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Review article

Overview about viruses in the aerosol of the wastewater treatment plants

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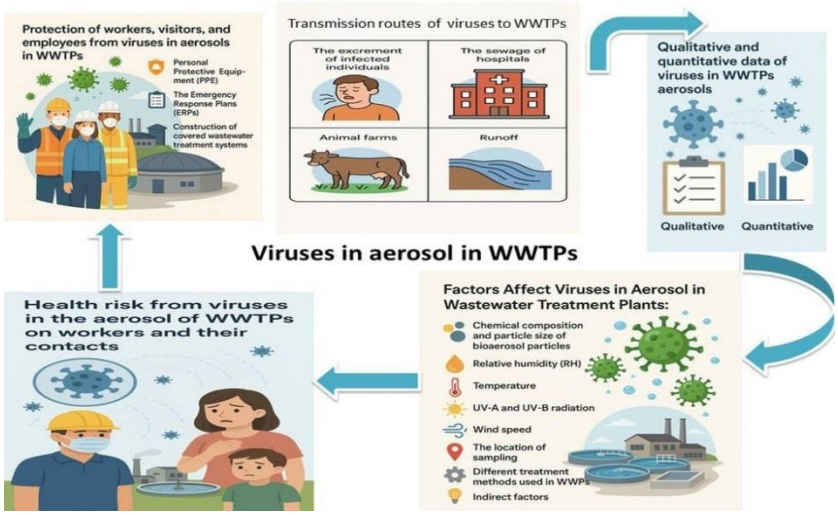
ABSTRACT

This study's first objective was to summarize the factors that affect the viability of the respiratory and enteric viruses in aerosols in wastewater treatment plants (WWTPs). Also, to determine the health hazards for workers in WWTPs and their contacts, and how to protect them and control viral spreading through their activities. The third objective was to analyze the efficiency of different virus concentration methods from aerosol samples. The review summarized sources of viruses in wastewater, such as human excreta from infected individuals, animal waste, untreated hospital wastewater in some developing countries, and the role of runoff in transporting viral contaminants from land to sewage systems. Also, the effect of environmental factors in treatment plants, such as relative humidity, temperature, wind speed, and the effect of natural ultraviolet (UV), were emphasized as a crucial determinants in the viability of viruses in the aerosols. The study also highlighted the health risks associated with aerosolized viral exposure to wastewater treatment workers, including respiratory and gastrointestinal diseases, and preventive measures such as emergency response plans (ERPs) and personal protective equipment (PPE) as recommended protection tools. Data about the efficiency of different sampling methods (active and passive) to concentrate viruses in aerosol on wastewater were summarized in this review. In conclusion, the development of the concentration methods for viruses in aerosol is greatly needed to increase the accuracy of qualitative and quantitative methods. Also, more research is needed to determine the best viral index for respiratory and enteric viruses in aerosols.

Graphical abstract



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1. Introduction

Aerosol is finely suspended liquid or solid particles in the air or gas that can spread for 8 meters, remain buoyant for hours, until falling. [1]. Another term, bioaerosol (biological aerosol), refers to aerosol particles of biological origin, such as microorganisms (e.g., bacteria, fungi, actinomycetes, and viruses), products of microorganisms (e.g., toxins, mycotoxins, and organic compounds), and active particles (e.g., allergens like pollen, a common aeroallergen), as well as spores and cellular fragments (e.g., DNA fragments or debris from plants or animals) [2,3]. Bioaerosols can be divided according to their source into natural bioaerosols, which originate from environmental origins (which have an essential role in the ecological system), like water (e.g., seas, oceans), soil, and plants (e.g., leaves). On the other hand, anthropogenic bioaerosols originate from anthropogenic origins and may contain pathogens that can affect human health. They are generated directly through human activities such as speaking, singing, laughing, coughing, breathing, or other respiratory activities, or through human-related processes in outdoor environments like water, wastewater treatment plants, and farms, or indoor environments, e.g., universities, prisons, health care centers, schools, restaurants, etc. [4, 5]. It is very important to differentiate between bioaerosol particles and droplets. Droplets are larger particles so settle down, causing infection directly or indirectly, at a distance > 1 m [6]. This differentiation provides important insights not only into the ability of these particles to travel and cause infection away from their source, but also into the severity of the infection. While aerosol particles are small enough to reach and infect the deep respiratory system, larger particles may settle in the gastrointestinal tract [7].

In our review, we focused on viruses in aerosol from wastewater treatment plants (WWTPs) as they are outdoor environments with a high risk of infection, the shortage of adequate techniques for sampling aerosol viruses, and the various strategies used to protect employees in WWPTs and manage viral aerosol.

2. Search Strategy

The materials used were previously published articles from various research groups working in the relevant field. Our methodology involved integrating and correlating the ideas presented in these studies to develop an independent perspective that effectively addresses the research objectives. The results of this review-based approach are presented in the following sections of the manuscript.

3. Key Points and discussion of integrated and correlated ideas

3.1. The transmission routes of viruses to WWTPs

The primary source of viruses in wastewater systems is the excrement of infected individuals from residential sewage. Patients with viral gastroenteritis can excrete approximately 10^5 to 10^{11} viral particles a day per gram of stool, including enteric viruses, such as rotaviruses, enteric adenoviruses, astroviruses, noroviruses, hepatitis A virus (HAV), and hepatitis E virus (HEV) [8-11]. Also, respiratory viruses e.g. Middle East respiratory syndrome coronavirus (MERS-CoV), severe acute respiratory syndrome coronavirus (SARS-CoV), severe acute respiratory syndrome coronavirus 2 **(**SARS-CoV-2), and influenza viruses could be detected in the sewage due to the excretions such as sputum [12], saliva [13], urine [14], and feces [15-18]. Another source is hospital sewage, which sometimes shows poor treatment efficacy, especially in developing countries. These viruses can preserve their viability partially until reaching WWTPs [19]. Animal farms are considered an important source of viral contaminants in sewage through animal feces [20]. A study compared the concentrations of chicken and turkey parvoviruses (ChPV/TuPV) in chicken stool samples and the downstream urban wastewater, indicating that ChPV/TuPV were detected in 73% of stool samples and 44% of downstream raw sewage samples, with a mean values of 9.07×10^8 genome copies (GC)/g and 2.65×10^2 GC/ml, respectively [21]. The high concentrations of ChPV/TuPV may explain how these pathogens can be introduced into the surrounding environment and the wastewater. Runoff, defined as water from rain, irrigation, or melted snow that runs over the land surfaces without penetrating the soil, can act as a carrier of human and animal viral pollutants. Reports indicated that SARS-CoV-2 could be transmitted to water systems like sewage [22]. Viruses such as enterovirus, adenovirus, and norovirus genogroup II (GII) have been collected from street runoff resulting from human excreta and represent a transport source of these viruses to drainage systems [23].

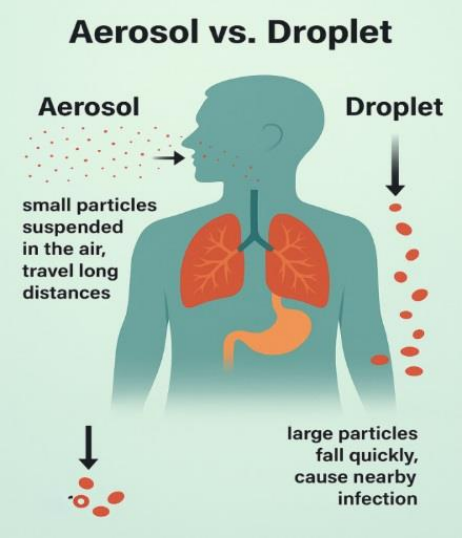

Figure 1: Aerosol versus Droplets. The illustration shows the difference between aerosols and droplets. Aerosols are small particles that remain suspended in the air and can travel long distances, whereas droplets are larger particles that quickly settle by gravity.

3.2. Factors affecting viruses in aerosols in wastewater treatment plants:

Many factors affect the bioaerosol formation from the source, the viability of viral aerosol, and the environmental factors of WWTPs (considered outdoor environments) that can affect the infectivity of the viruses [24, 25].

3.2.1. Factors affecting viral viability during survivability inside aerosol particles

3.2.1.1. Chemical composition of the bioaerosol particles:

Bioaerosol particles contain organic and inorganic pollutants from the emission sources, which are mainly anthropogenic [26]. These chemical pollutants may have a dual effect; first, the presence of organic matter in the surrounding layer may protect the viruses from the impact of environmental factors, increasing the viral infectivity for a long time [27]. Also, it may improve viral infectivity by causing stress on the respiratory tract and affecting the immune system of infected persons [28]. Second, organic compounds may go through photochemical reactions that produce oxidizing radicals that, in the case of non-enveloped viruses, cause damage to peptide bonds in surrounding proteins and subsequent structural change in the nucleobases of the nucleic acids like guanine, which compromise the virions' integrity, also reactive oxidant species react with the lipid envelope causing lipid peroxidation, which reduce the survival of enveloped viruses, so, the common chemical pollutants in aerosol particles may reduce the viability of associated virus [29].

