Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

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SARS-CoV-2 and other viruses in soil: An environmental outlook

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ABSTRACT

In the present review, the authors shed light on the SARS-CoV-2 impact, persistence, and monitoring in the soil environment. With this purpose, several aspects have been deepened: i) viruses in soil ecosystems; ii) direct and indirect impact on the soil before and after the pandemic, and iii) methods for quantification of viruses and SARS-CoV-2 monitoring in soil. Viruses are present in soil (i.e. up to 417×10^7 viruses per g TS^{-1} in wetlands) and can affect the behavior and ecology of other life forms (e.g. bacteria), which are remarkably important for maintaining environmental equilibrium. Also, SARS-CoV-2 can be found in soil (i.e. up to 550 copies·g^{-1}). Considering that the SARS-CoV-2 is very recent, poor knowledge is available in the literature on persistence in the soil and reference has been made to coronaviruses and other families of viruses. For instance, the survival of enveloped viruses (e.g. SARS-CoV) can reach 90 days in soils with 10% of moisture content at ambient. In such a context, the possible spread of the SARS-CoV-2 in the soil was evaluated by analyzing the possible contamination routes.

1. Introduction

The recent COVID-19 pandemic caused by SARS-CoV-2 has demonstrated how viruses or pathogenic microorganisms can generate epidemic diseases and subsequently cause socio-economic and environmental damage (Atar and Atar, 2020; Mofijur et al., 2021; Razzaq et al., 2020). SARS-CoV-2 is a viral strain of the coronaviruses (CoVs), which are named after their shape, as the spike proteins present on their surface resemble a crown, or corona in Latin (Acter et al., 2020). CoVs are positive single-stranded RNA viruses characterized by small size (i.e. 60–220 nm in diameter) and detected in many animals, and therefore, a CoV can reach humans causing potential epidemic outbreaks, as previously occurred with MERS-CoV and SARS-CoV (Lee and Hsueh, 2020; Prompetchara et al., 2020; Su et al., 2016).

To date (April 30th, 2021), SARS-CoV-2 has been detected in almost all countries of the world (i.e. 223) by infecting more than 150.1 million people and causing almost 3.2 million deaths (WHO). These data underscore COVID-19 rapid and vast spread, due to its high reproduction number (R_0) and long incubation period, factors that led to an easy and wide transmission of the infection (Cao et al., 2020; Liu et al., 2020). Also, the spread of SARS-CoV-2 variants, i.e. B.1.1.7, 501Y.V2, P.1 and B.1.617 by the United Kingdom, South Africa, Brazil and India, respectively, raises worries due to their alleged facility of transmission and wide mutations in the spike protein (Abdool Karim and de Oliveira, 2021; Moelling, 2021; Wang et al., 2021).

The main ways of virus spreads are aerosols and droplets, but it is well known that transmission can also occur via the fecal-oral route (Chen et al., 2020; Ding and Liang, 2020). Therefore, all the environmental compartments (i.e. water, air and soil) on which the virus can act should be necessarily monitored in order to control the spread of the infection (Cela-Dablanca et al., 2021; Núñez-Delgado, 2020a). However, the interactions between viruses and soil have received less attention compared to the other compartments, thus neglecting a possible contagion route. On the contrary, the relationship between water and wastewater, and the virus spread through the air particles, have been extensively studied (Anand et al., 2021b). Indeed, works published up to April 30th, 2021 and available on the Scopus database accounted for 1, 524, 692 and just 36 for air, water and soil compartments, respectively

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(the keywords used were "SARS-CoV-2" and "air", "SARS-CoV-2" and "water", and "SARS-CoV-2" and "soil", respectively).

Specifically, the possible interactions that can occur between wastewater and soil have received little investigation, but the interplay between these two matrices is continuous and frequent (Núñez-Delgado, 2020a). It is worth recalling that wastewater can be employed for secondary uses such as field irrigation, as well as the sewage sludge derived from wastewater treatment plants can be used as fertilizer in agricultural activities (Lamastra et al., 2018; Martínez-Puchol et al., 2020). This type of activity could allow the migration of the virus from wastewater and/or sewage sludge to the ground due to the fact that SARS-CoV-2 was found and proven to be present in these mentioned matrices (Balboa et al., 2020; Núñez-Delgado, 2020b). In addition, a source of interaction between the soil and SARS-CoV-2 can be due to the incorrect disposal of urban and hospital waste (Iyer et al., 2021), such as the protective devices used for the prevention of the virus (e.g. gloves and masks) (Rahman et al., 2020; Zand and Heir, 2020a).

