media throughout the world, in all languages and format, effective when and if the article is accepted for publication.

Authors also agree to the following terms:

- A. The article submitted is not subject to any prior claim or agreement and is not under consideration for publication elsewhere.
- B. The article contains no libelous or other unlawful statements and does not contain any materials that violate proprietary right of any other person, company, organization, and nation.
- C. If the article was prepared jointly with other authors, the author(s) agree with the authorship sequence.

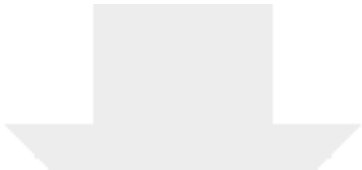
Please sign and date the document.

First Author: *Marco Panizzolo*

Corresponding author: *Marta Gea*

Date: *22/02/22*

Date: *22/02/22*



Click here to access/download  
**Supplementary Material**  
SM.docx