In conclusion, the different effects of the chemical composition of aerosol particles on viruses may vary depending on the type of chemicals, their concentration, their position related to the viruses, and the type of viruses.

3.2.1.2. Particle size

The particle size of aerosol is a very important physicochemical factor that affects the viability of the viruses on two axes,

The first is the settlement rate: The smaller the particles, the longer they remain airborne [30], increasing the probability of being inhaled and causing viral infection [31]. It was reported that large aerosol particles (> 4.1μm) needed 33 min to settle in 1 meter, while particles < 4.1 μm, which contain 42% of influenza A virus (IAV) RNA and respiratory syndrome virus (RSV) RNA, needed 8 hrs. to cover the same distance [32]. The second is related to protection: generally, larger particles provide protective effects and increase viral infectivity, while viruses in the small particles are more exposed to viral inactivation due to the enormous energy of the particles' surface [33]. For example, rhinovirus preserved longer in large particles than in smaller particles [34]. Another study concerned with three viruses, porcine epidemic diarrhea virus (PEDV), IAV, and porcine reproductive and respiratory syndrome virus (PRRSV) showed higher viability in the large particle size [35].

Although viruses prefer large particles, some studies indicated that some viruses, such as adenovirus (AdV), survive better in smaller particles than the larger ones [33].

3.2.2. Environmental factors of WWTPs that affect the infectivity of the viral aerosol.

The environmental factors of WWTPs (as an outdoor environment) are generally related to weather elements like temperature, relative humidity, natural UV radiation (sunlight), and wind speed.

3.2.2.1. Relative humidity (RH):

Relative humidity is a very important factor affecting the viability of the viruses in the bioaerosol [36, 37], the enveloped viruses show more stability at low RH as containing high lipid contents which tend more stable at low relative humidity [38], it is very important to link RH with three physicochemical processes which are hygroscopy, deliquescence, and efflorescence that depend on RH and affect the bioaerosol particles and consequently the vitality of viruses. First, Hygroscopy occurs when particles absorb water from the ambient air without turning into solutions in moderate humidity. Second, Efflorescence, in which particles lose their water molecules to the atmosphere, so, their weight is reduced. It occurs in dry air when the water vapor pressure of the air is lower than the water vapor pressure of the bioaerosol particles. Third, Deliquescence occurs when particles absorb water from the air and form solutions at high humidity when the water vapor pressure in the air is higher than the water vapor pressure of the bioaerosol particles [39]. Under hygroscopic conditions, enveloped viruses (e.g., influenza viruses and SARS-CoV-2) may absorb excess water, which perturbs their lipid bilayer envelopes and thereby increases their susceptibility to environmental stressors such as chemical disinfectants and ultraviolet light [37]. In contrast, non-enveloped viruses, such as norovirus (NoV) and rotavirus (RoV), possess robust protein capsids that resist water-induced structural damage, thus conferring prolonged environmental survivability [40]. In Efflorescence, the lipid envelope in enveloped viruses disrupts in dehydration and reduces their infectivity. For example, the viability of the influenza virus dropped significantly in dry air [41]. Non-enveloped viruses had less effect because of their hardy capsids. Infectivity of NoV is retained even after desiccation [42]. Finally in Deliquescence, high humidity may dissolve the lipid in the enveloped viruses, causing viral inactivation. SARS-CoV-2 stability declined in highly humid environments [36], while non-enveloped viruses, their capsids withstand dissolution; however, exposure to liquid phases may degrade internal genetic material over time [43].

Also, at the level of dispersion, high RH ≥80% can facilitate the dispersion of bioaerosol over long distances, increasing the risk of viral infection transmission [1].

3.2.2.2. Temperature:

Non-enveloped viruses show long-term persistence at low temperatures [44]. In contrast, enveloped viruses are more sensitive to temperature because of the presence of the lipid bilayer membrane (envelope) [45], for example, a previous study on SARS-CoV-2 (enveloped RNA virus) showed that a low survival period in the sewage at temperatures ≥ 20 ◦C and a high survival period reached 14 and 17 days at low temperatures of 4 °C and 20 °C respectively [46].

A combination of more than one factor can reduce the viability and transmission of viruses. It was detected that the rise in temperature and humidity above 30 °C and 80% by 1 °C and 1%, respectively, together could reduce the number of effective reproduction (R-value) by about 0.026 in China and 0.020 in the USA, as the high temperature accelerates viral aerosol evaporation, so reduced viral stability and transmission efficiency [47]. The high humidity also leads to viral inactivation due to deliquescence. Although the reduction in R-value is not high enough to stop pandemics, it can be a control strategy in

areas where non-pharmaceutical interventions are not strictly enforced.

3.2.2.3. Wind speed:

Wind speed can affect the bioaerosol emission, distribution, and the elimination of biological materials from surfaces [48]. It was indicated that the high wind speed caused high aerosolization of pathogens near the surface layer of wastewater to the air [49]. Conversely, another study reported that wind speed could have a negative effect on viral dispersion due to the dilution and dispersion of the wind [50]. For example, Fracchia et al. [51] found low levels of airborne contaminants at the downwind locations due to the dilution of bioaerosols as the distance from the sources increased. Ranga [1] showed that wind which dilutes bioaerosol particles continuously minimizes their effect as a source of infection.

3.2.3. Other factors can affect the viral titer in the aerosol particles

3.2.3.1. The location of sampling

Bioaerosol concentration at sites of pre-treatment, grit chamber, and primary treatment was higher than other sites such as the secondary treatment and final sedimentation sites, as the raw sewage flowed continuously to the pre-treatment chambers causing the generation of bioaerosol then dispersed by the wind, than the secondary sedimentation or the disinfection units in the WWTPs, as the amount of pathogens decreases with the progress in the treatment steps [52].

3.2.3.2. Different treatment methods used in WWTPs

The emission of virus-laden aerosol in the activated sludge treatment method is higher due to the use of air pumps to promote aerobic digestion, which in turn causes breaking in the water surface than other treatment methods, e.g., Anaerobic digesters, which have no mechanical aeration, so reduce aerosolization risks [53].

3.2.4. Indirect factors

Some factors indirectly affect the viral presence in the aerosol. In the outbreaks, the high viral titer occurs in the wastewater basins compared to the regular days, increasing the viral titer in aerosol [15, 54]. Also, the sampling time of the season may be an essential factor. An epidemiological study has shown a higher titer of respiratory viruses such as influenza virus, RSV, human metapneumovirus, and human coronavirus in the air during winter [55]. This highlights that winter is a critical season in pathogen transmission, especially in temperate regions, which is associated with enhanced pathogen survival at lower temperatures [6].

All these factors affect the number of viruses in the wastewater units, which indirectly affects the occurrence of viruses in the bioaerosol. Also, each virus reacts differently to each factor or combination of factors [56].

3.3. Health risk from viruses in the aerosol of WWTPs on workers and their contacts.

As mentioned above, WWTPs are considered a primary source of emissions of bioaerosol [57], as the sewage in the WWTPs can contain a high load of viruses, especially viable ones that during the process of aeration at activated sludge tank or any mechanical agitation, can be aerosolized to the air forming viral aerosols. These viruses can cause viral infection to the workers by either the fecal-inhalation way of the aerosol particles or the fecal-oral way after contact with contaminated surfaces. There were studies on the health condition of sewage workers who had frequent symptoms like weakness, fatigue, headache, fever, dizziness, and respiratory diseases, which are known as "sewage workers' syndrome", compared to non-sewage workers, and explaining it due to the pathogens in the aerosol [58]. Workers have a higher probability of being infected with several respiratory and enteric diseases with viral causes such as gastroenteritis (by rotaviruses, astroviruses, enteric adenoviruses, and noroviruses), hepatitis (by hepatitis A and E viruses), myocarditis, conjunctivitis, rash, and maybe diabetes (by Coxsackieviruses A and B and echoviruses), influenza (by different types of influenza viruses), SARS (by SARS-CoV and SARS-CoV-2) in regular periods and even in pandemics [59]. Although persons must be exposed to minimal infectious doses of viruses to be infected, regular exposure (daily, weekly, monthly) may achieve these doses for respiratory diseases (by inhalation) and even enteric diseases by oral route, aerosol entrance, or dealing with contaminated surfaces [60, 61]. It has been shown that the places surrounding the units in the wastewater treatment plants contained up 2×10^6 GC/m^3 of AdV in the air samples [62]. The problem is not only workers may have an exposure rate to these diseases but also their families or individuals in their social environment may have the same exposure rate if they do not follow the safety instructions during their working time [63]. Many transmission routes from workers to their contacts were reported, such as direct contact, contaminated clothes, food, drink, and belongings [64].