Hence, this review paper is aimed at illustrating the possible impacts and consequences that the COVID-19 pandemic could have on the soil ecosystem. The manuscript is aimed at analyzing: i) the soil ecosystem and its health status before and after the pandemic; ii) the persistence of SARS-CoV-2 and other viruses in the soil; iii) the methods for SARS-CoV-2 detection and quantification in soil and iv) the monitoring and management of soil impacted by viruses.

2. Viruses in soil

During the COVID-19 pandemic, different basic essential parameters (e.g. national economics, public health) are assessed (Hsiang et al., 2020; Rundle et al., 2020; Siche, 2020). However, among all the environmental concerns of the current situation, soil health can be predominant for living organisms. Indeed, soil plays an important role in the decomposition degree of organic compounds, maintaining the biogeochemical cycle (Nannipieri et al., 2017). Also, the soil is one of the greatest reservoirs of microorganisms (Breitbart and Rohwer, 2005; Rohwer et al., 2009), which are responsible for a series of environmental chemistry reactions (Douglas, 2006). However, viruses (e.g. bacteriophages) can affect the bacterial population, causing detrimental effects on soil quality (Srinivasiah et al., 2013) (see section 2.2).

Viruses found in the soil environment can have an impact on economy and production likely due to an infectious cycle that helps the gene transformation process (Breitbart and Rohwer, 2005; Jain, 2003; Jones et al., 2007). In addition, specific viruses such as SARS-CoV-2 can be transmitted to the soil through infected water bodies, by means of sewage sludge and irrigation water, deeply affecting soil health, and with potential to eventually suffer adsorption-desorption phenomena in function of various parameters (e.g. pH, temperature, surface charges, moisture content) (see section 2.3).

2.1. Soil structure

The soil is not a unique and indistinct system, but is referred to individual ecosystems offering several habitats (e.g. rhizosphere, drilosphere), which jointly describe the whole and porous structure of this tridimensional entity that covers a high proportion of the world (George et al., 2019; Reich, 2014). The soil can be defined as a heterogeneous structure composed of different phases (i.e. solid, liquid and gaseous), whose aliquots and specific elements can be remarkably varied along with space and over time (Rabot et al., 2018). The content of organic matter (OM), i.e. mostly a mixture of partially or totally decomposed plants and animals, also characterizes the soil composition.

In such context, and regarding microbiota, the complexity of the soil ecosystems is meaningful to be understood, due to the fact that all the interactions between the host and virus or pathogens can occur in the soil through contact with host cells (Emerson, 2019; Munson-Mcgee et al., 2018), which leads to virus replication and progeny release (Iwanami et al., 2020). In addition, the physical-chemical demand of soil ecosystems considerably affected the virus-host interactions in the soil, allowing virus mutation due to the host resistance phenomena over the years (Sime-Ngando, 2014).

Several extraction methods (e.g. dispersed soil, aqueous two-phase partitioning) evaluating the bacterial number expressed as average per unit of inorganic mass are reported in studies on soil microbiology to cope with soil microheterogeneity, returning results fluctuating with various orders of magnitude (i.e. from 1×10^8 to 1×10^{10}) (Insam, 2001). In addition, recent analytical methods focusing on the soil virome (i.e. the assembly of viruses) led to examine the virus role (e.g. coronaviruses) in the soil microbial communities (Gundy et al., 2009; Srinivasiah et al., 2008). Notwithstanding, the majority of studies are limited to the topsoil (i.e. up to 10 cm), and therefore, future attention should be addressed to increase the knowledge of viruses in deeper layers of soils within the overall biosphere (Williamson et al., 2017). Indeed, previous studies showed that viruses can percolate into soil reaching depths well below 10 m (De Serres et al., 1999; Keswick et al., 1984).