Another problem is that WWTPs are reservoirs for human and animal viruses, due to the transmission of viruses (pathogens) from animal feces to sewage via runoff. The problem is that the zoonotic nature of some viruses, which naturally infect animal species and can cross the species barrier from animals to humans, such as HEV-3 and HEV-4, and avian influenza [65, 66]. So these zoonotic viruses may represent an extra health risk for workers in addition to the risk of their exposure to human viruses.

3.4. Protection of workers, visitors, and employees from viruses in aerosols in WWTPs.

The effect of drugs to treat viruses doesn't cover all viruses; only a few drugs for a few viruses were discovered and approved, such as sofosbuvir (sovaldi) drug for hepatitis C [67] and acyclovir drug for herpes simplex virus (HSV) [68]. On the other hand, although vaccine technology is very advanced now, fast mutation, emergencies, and new strains may limit these means of defense [69-72]. So let us return to the golden rule: Prevention is better than cure. Depending on that, WWTP workers and their contacts are groups exposed to high threat during their daily work by exposure to many enteric and respiratory microbes (viruses, bacteria, fungi, parasites). So, the protocols that protect workers and consequently their contacts are of great concern. This may be the first barrier for preventing viral spreading and also goes in parallel with vaccination as a means of deep defense and protection.

There are multiple preventive and precautionary measures available to minimize the chances of contracting the viral infection, such as personal protective equipment (PPE), emergency response plans (ERPs), and construction of covered wastewater treatment systems [73-75], which we explain in some detail as follows:

3.4.1. Construction of covered wastewater treatment systems

A study was conducted on the wastewater treatment sectors of three hospitals: Jinyintan Hospital, the first one receiving COVID-19 patients; Wuchang Cabin Hospital, a temporary hospital that was opened to receive COVID-19 patients; and Huoshenshan Hospital, built emergently to receive COVID-19 patients, in Wuhan, China. The study reported that SARS-CoV-2 was found in all aerosol particles and soil surrounding the wastewater treatment tank in Huoshenshan Hospital and Jinyintan Hospital. At the same time, there were negative results for SARS-CoV-2 in the aerosol particles or the surrounding soil in Wuchang Cabin Hospital because the wastewater treatment unit is an enclosed system that prevents aerosolization of aerosol containing SARS-CoV-2 from the wastewater [76].

According to these results, the enclosed wastewater treatment system can be an effective solution, as it is designed to contain an enclosed or isolated structure to protect against viral spread over long distances. However, applying this solution to all wastewater treatment plants worldwide is difficult because of the high cost and long duration of construction. This may represent a big challenge, especially in developing countries.

3.5. Effect of solar radiation on viral survivability in aerosol of WWTPS

Although WWTPs use physical/chemical and/or biological processes to treat their effluents [77], it is also essential to try to reduce the possible risk of viruses in aerosol by different strategies. Several studies discussed the effect of natural ultraviolet (UV) on viruses in aerosol. Senatore et al. [27] reported that one of the promising strategies is the effect of UV-A from the sun as it requires no cost, can be useful in outdoor environments like WWTPs, and is available most days of the year, especially in tropical [sunny) countries or semi-arid countries with a sunny climate**.** Ali et al. [24] found that the survival of airborne pathogens, e.g., viruses, is greatly negatively affected by solar radiation because pathogens can be inactivated by ultraviolet irradiation. As a result, this causes the death of pathogens in bioaerosols. It was also reported that UVA at 513.30 J/cm^2 helped in virus inactivation, reducing aerosolizing Escherichia phage T4 (T4 phages) to 20% [78]. UV-B (λ=280–315 nm) and UV-A (λ=315–400 nm) solar radiation could penetrate the Earth's atmosphere to a certain degree, reducing the risk of viral transmission, especially in the Mediterranean region, as solar radiation is abundant [79]. From these results, we could conclude that countries with intense sunlight on most days of the year have a high opportunity to use this solar energy to inactivate viruses in aerosol in WWTPs. The recommended distance away from human activities must be followed by open (not covered) WWTPs.

More studies are needed to evaluate the effect of UV-A radiation on a wide range of viruses in correlation to the impact of different environmental factors such as seasonal variation in an open environment.

3.6. Qualitative and quantitative data of viruses in WWTPs aerosols.

The techniques used in viral sampling from the aerosols of WWTPs are considered a crucial phase in the process of viral investigation. The air is a biologically diluted environment, so it usually contains fewer pathogens than other matrices like raw wastewater in WWTPs. So it is essential to concentrate viruses from aerosol samples before any analysis [80]. Many studies on different sampling methods were performed. These studies included the active methods, which used mechanical components (pumps, power requirements, and flow monitoring to quantify the viral concentration), and the passive methods, which depend on natural processes or environmental factors without using active mechanical systems to allow viral aerosols to accumulate or concentrate [81]. The studies conducted on viruses in the aerosol of the WWTPs in the last fifty years with different sampling methods are summarized in Table 1.

Table 1 outlines active and passive sampling methods for collecting viral particles from aerosols in WWTPs. Active methods include impingers (e.g., Biosampler Impinger or All-Glass Impinger [AGI]), cyclones, impactors (single-stage or multi-stage), large-volume air samplers (LVAS), filters (polycarbonate, gelatin, or nylon), and high-volume electrostatic precipitator samplers such as the Large Electrostatic Aerosol Precipitator (LEAP). Although condensation samplers represent another active method, none have yet been applied to WWTP environments.

Passive sampling encompasses sedimentation on open plates containing agar or filters as collection media. Table 1 also summarizes the limitations inherent to methods and their impact on sampling efficiency. In Brenner et al.'s study [85], coliphages were recovered using a Biosampler, whereas animal viruses were undetected. This failure may be due to:

1. Unlike the abundant coliphages, insufficient animal virus concentrations in the influent generate detectable aerosols by the XM2 Biological Sampler.
2. The sampling duration was too short to capture low-abundance animal viruses.
3. Particle size distributions (< 2 μm or > 12 μm) of animal viruses which not retained by the XM2.

Biological Sampler needs redesign of the inlet geometry and airflow dynamics. Moreover, air passages accommodating 12 μm particles could not be sterilized.

Another limitation involves the small air volume collected by samplers, reducing sensitivity for low airborne virus concentrations [62]. It was estimated that the expected concentration of animal virus in aerosol is approximately 6.5×10^{-5} MPN m^{-3}. Therefore, at least 3.5×10^3 m^3 of air must be sampled to isolate animal viruses at rates comparable to coliphages. However, the maximal sampled volume by the sampler was only 1.2×10^2 m^3, which was insufficient for such detection [83].

Table 1. Summary of viruses sampling in the aerosol of WWTPs

Sampling method	Location	Time of Sampling & Duration	Virus	Sampling Flow rate & air volume	Pos./T. samples	Genome copies/m^3	Number of infectious unit (CPU/m^3) /(PFU/m^3)	Comment	Year of publication	Ref.
Andersen viable type samplers	A spray irrigation site with wastewater in Fort Huachuca, Arizona	From 19 to 31 October during day and night periods	Coliphage f2	28.3l L/min	NR	NR	-	Coliphage F2 was detected in air samples collected up to 563 m. downwind from the spray irrigation source	1975	[82]
A high-volume electrostatic precipitator samplers				1000L/min						
Large Volume Air Samplers (LVAS)	Two WWTPs/south eastern Michigan.	-	animal viruses	The maximum volume of air sampled was 1.20×10^2 m^3	NR	ND	-	The mean animal virus concentration per liter was about 3.6-3.7 logs lower than the mean coliphage level. The expected airborne animal virus concentration would be approximately 6.2×10^{-5} or 6.5×10^{-5} MPN m^{-3}.	1976	[83]
			E. coli C3000 phages			3.0×10^{-1} MPN m^{-3}				
			E. coli KI2HfrD phages.			2.3×10^{-1} MPN m^{-3}				
Large-volume scrubbers (LVS)	The O'Hare Water Reclamation sewage plant/ Chicago, USA	During the day (800 to 1959 hrs.) and night (2000 to 759 hrs.).	Enteric viruses (coxsackievirus B-1)	0.6 to 0.9 m^3/min. Air volume 30-60 m^3 for coliphage & 400-600 m^3 enteric viruses	2/9	-	4.7×10^3 to 1.0×10^2 at <150 m	EVs were detected during the night only within 150 m, of the operating aeration tanks. No viruses were detected during the daytime in air sample volumes of up to 428 m3. In general, the densities of microorganism-containing aerosols were higher at night than during the day.	1985	[84]
			Coliphages		-		7.3×10^2 at >150 m 4.9×10^2 at 150 to 250 m 7.6×10^2 at >250 m			
XM2 Biological	The Muskegon		Coliphages	1,050	-	-	From 0 to 9 PFU/m^3	Coliphages were recov-	1988	[85]

Sampler/Collector.	County Wastewater Management System Number 1 spray irrigation site /Michigan, USA	From May to August in addition to October Two 2-h samples were collected.	Animal viruses	L/min.	ND	ND	ND			ered from Muskegon aerosols with this sampler, animal viruses were not, coliphages, present in much larger quantities in the wastewater than animal viruses.		
Impactor sampler with contact agar plates	WWTPs/Pisa, Italy	Twice-monthly from May 1992 to April 1993	Coliphages	NR	-	-	At 2 meters	At 20 meters	At 30 meters	The sample volume was 0-9 m3 of air. Enteroviruses were detected in 25% of the aerosol samples, and positive were only from August to November. And detection only at a height of 2 meters.	1995	[86]
							8-13 pfu/m^3, summer & 0-3 pfu/m^3, winter	1-4 pfu/m^3, summer & 1-4 pfu/m^3 October only	ND			
Sedimentation on open Petri dishes method (30 min exposure, about 50 cm from ground level)	Three WWTPs/ in the City of Leghorn, Livorno, Italy	Monthly aerosol samples were collected from January to November 1996	Enterovirus	-	9%	-	-			The virological analysis has been only qualitative. Enterovirus is always in conjunction with reovirus.	1999	[87]
			Reovirus		46%							
Bio Sampler liquid Impinger	Seven Finnish wastewater	A nine-month	Somatic coliphages	12.5 L/min	-	-	Max. Conc. 380 pfu/m^3			-	2009	[88]
			F-specific coliphages		-	-	Max. Conc. 70 pfu/m^3					
Inhalable GSP samplers with polycarbonate filters	WWTP in / Copenhagen, Denmark	27 May 2010	NoV-GI	For 242 min	1/1	1,420	-			The GSP samplers used in the study have not been applied for the collection of airborne viruses but have a high sampling efficiency for particles with aerodynamic diameters <50 μm at both high and low wind speeds	2011	[89]
			NoV-GII		0/1	ND						
			AdV		0/1	ND						

Gelatin filters embedded in standard cassettes	The 79 WWTPs in the Canton of Zurich/ Switzerland	Once in winter and once in summer	AdV	4 L/min	104/123	$2.27x10^6$	-	NoV levels are lower than those to AdV because NoV is known to survive very well in adverse environments. HEV was found in very low concentrations (not quantifiable) in wastewater compared to the waterborne concentrations of AdV and NoV, and therefore aerosolization was less probable, also be explained by the small collected volume of air which limits the probability of detecting low airborne virus concentrations.	2014	[62]
			NoV-GII		3/123	$6.55x10^2$				
			HEV		0/123	ND				
An impinger (AGI-4)	3 sites 1- Clermont-Ferrand WWTP, 2- Agricultural area irrigated with WW and, 3- Puy de Dôme Mountain summit (15 km southwest of the WWTP)/ France	3/7/2014, 26–27/3/2015, 21/5/2015, 25–26/6/2015, 30/7/2015 The sampling duration ranged from 5 to 24 hr, depending on the weather conditions (shorter periods on hot days for reduced evaporation)	NoV	4-6 L/min	-	$>10^3$	-	- Norovirus were the most abundant enteric viruses found in water, at a concentration up to 10^6 gc/l. - Other enteric viruses such as rotavirus or enteroviruses were detected in water but their quantities found were low and close to the detection limit. - HEV was detected in all sampled compartments (water, air, cloud, raw, treated wastewater, water from decantation pond, and the air above irrigated fields)	2017	[90]
			HEV			NR				
Petri dishes containing different filters			-	-	-	-	-			
Inhalable samplers with nylon filters	The new full-scale pilot WWTP at Herlev Hospi-	On May 27 and June 23, 2015.	NoV-GII	3.5 L/min	-	At the wastewater outlet air / At the air exhaust	-	Higher levels of aerosolized NoV detected during sampling in May and June are therefore likely to	2017	[91]

	tal in the Capital Region of Denmark	The average sampling period is 409 min.	NoV-GI		-	15.5 ND	1.8 -		occur during winter when NoV epidemics occur in the population.		
Gelatin filters	The sludge was obtained from the Christiansburg Wastewater Treatment Plant (WWTP) in Virginia.	-	MS2 bacteriophage Phi6 Bateriophage	2 L/min	-	-		Aeration basin 547 PFU /min 3.8 PFU /min	Sewer pipes 79 PFU /min 0.3 PFU /min	Aeration basin and sewer pipe are lab-scale models. Also, MS2 and Phi6 are Ebola virus surrogates	2017	[92]
Liquid cyclone	4 different wastewater treatment centers /Eastern Canada	During the summer Cyclone for 10 min and 5hrs. for Impactor.	RoV HAV	200 L/min for a total of 2 m^3 of air/sample	6/10 1/10	$7.04x10^4$ 4.7×10^3		-		The genome copies were expressed by the mean of the values.	2018	[93]
Cascade Impactor (8 stages with filters as collection media)			RoV HAV	2 L/min, for a total of 0.6 m^3 collected	7/8 0/8	$1.26x10^6$ ND		-				
A mixed cellulose membrane in an active air sampler	wastewater treatment plant /Japan	November, December 2007 January 2008	NV GII NV GI AdV F-RNA bacteriophages GIII Enteroviruses	4 L/min	9/16 6/16 4/16 3/16 3/16	$3.2\ 10^3$ NR NR NR NR		-		The active air sampler is capable of testing 0.7–1.6 m^3 air after 3–7 h sampling with a detection limit of 10^2 copies/m^3 air in the field.	2019	[94]
Impinger	The west of Tehran/ Iran	12 months (9:00 AM-13:00 PM)	NoV RoV	4L/min	-	27 3099		-		-	2019	[95]
Dry Filter Continuous Air Sampler	Near open wastewater canal/ in Kanpur/ India	From May to August	Pan-enterovirus AdV (A−F) NoV-GI NoV-GII MS2	200 L/min with air volume 47.5 m^3 in La Paz , 36.3 m^3 in Kanpur and 28.3 m^3 in Atlanta	0/53 0/53 33/53 1/53 1/53	- - 320 150 NR		-		-	2021	[96]
	Near open wastewater canal/ La Paz, Bolivia	From December to March and from May to August	Pan-enterovirus AdV (A−F) NoV-GI NoV-GII		3/75 1/75 3/75 3 /75	NR NR 13 2.4						

			MS2		0/75	-				
	Near open wastewater canal in Atlanta, GA/ USA	From March to January	NF		0/15	-				
All-glass Impingers	Two WWTPs/ Isfahan, Iran	From 4March to 17March, 2020	SARSCoV-2	7.5–8.5 L/min and air samples (3500-4500 L)	6/15	Range: 5-188	-	-	2021	[97]
A cyclone-based Coriolis μ impinger	Five different wastewater	In March 2021, 10 min. for cyclone & 20 min. for impactor	HAdV	100 L/min	12 / 26	7.03×10^3	1.54×10^3	-	2022	[98]
			HBOV		2 / 26	8.1×10^2	4.73×10^1			
			RoVs		9 / 26	1.52×10^4	1.08×10^3			
			NoV-GI		2 / 26	1.06×10^3	1.05×10^2			
			NoV-GII		6 / 26	2.22×10^3	3.97×10^2			
			IAV		0 / 26	BDL	BDL			
			SARS-CoV-2		0 / 26	BDL	BDL			
			SARS-CoV-2/P		8 / 26	5.32×10^3	2.44×10^2			
A single-stage impactor			HAdV		8 / 26	4.73×10^3	9.48×10^2			
			HBOV		0 / 26	BDL	BDL			
			RoVs		5/26	3.89×10^3	8.74×10^2			
			NoV-GI		0 / 26	BDL	BDL			
			NoV-GII		0 / 26	BDL	BDL			
			IAV		0 / 26	BDL	BDL			
			SARS-CoV-2		0 / 26	BDL	BDL			
			SARS-CoV-2/P		5 / 26	4.5×10^2	6.05×10^1			
Cyclone (a Coriolis μ) air sampler	WWTP /Catalonia	Winter and summer. Sampling was for 60 min.	SARS-CoV-2	300 L/min	0/10	ND	-	-	2024	[59]
			HAdV		6/10	$4.41x10^3$				

Notes: Pos.: positive. T: total. AdV: adenovirus. NoV-GI: norovirus genogrouping I. NoV-GII: norovirus genogrouping II. NR: not reported. ND: not detected. HEV: hepatitis E virus. WWTP: wastewater treatment plant. RoV: rotavirus. MPN: most probable number. BDL: below the detection limit. SARS-CoV-2: severe acute respiratory syndrome coronavirus 2. SARS-CoV-2/P: presumptive SARS-CoV-2 positive/other coronaviruses positive. HBoV: human bocavirus. HAdV: human adenovirus. IAV: Influenza A virus. PFU: plaque-forming unit. CPU: cytopathogenic unit. Max. Conc.: maximum concentration.