2.2. Viral ecology

A virus is not able to absorb and save energy, or even enabled out of their host target. Hence, a virus is not considered as an independent living organism in biology but as an infectious agent encompassed by a protein capsid, and is distinguished by internal (e.g. diameter, genetic material) and relational (e.g. host, other objects) characteristics (Regenmortel, 2000).

2.2.1. Bacteriophages

Bacteriophages (phages) are the main viruses in the aquatic ecosystems, and this statement should not be easily extended to soil environments due to the scarcity of bacterial biomasses in the bulk soils, which instead are abundant in the rhizosphere (Buée et al., 2009). Bacteriophages are classified into 21 morphotypes, being part of 13 virus families and almost 140 bacterial genera (Kimura et al., 2008). Phages also co-exist with bacteria by showing a predator-prey relationship in a sort of "arms race" explained by the theory of "coevolution", as proposed by Alexander (1981). However, the coevolution can be affected by lytic and lysogenic phenomena.

Lytic phages quickly start a prolific cycle, where the phage genome is replicated and descendant phages are released via bacterial lysis (Feiner et al., 2015). Moderate phages start a lysogenic cycle when the phage genome is merged into the bacterial chromosome as prophages, which are concomitantly replicated with the bacterial host chromosome during its reproduction, and can enter a lytic cycle (Miller and Day, 2009). Pseudolysogeny is a precarious state where the phage genome is not able to start a lytic or lysogenic, mostly occurring under limited nutrient conditions (Feiner et al., 2015). Hence, lysogeny can be seen as the persistence approach by viruses at poor host abundance and activity, as also occurs in bulk soils (Trubl et al., 2018).

2.2.2. Virus abundance in soil

Viruses are notably abundant in various soils and are characterized by exhibiting more limited variability compared to host bacteria in the function of environmental circumstances. Indeed, viruses abound in soils with high OM and moisture content compared to dried and desert soils (Srinivasiah et al., 2008). In cold deserts, virus copiousness was composed of 23–64 $\times 10^7$ viruses per g of soil total solids (TS^{-1}) (Fig. 1), which was lower compared to wetlands and agricultural soils (i.e. 87–417 $\times 10^7$ viruses per g TS^{-1}, Fig. 1) (Williamson et al, 2005, 2007). On the contrary, the bacterial abundance shows a soil population considerably mutable in copiousness, ranging from 0.035 to 338 $\times 10^6$ cells·g mL^{-1} (Fig. 1), which was 10,000-fold greater in wetlands compared to cold deserts (Fig. 1) (Williamson et al, 2005, 2007).

Furthermore, pH and temperature may also play a significant role in

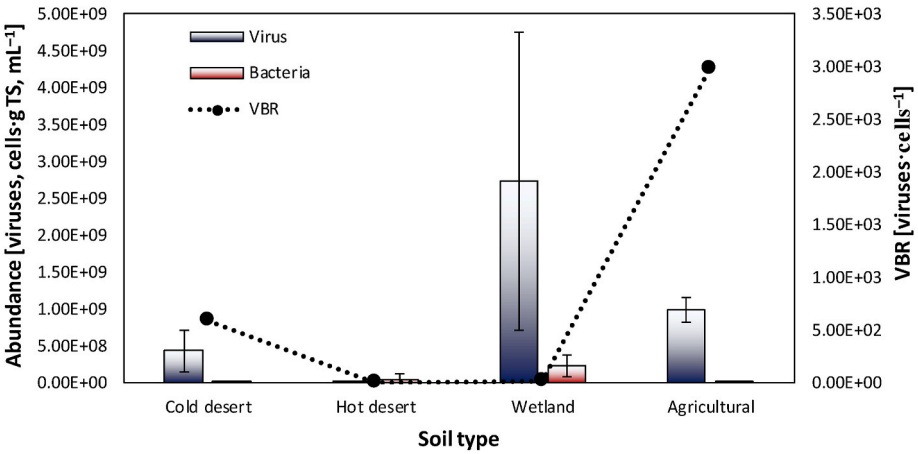

Fig. 1. Viral (viruses·g TS^{-1}) and bacterial (cells·mL^{-1}) abundance in various soil types (i.e. cold and hot deserts, wetlands and agricultural soils). The values are the means and standard deviations of the reported data for each soil. The virus to bacteria ratio (VBR) was calculated assuming a conversion factor of 1 g mL^{-1} for bacterial abundance (Srinivasiah et al., 2008). Data were taken from Williamson et al. (2017, 2007, 2005).