Sampler design and virus type further influence the recovery rate. Comparing cyclone (filtration) and impactor (impaction) systems for rotavirus aerosol capture revealed that both samplers have limitations when considering their use and induce particle loss. For impactors, the bouncing of particles on the surface can lead to the destruction of collected particles and a decrease in collection efficiency. In turn, the cyclonic samplers, evaporation of the sampling liquid, and reaerosolization of already trapped particles may impact the sampling results. Rotavirus was detectable more frequently with the impactor compared to the cyclone. As rotavirus is a double-stranded RNA virus, there is a chance that samplers with a flow rate higher than 200L/min tend to lead to the degradation of the RNA virus species during collection. [93]. On the other hand, when the flow rate is the same for both samplers, 100 L/min (< 200 L/min), the recovery of viruses from aerosol of WWTPs using a cyclone was greater than the recovery using impactor. Although impactor operates at longer sampling times, surface "bounce" destroys the viral particles, so reducing its efficiency [98]. While passive methods are cost-effective and facile for broad site screening, the lack of quantitative analysis and the failure to detect real airborne aerosols represent limitations for these methods [87].

No standard aerosol-virus collection protocol has existed since 1982 and until 2025 [99]. Current methodologies must be developed to improve efficiency of sampling methods. Future studies must address factors affecting viral-aerosol sampling techniques, such as optimizing collection media beyond empirical experiences, enhancing sampler efficiency across virus size ranges, and minimizing re-aerosolization, particle bounce, and inlet/wall losses of the samplers aiming to establish standardized procedures [100].

On the other hand, some studies [83,101] have suggested bacteriophages as a suitable indicator of airborne animal viral contamination from WWTPs than coliform bacteria, as they behave similarly to the enteric viruses, are more resistant to environmental stresses, and are more stable in the air than coliforms. Relying on coliform bacteria as indicators of viral contamination in WWTPs may lead to an inaccurate assessment of the actual presence of viruses. This is supported by findings showing that coliform recovery was significantly reduced under low wind velocity, high ambient air temperature, and increased distance from the emission source. In contrast, coliphage recovery was not found to be affected by these environmental conditions. Numerous studies have also demonstrated the failure of indicator bacteria to detect viral contamination in wastewater [102,103]. The presence of bacteriophages in aerosol samples, alongside the absence of animal viruses despite their confirmed presence in the wastewater of WWTPs [83, 85] raises questions about the accuracy and validity of proposing bacteriophages as reliable indicators of airborne animal viral contamination. These studies have attributed the possible failure to detect animal viruses in aerosol samples to the low sensitivity of the sampling methods. Still, the detection of only bacteriophages, which were sampled with the same methods, may indicate the unsuitability of bacteriophages as an index of enteric viruses in aerosols of WWTPs. This may be explained by the high number of bacteriophages in the aerosol samples in parallel to a low number or complete absence of the enteric viruses in the same samples [104]. Moreover, some studies have detected the presence of enteric viruses (coxsackievirus B-1) in aerosol samples and the absence of bacteriophages [84]. Other studies have demonstrated the efficiency of bacteriophages and adenoviruses as indices of viral contamination in wastewater [105-107]. Adenoviruses have also not been proven to be a reliable index for viral contamination in air samples. Several studies have reported the non-detection of adenovirus despite the presence of NoV-GI, NoV-GII, Pan-enterovirus, and MS2 bacteriophage [89, 96], thereby limiting its accuracy and reliability as a biological indicator for airborne viral contamination. More studies are needed to select a suitable index for viral pollution in the aerosol samples of WWTPs.

4. Conclusion

From this review, we could conclude that the development of concentration methods of viruses in aerosol is a critical point to increase the accuracy of qualitative and quantitative methods and consequently to have accurate results about the presence/ absence of respiratory and enteric viruses in aerosol of WWTPs in addition to number of genome copies, infectious units and different viral genotypes in aerosols. Also, more research is needed to determine the suitable viral index for respiratory and enteric viruses in aerosols of WWTPs. Natural UV radiation may affect the viability of viruses in aerosols. Finally, protection tools for WWTP workers may decrease the spread of the viruses and, consequently, the spread of viral diseases for their contacts.

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References

1. U. Ranga, *SARS-CoV-2 aerosol and droplets: an overview*. VirusDisease. 32(2021) 190–197. https://doi.org/10.1007/s13337-021-00670-0.
2. J. Mandal, H. Brandl, *Bioaerosols in indoor environment—a review with special reference to residential and occupational locations.* Open Environ. Biol. Monit. J. 4(2011) 83–96. https://doi.org/10.5167/uzh-53633.
3. H. Mbareche, M. Veillette, G. J. Bilodeau, C. Duchaine, *Fungal aerosols at dairy farms using molecular and culture techniques.* Sci. Total Environ. 653(2019) 253–263. https://doi.org/10.1016/j.scitotenv.2018.10.345

4. S. Saibu, I. U. Perera, S. Suzuki, X. Rodó, S. Fujiyoshi, F. Maruyama, *Resistomes in freshwater bioaerosols and their impact on drinking and recreational water safety: a perspective*. Environ. Int. 183(2024) 108377. https://doi.org/10.1016/j.envint.2023.108377.
5. X. Feng, X. Xu, X. Yao, Y. Zhao, Y. Tang, Z. Zhao, Y. Wei, T. Mehmood, X. S. Luo, *Sources, compositions, spatio-temporal distributions, and human health risks of bioaerosols: A review.* Atmos. Res. 294(2024) 107453. https://doi.org/10.1016/j.atmosres.2024.107453.
6. Z. Y. Han, W. G. Weng, Q. Y. Huang, *Characterizations of particle size distribution of the droplets exhaled by sneeze.* J. R. Soc. Interface. 10(88)(2013) 20130560. https://doi.org/10.1098/rsif.2013.0560.
7. J. H. Brown, K. M. Cook, F. G. Ney, T. Hatch, *Influence of particle size upon the retention of particulate matter in the human lung*. Am. J. Public Health Nations Health 40(4) (1950) 450–480. https://doi.org/10.2105/AJPH.40.4.450.
8. R. Sinclair, S. A. Boone, D. Greenberg, P. Keim, C. P. Gerba, *Persistence of category A select agents in the environment.* Appl. Environ. Microbiol. 74(3)(2008) 555–563. https://doi.org/10.1128/AEM.02167-07.
9. A. I. Okoh, T. Sibanda, S. S. Gusha. *Inadequately treated wastewater as a source of human enteric viruses in the environment.* Int. J. Environ. Res. Public Health. 7(6)(2010) 2620-37.
10. Y. Aoki, A. Suto, K. Mizuta, T. Ahiko, K. Osaka, Y. Matsuzaki, *Duration of norovirus excretion and the longitudinal course of viral load in norovirus-infected elderly patients*. J. of Hospital Inf. 75(1)(2010) 42-6. https://doi.org/10.1016/j.jhin.2009.12.016
11. G. La Rosa, M. Fratini, S. della Libera, M. Iaconelli, M. Muscillo. *Emerging and potentially emerging viruses in water environments.* Annali dell'Istituto superiore di sanità. 397(2012) 406.
12. A. R. Branche, E. E. Walsh, M. A. Formica, A. R. Falsey, *Detection of respiratory viruses in sputum from adults by use of automated multiplex PCR*. J. of clinical Mic. 52(10)(2014) 3590-3596.
13. K. K. To, L. Lu, C. C. Yip, R. W. Poon, A. M. Fung, A. Cheng, D. H. Lui, D. T. Ho, I. F. Hung, K. H. Chan, K. Y. Yuen, *Additional molecular testing of saliva specimens improves the detection of respiratory viruses.* Emerging Mic. & Inf. 6(1)(2017) 1-7.
14. M. Niedrig, P. Patel, A. A. El Wahed, R. Schädler, S. Yactayo, *Find the right sample: A study on the versatility of saliva and urine samples for the diagnosis of emerging viruses*. BMC infectious diseases. 18 (2018) 1-4.
15. D. Paul, P. Kolar, S. G. Hall, *A review of the impact of environmental factors on the fate and transport of coronaviruses in aqueous environments*. NPJ Clean Water. 4(1)(2021) 7.
16. S. A. Lowry, M. K. Wolfe, A.B. Boehm, *Respiratory virus concentrations in human excretions that contribute to wastewater: a systematic review and meta-analysis*. J. of Water and Health. 21(6)(2023) 831-848.
17. A. S. van Doorn, B. Meijer, C. M. Frampton, M. L. Barclay, N. K. de Boer, *Systematic review with meta-analysis: SARS-CoV-2 stool testing and the potential for faecal-oral transmission.* Alimentary pharmacology & therapeutics. 52(8)(2020) 1276-1288.
18. D. L. Jones, M. Q. Baluja, D. W. Graham, A. Corbishley, J. E. McDonald, S. K. Malham, L. S. Hillary, T. R. Connor, W. H. Gaze, I. B. Moura, M. H. Wilcox, *Shedding of SARS-CoV-2 in feces and urine and its potential role in person-to-person transmission and the environment-based spread of COVID-19*. Sci. of the Tot. Env. 749(2020) 141364.
19. R. C. Picão, J. P. Cardoso, E. H. Campana, A. G. Nicoletti, F. V. Petrolini, D. M. Assis, L. Juliano, A. C. Gales, *The route of antimicrobial resistance from the hospital effluent to the environment: focus on the occurrence of KPC-producing Aeromonas spp*. and Enterobacteriaceae in sewage. Diagn. Microbiol. Infect. Dis. 76(1)(2013) 80–85. https://doi.org/10.1016/j.diagmicrobio.2013.02.001.
20. 20 J. Li, J. Liu, H. Yu, W. Zhao, X. Xia, S. You, et al., *Sources, fates and treatment strategies of typical viruses in urban sewage collection/treatment systems: A review.* Desalination. 534(2022) 115798. https://doi.org/10.1016/j.desal.2022.115798.
21. 21 A. Carratalà, M. Rusinol, A. Hundesa, M. Biarnes, J. Rodriguez-Manzano, A. Vantarakis, et al., *A novel tool for specific detection and quantification of chicken/turkey parvoviruses to trace poultry fecal contamination in the environment.* Appl. Environ. Microbiol. 78(20) (2012) 7496–7499. https://doi.org/10.1128/AEM.01283-12.
22. 22 Environmental Protection Agency (EPA), Polluted Runoff: Nonpoint Source (NPS) Pollution. https://www.epa.gov/nps, 2025 (accessed 8 May 2025).
23. 23 K.E. Graham, C.E. Anderson, A.B. Boehm, *Viral pathogens in urban stormwater runoff: Occurrence and removal via vegetated biochar-amended biofilters.* Water Res. 207(2021) 117829. https://doi.org/10.1016/j.watres.2021.117829.
24. 24 B.H. Ali, M.S. Shahin, M.M. Sangani, M. Faghihinezhad, M. Baghdadi, *Wastewater aerosols produced during flushing toilets, WWTPs, and irrigation with reclaimed municipal wastewater as indirect exposure to SARS-CoV-2.* J. Environ. Chem. Eng. 9(5)(2021) 106201. https://doi.org/10.1016/j.jece.2021.106201.
25. M. Lou, S. Liu, C. Gu, H. Hu, Z. Tang, Y. Zhang, et al., *The bioaerosols emitted from toilet and wastewater treatment plant: a literature review.* Environ. Sci. Pollut. Res. 28(2021) 2509–2521.
26. K.R. Daellenbach, G. Uzu, J. Jiang, L.E. Cassagnes, Z. Leni, A. Vlachou, et al., *Sources of particulate-*

matter air pollution and its oxidative potential in Europe. Nature 587(2020) 414–419.
27. V. Senatore, T. Zarra, A. Buonerba, K.H. Choo, S.W. Hasan, G. Korshin, et al., *Indoor versus outdoor transmission of SARS-CoV-2: environmental factors in virus spread and underestimated sources of risk.* Euro-Mediterr. J. Environ. Integr. 6(2021) 1–9.
28. Z. Gu, J. Han, L. Zhang, H. Wang, X. Luo, X. Meng, et al., *Unanswered questions on the airborne transmission of COVID-19.* Environ. Chem. Lett. 21(2)(2023) 725–739.
29. K. Ram, R.C. Thakur, D.K. Singh, K. Kawamura, A. Shimouchi, Y. Sekine, et al., *Why airborne transmission hasn't been conclusive in case of COVID-19? An atmospheric science perspective.* Sci. Total Environ. 773(2021) 145525.
30. A. Adam, F.M. El-Hussainy, A.A. Ali, M.M. Eid, *Identification chemical air pollutant sources over Egypt on 10 May 2010.* Al-Azhar Bull. Sci. 24(2)(2013) Article 35. https://doi.org/10.21608/absb.2013.6589
31. P. Khare, L.C. Marr, *Simulation of vertical concentration gradient of influenza viruses in dust resuspended by walking. Indoor Air* 25(4)(2015) 428–440. https://doi.org/10.1111/ina.12156
32. W. G. Lindsley, F. M. Blachere, K. A. Davis, T. A. Pearce, M. A. Fisher, R. Khakoo, et al., *Distribution of airborne influenza virus and respiratory syncytial virus in an urgent care medical clinic.* Clin. Infect. Dis. 50(5)(2010) 693–698. https://doi.org/10.1086/650457
33. J. Appert, P. C. Raynor, M. Abin, Y. Chander, H. Guarino, S. M. Goyal, et al., *Influence of suspending liquid, impactor type, and substrate on size-selective sampling of MS2 and adenovirus aerosols*. Aerosol Sci. Technol. 46(3)(2012) 249–257. https://doi.org/10.1080/02786826.2011.619224.
34. D. A. Tyrrell, *The spread of viruses of the respiratory tract by the airborne route*. Symp. Soc. Gen. Microbiol. 17 (1967) 286–306.
35. C. Alonso, P.C. Raynor, P. R. Davies, M. Torremorell, *Concentration, size distribution, and infectivity of airborne particles carrying swine viruses*. PLoS One. 10(8)(2015) e0135675. https://doi.org/10.1371/journal.pone.0135675
36. K. Lin, C. R. Schulte, L. C. Marr, *Survival of MS2 and Φ6 viruses in droplets as a function of relative humidity, pH, and salt, protein, and surfactant concentrations*. PLoS One. 15(12)(2020) e0243505. https://doi.org/10.1371/journal.pone.0243505
37. W. Yang, S. Elankumaran, L. C. Marr, *Relationship between humidity and influenza A viability in droplets and implications for influenza's seasonality*. PLoS One. (2012). https://doi.org/10.1371/journal.pone.0046789.
38. P. Vasickova, I. Pavlik, M. A. Verani, A. N. Carducci, *Issues concerning survival of viruses on surfaces.* Food Environ. Virol. 2(2010) 24–34.
39. C. Peng, L. Chen, M. Tang, *A database for deliquescence and efflorescence relative humidities of compounds with atmospheric relevance*. Fundamental Res. 2(4)(2022) 578-87.
40. J. L. Cannon, E. Papafragkou, G. W. Park, J. Osborne, L.A. Jaykus, J. Vinjé, *Survival of norovirus on surfaces under desiccated conditions.* Appl. Environ. Microbiol. 72(12)(2006) 7894–7896.
41. A. C. Lowen, S. Mubareka, J. Steel, P. Palese, *Influenza virus transmission is dependent on relative humidity and temperature. PLoS Pathog.* 3(10)(2007) e151. https://doi.org/10.1371/journal.ppat.0030151
42. E. Robilotti, S. Deresinski, B. A. Pinsky, *Norovirus*. Clin. Microbiol. Rev. 28(1)(2015) 134–164. https://doi.org/10.1128/cmr.00075-14
43. S. A. Ansari, V. S. Springthorpe, S. A. Sattar, S. Rivard, M. Rahman, *Potential role of hands in the spread of respiratory viral infections: studies with human parainfluenza virus 3 and rhinovirus 14*. J. Clin. Microbiol. 29(10)(1991) 2115–2119. https://doi.org/10.1128/jcm.29.10.2115-2119.1991
44. S. Jia, X. Zhang, Biological HRPs in wastewater. High-Risk Pol. in WW. (2020) 41–78. https://doi.org/10.1016/B978-0-12-816448-8.00003-4.
45. Y. Ye, P. H. Chang, J. Hartert, K. R. Wigginton, *Reactivity of enveloped virus genome, proteins, and lipids with free chlorine and UV254.* Environ. Sci. Technol. 52(14)(2018) 7698–7708. https://doi.org/10.1021/acs.est.8b00824.
46. M. Patel, A. K. Chaubey, C. U. Pittman Jr., T. Mlsna, D. Mohan, *Coronavirus (SARS-CoV-2) in the environment: occurrence, persistence, analysis in aquatic systems and possible management*. Sci. Total Environ. 765(2021) 142698. https://doi.org/10.1016/j.scitotenv.2020.142698.
47. J. Wang, K. Tang, K. Feng, X. Lin, W. Lv, K. Chen, et al., *Impact of temperature and relative humidity on the transmission of COVID-19: a modelling study in China and the United States.* BMJ Open 11(2)(2021) 43863. https://doi: 10.1136/bmjopen-2020-043863.
48. J. Tian, C. Yan, S.G. Alcega, F. Hassard, S. Tyrrel, F. Coulon, Z. A. Nasir, *Detection and characterization of bioaerosol emissions from wastewater treatment plants: Challenges and opportunities*. Front. Microbiol. 13(2022) 958514. https://doi.org/10.3389/fmicb.2022.958514.
49. Y. Han, K. Yang, T. Yang, M. Zhang, L. Li, *Bioaerosols emission and exposure risk of a wastewater treatment plant with A2O treatment process.* Ecotoxicol. Environ. *Saf.* 169(2019) 161–168. https://doi.org/10.1016/j.ecoenv.2018.11.018.
50. K. Yang, L. Li, Y. Wang, S. Xue, Y. Han, J. Liu, *Airborne bacteria in a wastewater treatment plant: emission characterization, source analysis and health risk assessment*. Water Res. 149(2019) 596–606. https://doi.org/10.1016/j.watres.2018.11.027.