viral and bacterial abundance in soil (Qu et al., 2020). Williamson et al. (2017) reported a reduction of virus copiousness in soil with extreme pH values (Table 1), thus affecting the virus persistence in soil. A substantial difference can be also seen by comparing the viral abundance in cold (Fig. 1) and hot deserts (i.e. 2.2×10^3–1×10^7 viruses per·g TS^{-1}, Fig. 1), with lower temperatures probably promoting the virus persistence (Williamson et al., 2017). This should be seen as an important aspect, being real the possibility of reviving frozen pathogenic microorganisms such as the endospores of Siberian anthrax which can remain in the Permafrost for many years (Steffan et al., 2020).

2.3. Human viruses in soil environment

2.3.1. Enveloped viruses

A CoV refers to an extended group of enveloped viruses (e.g. SARS-CoV-1, MERS-CoV, SARS-CoV-2) with a +ssRNA and crown-like heads on their egg-shaped surfaces (La Rosa et al., 2020) with diameters comprised between 60 and 220 nm (Table 1) (Cascella et al., 2020), which can influence the virus mobility in the soil environment. The genome sequence of SARS-CoV-2 was positively correlated with SARS-CoV-1 (i.e. 82%) (Chan et al., 2020), which was listed with MERS-CoV as a pathogenic agent with high mortality by causing severe pneumonia and respiratory failure in an infected subject (Qu et al., 2020). CoV can be spread via direct contact between humans (i.e. symptomatic or asymptomatic people) through the inhalation of the breathed virus in droplets (e.g. coughs or sneezes) or with infected surfaces (e.g. skin-to-skin, objects) (Qu et al., 2020). However, other sources of contamination such as the environmental compartments (e.g. soil) should be clarified.

Skeletal similarities between SARS-CoV and SARS-CoV-2 would suggest the possible use of already found data for SARS-CoV and its surrogates, in order to suppose the environmental fate of SARS-CoV-2 (Kumar et al., 2020c). The enveloped viruses differ from non-enveloped viruses due to genome, structure, replication, pathogenicity and persistence (Wigginton and Boehm, 2020). This aspect can be significant to determine the interactions (e.g., hydrophobic) that can allow the adsorption of CoV onto solid particles.

The time needed to reduce 90% the initial viral infectivity (T_{90}) was reported to reach almost 40 d for SARS-CoV-2 in wastewater (Table 1) (Mohapatra et al., 2020). In addition, T_{90} of human CoV was comprised between 200 and 400 d at 4 °C in different water matrices (Table 1) (Kumar et al., 2020c), and can reach a value of 588 d in filtered tap water (Table 1), as reported by Franklin and Bevins (2020). The endurance of enveloped viruses such as the influenza virus (H1N1) in the water was determined as almost 200 days at 4 °C (Table 1) (Dublineau et al., 2011). Gutiérrez and Buchy (2012) reported that avian influenza (H5N1) could not maintain the viral load in sandy topsoil, but could remain in soil-based compost, thus indicating that different soil properties considerably influence the survival of the virus.

Although the World Health Organization (WHO) suggested that there was no evidence regarding the persistence of SARS-CoV-2 in wastewater or drinking water (Baldovin et al., 2021; Kitajima et al., 2020), the viral load of enveloped viruses such as CoV could be maintained for a prolonged time in the soil environment in the function of different parameters such as the temperature, moisture content, pH, OM, sunlight radiation, and occurrence of clays and nutrients (see section 2.5) (Anand et al., 2021a). For example, Bivins et al. (2020) recently evaluated that the increase of temperature from ambient to 50 °C can drastically reduce the T_{90} to 15 min (Table 1). Likewise, the survival of enteric viruses such as SARS-CoV can be decreased in dried soils (i.e. 15–25 d) compared to soils with 10% of moisture content (i.e. 60–90 d) at ambient temperature (i.e. 20 °C) (Bosch et al., 2006). In addition, the experiments performed on frozen and thawed samples could affect the microbial community involved, thus contributing to the reduction of the CoV particles in the investigated matrix (Bivins et al., 2020).

2.3.2. Non-enveloped viruses and other pathogenic microorganisms

Adenovirus, enterovirus, and orthoreovirus are non-enveloped viruses that can cause respiratory tract disease (Michen and Graule, 2010). These non-enveloped viruses are characterized by diameter size of 25–100 nm (Table 1) and could have a great potential to infect a large variety of environmental compartments such as the soil, being resistant to hostile conditions and water treatments (Kumar et al., 2020c).

For example, Adenovirus arrives in the soil environment after sewage sludge amendment (Horswell et al., 2010), and are subsequently sorbed by the soil particles through electrostatic interactions due to its isoelectric point (Table 1), as suggested by Wong et al. (2013). Adenovirus showed a T_{90} value comprised between almost 9 and 51 d at 4 °C (Table 1) depending on the biosolid considered (i.e. dairy and swine manure, respectively) (Wei et al., 2009). Also, in this case, an increase of temperature can allow a reduction of T_{90} to approximately 4 d (Table 1) (Wei et al., 2009). For Enterovirus, i.e. a virus of the Enterovirus genome, the survival in soil was estimated at approximately 14 d (Table 1) (Pourcher et al., 2007). Moreover, Orthoreovirus are reported to have a unique impact on mangrove soil (e.g. in river virome) (Jin et al., 2019), with a T_{90} ranging between 4 and 807 d (Table 1) depending on temperature, pH and ammonia nitrogen concentrations (Magri et al., 2015).

In addition, pathogenic soil bacteria or fungi can quickly infiltrate humans via cutaneous wounds, ingestion of infected foods or soils, and

Table 1
Persistence (T_{90}) of different enveloped (i.e. CoV and H1N1) and non-enveloped (i.e. Adenovirus, Enterovirus and Orthorevirus) virus causing respiratory diseases (e.g., pneumonia) reported for various matrices (i.e. water, tap water, biosolid and soil) in the function of the temperature (i.e. ambient, 4, 37 and 50 °C) and pH values. T_{90} is the time (d) needed for 1 $\log_{10}$ unit reduction.

Viruses	Characteristics					T_{90} [d]					References
	Envelope	Genome type	Genome size [bases]	Diameter [nm]	Isoelectric point	4 °C	Ambient	37 °C	50 °C	pH	Matrices	
CoV[a]	Enveloped	+ve sense single strand RNA	30,000	60–220	6.24	28–588	1.6–59	6–9	15 min	1[b]	Wastewater, tap water	(Ahmed et al., 2020b; Bivins et al., 2020; Cascella et al., 2020; Franklin and Bevins, 2020; Kumar et al., 2020c; Mohapatra et al., 2020)
H1N1	Enveloped	Single RNA	13,500	80–120	6.50–7.00	200	–	–	–	–	Water	(Dublineau et al., 2011; Michen and Graule, 2010)
Adenovirus	Non-enveloped	Double strand RNA	26,000–48,000	90–100	4.50	9–51	4.3–214	–	–	14–98[c]	Biosolids (i.e. manure, sludge)	(Magri et al., 2015; Michen and Graule, 2010; Wei et al., 2009)
Enterovirus	Non-enveloped	+ve sense single strand RNA	7200–8500	25–30	3.80–3.90	–	14	–	–	–	Soil	(Michen and Graule, 2010; Pourcher et al., 2007)
Orthoreovirus	Non-enveloped	Double strand RNA	23,500	70–85	4.00–5.50	807	66–151	–	–	14–98[c]	Biosolid (i.e. sludge)	(Magri et al., 2015; Michen and Graule, 2010)

a = SARS-CoV, SARS-CoV-2, MERS-CoV; b = pH of 2–3 and 11–12; c = pH of 9.

inhalation of respiratory droplets (Steffan et al., 2020). *Yersinia pestis* and *Legionella* spp. are the causative agents of pneumonic diseases with a high mortality rate (Baumgardner, 2012; Lynteris, 2017). For example, *Legionella* spp. can persist for months (i.e. 90–300 d) in a potting soil kept at temperatures comprised between −20 and 35 °C. Although bacterial diseases are not listed as highly contagious (Steffan et al., 2020), these aspects should also be considered in future policies to prevent new pandemics.