51. L. Fracchia, S. Pietronave, M. Rinaldi, M. G. Martinotti, *Site-related airborne biological hazard and seasonal variations in two wastewater treatment plants*. Water Res. 40(10)(2006) 1985–1994. https://doi.org/10.1016/j.watres.2006.03.016.
52. L. Pascual, S. Pérez-Luz, M. A. Yáñez, A. Santamaría, K. Gibert, M. Salgot, et al., *Bioaerosol emission from wastewater treatment plants*. Aerobiologia 19(2003) 261–270.
53. M. Michałkiewicz, A. Pruss, Z. Dymaczewski, J. Jeż-Walkowiak, S. Kwaśna, *Microbiological air monitoring around municipal wastewater treatment plants.* Pol. J. Environ. Stud. 20(5) (2011).
54. M. Kitajima, W. Ahmed, K. Bibby, A. Carducci, C.P. Gerba, K.A. Hamilton, et al., *SARS-CoV-2 in wastewater: State of the knowledge and research needs.* Sci. Total Environ. 739(2020) 139076. https://doi.org/10.1016/j.scitotenv.2020.139076.
55. M. Moriyama, W. J. Hugentobler, A. Iwasaki, *Seasonality of respiratory viral infections*. Annu. Rev. Virol. 7(1)(2020) 83–101. https://doi.org/10.1146/annurev-virology-012420-022445.
56. M. Vittecoq, M. Ottmann, F. Renaud, F. Thomas, M. Gauthier-Clerc, *Persistence of influenza A viruses in relation with environmental factors*. Virologie. 15(6)(2011) 371-379. https://doi.org/10.1684/vir.2011.0425.
57. M. Michałkiewicz, *Wastewater Treatment Plants as a Source of Bioaerosols*, Pol. J. Environ. Stud. 28(4) (2019) 2261–2271. https://doi.org/10.15244/pjoes/90183.
58. E. Brisebois, M. Veillette, V. Dion-Dupont, J. Lavoie, J. Corbeil, A. Culley, C. Duchaine, *Human viral pathogens are pervasive in wastewater treatment center aerosols.* J. of Environ. Sci. 67(2018) 45-53.
59. M. Itarte, M. Calvo, L. Martínez-Frago, C. Mejías-Molina, S. Martínez-Puchol, R. Girones, G. Medema, S. Bofill-Mas, M. Rusiñol, *Assessing environmental exposure to viruses in wastewater treatment plant and swine farm scenarios with next-generation sequencing and occupational risk approaches*. Int. J. Hyg. Environ. Health 259(2024) 114360. https://doi.org/10.1016/j.ijheh.2024.114360.
60. P. Stellacci, L. Liberti, M. Notarnicola, C. N. Haas, *Hygienic sustainability of site location of wastewater treatment plants: A case study. I. Estimating odour emission impact*. Desalination. 253(1-3)(2010) 51-56. https://doi.org/10.1016/j.desal.2009.11.034.
61. H. F. Hung, Y. M. Kuo, C. C. Chien, C. C. Chen, *Use of floating balls for reducing bacterial aerosol emissions from aeration in wastewater treatment processes.* J Hazard Mater. 175(1-3)(2010) 866-871. https://doi.org/10.1016/j.jhazmat.2009.10.090.
62. F. G. Masclaux, P. Hotz, D. Gashi, D. Savova-Bianchi, A. Oppliger, *Assessment of airborne contamination in wastewater treatment plants*. Environ Res. 133(2014) 260-265. https://doi.org/10.1016/j.envres.2014.06.002.
63. Y. H. Chen, C. Yan, Y. F. Yang, J. X. Ma, *Quantitative microbial risk assessment and sensitivity analysis for workers exposed to pathogenic bacterial bioaerosols under various aeration modes in two wastewater treatment plants*. Sci Total Environ. 755(2021) 142615. https://doi.org/10.1016/j.scitotenv.2020.142615.
64. P. Grisoli, M. Rodolfi, S. Villani, E. Grignani, D. Cottica, A. Berri, A. M. Picco, C. Dacarro, *Assessment of airborne microorganism contamination in an industrial area characterized by an open composting facility and a wastewater treatment plant.* Environ Res. 109(2)(2009) 135-142. https://doi.org/10.1016/j.envres.2008.11.001
65. V. Doceul, E. Bagdassarian, A. Demange, N. Pavio, *Zoonotic hepatitis E virus: classification, animal reservoirs and transmission routes*. Viruses. 8(10)(2016) 270. https://doi.org/10.3390/v8100270.
66. B. Kumar, A. Manuja, B. R. Gulati, N. Virmani, B. N. Tripathi, *Zoonotic viral diseases of equines and their impact on human and animal health.* Open Virol J. 12 2018) 80. doi: 10.2174/1874357901812010080.
67. B. Lam, L. Henry, Z. Younossi, *Sofosbuvir (Sovaldi) for the treatment of hepatitis C.* Expert review of clinic. pharm. 7(5)(2014) 555-566. https://doi.org/10.1586/17512433.2014.928196
68. G. B. Elion, *Mechanism of action and selectivity of acyclovir*. The American J. of medicine. 73(1)(1982) 7-13.https://doi.org/10.1016/0002-9343(82)90055-9
69. M. A. Mostafa, *Role of Zidovudine and Candesartan combination in novel SARS-CoV-2 management, prophylaxis and treatment trials*. Al-Azhar Univ J Virus Res Stud. 2(1)(2020) 1-6. https://doi.org/10.21608/AUJV.2020.106719
70. A. M. M. Youssef, Z. A. S. EL-Swaify, D. A. M. Maaty, M. M.Youssef, *Phytochemistry and antiviral Properties of two Lotus species growing in Egypt.* Vitae [online]. 28(3)(2022). https://doi.org/10.17533/udea.vitae.v28n3a348069.
71. F. Abo-Elghiet, A. Rushdi, M. H. Ibrahim, S. H. Mahmoud, M. A. Rabeh, S. A. Alshehri, N.G. El Menofy, *Chemical profile, antibacterial, antibiofilm, and antiviral activities of Pulicaria crispa most potent fraction: an in vitro and in silico study*. Molecules. 28(10)(2023) 4184. https://doi.org/10.3390/molecules28104184
72. H. S. Mohammed, E. F. S. Taha, F. S. Mahrous, R. Sabour, M. M. *Abdel-Aziz, L. D. Ismail, Antimicrobial and antiviral evaluation of compounds from Holoptelea integrifolia: in silico supported in vitro study*. RSC Adv. 13(46)(2023) 32473-32486. https://doi.org/10.1039/d3ra05978b
73. L. D. Nghiem, B. Morgan, E. Donner, M. D. Short, *The COVID-19 pandemic: considerations for the*

waste and wastewater services sector. Case Stud Chem Environ Eng. 1(2020) 100006. https://doi.org/10.1016/j.cscee.2020.100006.

74. L. Warren, S. Davis, C. T. Cyr, *Emergency response planning for water and/or wastewater systems.* Wiley Handb Sci Technol Homel Secur. 15(2008) 1-24. https://doi.org/10.1002/9780470087923.hhs205
75. R. N. Zaneti, V. Girardi, F. R. Spilki, K. Mena, A. P. Westphalen, E. R. da Costa Colares, A. G. Pozzebon, R. G. Etchepare, *Quantitative microbial risk assessment of SARS-CoV-2 for workers in wastewater treatment plants.* Sci Total Environ. 754 (2021) 142163. https://doi.org/10.1016/j.scitotenv.2020.142163
76. D. Zhang, H. Ling, X. Huang, J. Li, W. Li, C. Yi, T. Zhang, Y. Jiang, Y. He, S. Deng, X. Zhang, *Potential spreading risks and disinfection challenges of medical wastewater by the presence of Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) viral RNA in septic tanks of Fangcang Hospital.* Sci Total Environ. 741(2020) 140445. https://doi.org/10.1016/j.scitotenv.2020.140445
77. V. Senatore, T. Zarra, G. Oliva, V. Belgiorno, V. Naddeo, *Volatile organic compounds (VOCs) control by combining bio-scrubber and ozone pretreatment*, Glob. Nest J. (2020) 1–6.
78. A. Mohamadi Nasrabadi, D. Eckstein, P. Mettke, N. Ghanem, R. Kallies, M. Schmidt, F. Mothes, T. Schaefer, R. Graefe, C. D. Bandara, M. Maier, *A Virus Aerosol Chamber Study: The Impact of UVA, UVC, and H2O2 on Airborne Viral Transmission.* Envir. & Health. (2025) 7.
79. J. L. Sagripanti, C. D. Lytle, *Estimated inactivation of coronaviruses by solar radiation with special reference to COVID-19. Photochem. Photobiol*. 96(4) (2020) 731–737. https://doi.org/10.1111/php.13293.
80. H. Mbareche, E. Brisebois, M. Veillette, C. Duchaine, *Bioaerosol sampling and detection methods based on molecular approaches: No pain no gain*. Sci Total Environ. 599(2017) 2095-2104. https://doi.org/10.1016/j.scitotenv.2017.05.076
81. C. W. Haig, W. G. Mackay, J. T. Walker, C. Williams, *Bioaerosol sampling: sampling mechanisms, bioefficiency and field studies*. J Hosp Infect. 93(3)(2016) 242-255. doi: 10.1016/j.jhin.2016.03.017
82. H. T. Bausum, S. A. Schaub, K. F. Kenyon, M. J. Small, *Comparison of coliphage and bacterial aerosols at a wastewater spray irrigation site*. Appl Environ Microbiol. 43(1)(1982) 28-38. https://doi.org/10.1128/aem.43.1.28-38.1982
83. K. F. Fannin, J. J. Gannon, K. W. Cochran, J. C. Spendlove, *Field studies on coliphages and coliforms as indicators of airborne animal viral contamination from wastewater treatment facilities*. Water Res. 11(2)(1977):181–188. https://doi.org/10.1016/0043-1354(77)90124-5
84. K. F. Fannin, S. C. Vana, W. Jakubowski, *Effect of an activated sludge wastewater treatment plant on ambient air densities of aerosols containing bacteria and viruses*. Appl. Environ. Microbiol. 49(5)(1985) 1191–1196. https://doi.org/10.1128/aem.49.5.1191-1196.1985.
85. K. P. Brenner, P. V. Scarpino, C. S. Clark, *Animal viruses, coliphages, and bacteria in aerosols and wastewater at a spray irrigation site*. Appl. Environ. Microbiol. 54(2)(1988) 409–415. https://doi.org/10.1128/aem.54.2.409-415.1988
86. A. Carducci, S. Arrighi, A. Ruschi, *Detection of coliphages and enteroviruses in sewage and aerosol from an activated sludge wastewater treatment plant*. Lett. Appl. Microbiol. 21(3)(1995) 207–209. https://doi.org/10.1111/j.1472-765X.1995.tb01042.x.
87. A. Carducci, E. Tozzi, E. Rubulotta, B. Casini, L. Cantiani, E. Rovini, M. Muscillo, R. Pacini, *Assessing airborne biological hazard from urban wastewater treatment*. Water Res. 34(4)(2000) 1173–1178. https://doi.org/10.1016/S0043-1354(99)00264-X.
88. H. Heinonen-Tanski, T. Reponen, J. Koivunen, *Airborne enteric coliphages and bacteria in sewage treatment plants*. Water Res. 43(9)(2009):2558–2566. https://doi.org/10.1016/j.watres.2009.02.025.
89. K. Uhrbrand, A. C. Schultz, A. M. Madsen, *Exposure to airborne noroviruses and other bioaerosol components at a wastewater treatment plant in Denmark*. Food Environ. Virol. 3(3)(2011) 130–137.
90. D. Courault, I. Albert, S. Perelle, A. Fraisse, P. Renault, A. Salemkour, P. Amato, *Assessment and risk modeling of airborne enteric viruses emitted from wastewater reused for irrigation*. Sci. Total Environ. 592(2017) 512–526. https://doi.org/10.1016/j.scitotenv.2017.03.105.
91. K. Uhrbrand, A. C. Schultz, A. J. Koivisto, U. Nielsen, A. M. Madsen, *Assessment of airborne bacteria and noroviruses in air emission from a new highly-advanced hospital wastewater treatment plant*. Water Res. 112(2017) 110–119. https://doi.org/10.1016/j.watres.2017.01.046
92. K. Lin, L. C. Marr, *Aerosolization of Ebola virus surrogates in wastewater systems*. Environ. Sci. Technol. 51(5)(2017) 2669–2675. https://doi.org/10.1021/acs.est.6b04846.
93. E. Brisebois, M. Veillette, V. Dion-Dupont, J. Lavoie, J. Corbeil, A. Culley, C. Duchaine, *Human viral pathogens are pervasive in wastewater treatment center aerosols*. J. Environ. Sci. 67(2018) 45–53. https://doi.org/10.1016/j.jes.2017.07.015
94. K. Matsubara, H. Katayama, *Development of a portable detection method for enteric viruses from ambient air and its application to a wastewater treatment plant*. Pathogens 8(3)(2019) 131. https://doi.org/10.3390/pathogens8030131.
95. H. Pasalari, A. Ataei-Pirkooh, M. Aminikhah, A. J. Jafari, M. Farzadkia, *Assessment of airborne enteric viruses emitted from wastewater treatment plant: atmospheric dispersion model, quantitative microbial risk assessment, disease burden*. Environ. Pollut.

253(2019) 464–473. https://doi.org/10.1016/j.envpol.2019.07.010.

96. O. Ginn, L. Rocha-Melogno, A. Bivins, S. Lowry, M. Cardelino, D. Nichols, S.N. Tripathi, F. Soria, M. Andrade, M. Bergin, M.A. Deshusses, *Detection and quantification of enteric pathogens in aerosols near open wastewater canals in cities with poor sanitation*. Environ. Sci. Technol. 55(21)(2021) 14758–14771. https://doi.org/10.1021/acs.est.1c05060.
97. S. Gholipour, F. Mohammadi, M. Nikaeen, Z. Shamsizadeh, A. Khazeni, Z. Sahbaei, S. M. Mousavi, M. Ghobadian, H. Mirhendi, *COVID-19 infection risk from exposure to aerosols of wastewater treatment plants*. Chemosphere 273(2021) 129701. https://doi.org/10.1016/j.chemosphere.2021.129701.
98. A. Stobnicka-Kupiec, M. Gołofit-Szymczak, M. Cyprowski, R.L. Górny, *Detection and identification of potentially infectious gastrointestinal and respiratory viruses at workplaces of wastewater treatment plants with viability qPCR/RT-qPCR*. Sci. Rep. 12(1)(2022) 4517.
99. L. J. Justen, S. L. Grimm, K. M. Esvelt, W. J. Bradshaw, *Indoor air sampling for detection of viral nucleic acids*. J. Aerosol Sci. (2024) 106549. https://doi.org/10.1016/j.jaerosci.2025.106549.
100. M. Pan, J. A. Lednicky, C.Y. Wu, *Collection, particle sizing and detection of airborne viruses*. J. Appl. Microbiol. 127(6)(2019) 1596–1611. https://doi.org/10.1111/jam.14278.
101. Y. Kott, H. B. Ari, N. Buras, *The fate of viruses in a marine environment*. Adv. Water Pollut. Res. (1969) 823–829.
102. F. Baggi, A. Demarta, R. Peduzzi, *Persistence of viral pathogens and bacteriophages during sewage treatment: lack of correlation with indicator bacteria*. Res. Microbiol. 152(8)(2001) 743–751. https://doi.org/10.1016/S0923-2508(01)01255-4.
103. N. Montazeri, D. Goettert, E. C. Achberger, C. N. Johnson, W. Prinyawiwatkul, M.E. Janes, *Pathogenic enteric viruses and microbial indicators during secondary treatment of municipal wastewater*. Appl. Environ. Microbiol. 81(18)(2015) 6436–6445. https://doi.org/10.1128/AEM.01218-15
104. T.G. Metcalf, J.M. Vaughn, W.C. Stiles, The occurrence of human viruses and coliphage in marine waters and shellfish, FAO Technical Conference on Marine Pollution and its Effects on Living Resources and Fishing, FAO United Nations, Rome, Italy, 1970, pp. 570–574.
105. Y. Kott, N. Roze, S. Sperber, N. Betzer, *Bacteriophages as viral pollution indicators*. Water Res. 8(3)(1974) 165–171. https://doi.org/10.1016/0043-1354(74)90039-6.
106. E. Rames, A. Roiko, H. Stratton, J. Macdonald, *Technical aspects of using human adenovirus as a viral water quality indicator*. Water Res. 96(2016) 308–326. https://doi.org/10.1016/j.watres.2016.03.042.
107. M. K. Rashed, W. M. El-Senousy, E. T. Sayed, M. AlKhazindar, *Infectious pepper mild mottle virus and human adenoviruses as viral indices in sewage and water samples*. Food Environ. Virol. 14(3)(2022) 246–257.