2.3.3. Fate of CoV in soil and crosstalk between ecosystems

The soil could be a viral sink acting as a secondary source for the spread of SARS-CoV-2 over an extended period (D. Zhang et al., 2020), likely due to inappropriate sanitization actions that can allow soil contamination by viruses (Foladori et al., 2020). In addition, the detection of a not negligible viral load of the human excrements has increased the concerns regarding the potential spread of SARS-CoV-2 through the soil and other environmental compartments within the ecosystems (e.g. plant, animal, ground-water) (Patel et al., 2020). Indeed, Wang et al. (2020) showed a viral load of human feces around $2.6{\cdot}10^4$ copies·mL^{-1}. Afterwards, SARS-CoV-2 can reach wastewater treatment plants (WWTPs) via sewer systems, with various virus dilutions in the function of the degree of affectation of the pandemic (Foladori et al., 2020), population number, and other parameters such as temperature and travel time (Hart and Halden, 2020).

Ahmed et al. (2020) firstly showed the presence of SARS-CoV-2 in untreated wastewater ranging from 1.9 to 12 copies·100 mL^{-1}. Zaneti et al. (2021) recently conducted a quantitative microbial risk assessment of COVID-19 in wastewater obtaining a viral load comprised between 1.03×10^2 and 1.31×10^4 copies mL^{-1}. The disinfection of wastewater is strongly recommended by the WHO in order to avoid any virus discharge in the environment (Collivignarelli et al., 2020). However, Bogler et al. (2020) reported a partial removal of SARS-CoVs in conventional WWTPs, thus questioning the efficiency of some disinfection treatments. Indeed, 20% of soil samples collected nearby to the hospital receiving COVID-19 subjects and wastewater plant in Wuhan, recently resulted positive to SARS-CoV-2 RNA with an abundance comprised between 205 and 550 copies·g^{-1} (Wiktorczyk-Kapischke et al., 2021).

Hence, a substantial screening should be conducted on the wastewater effluents and sewage sludge before their application in soils to prevent the COVID-19 migration to other environmental compartments (Núñez-Delgado, 2020a). The use of infected wastewater and sewage sludge for soil irrigation and fertilization, respectively, could have a significant impact on the virus spread due to the groundwater contamination by SARS-CoV-2, and its potential uptake in crops (Fig. 2) (Usman et al., 2020), thus entering in the human food chain. A plant can use phosphorus incorporated by viruses after infecting and lysing microbial cells (Kuzyakov and Mason-Jones, 2018). This phenomenon can have a more pronounced harmful effect when untreated wastewater is improperly introduced into the environment due to the persistence of SARS-CoV-2 RNA (Ahmed et al., 2020b). Otherwise, the plant viral internalization can be reduced by the colloid fraction of soil (e.g. OM) (Mancuso et al., 2021). For example, Badawy et al. (1990) revealed that an enterovirus such as coxsackie virus can remain viable in crops for about 3–5 weeks.

Evapotranspiration phenomena can also accelerate the shift of the virus from soil or plants to the air moisture affecting the disease transmission (Lian et al., 2020). However, the viral particles could be strongly sorbed by soil due to envelope hydrophobicity of the SARS-CoV-2 (Fig. 2), affecting the survival (Table 1) and potential transmission of the virus (Mohapatra et al., 2020).

SARS-CoV-2 contains a further lipid membrane enveloping the capsid protein (Kumar et al., 2020b), probably impacting virus adsorption onto a solid fraction of soil (Fig. 2). A virus can be adsorbed by the soil through electron donor-acceptor (e.g. π-π) and hydrophobic (e.g. Van der Waals) interactions depending on the characteristics of the virus (e.g. lipophilicity, presence of functional groups) and soil

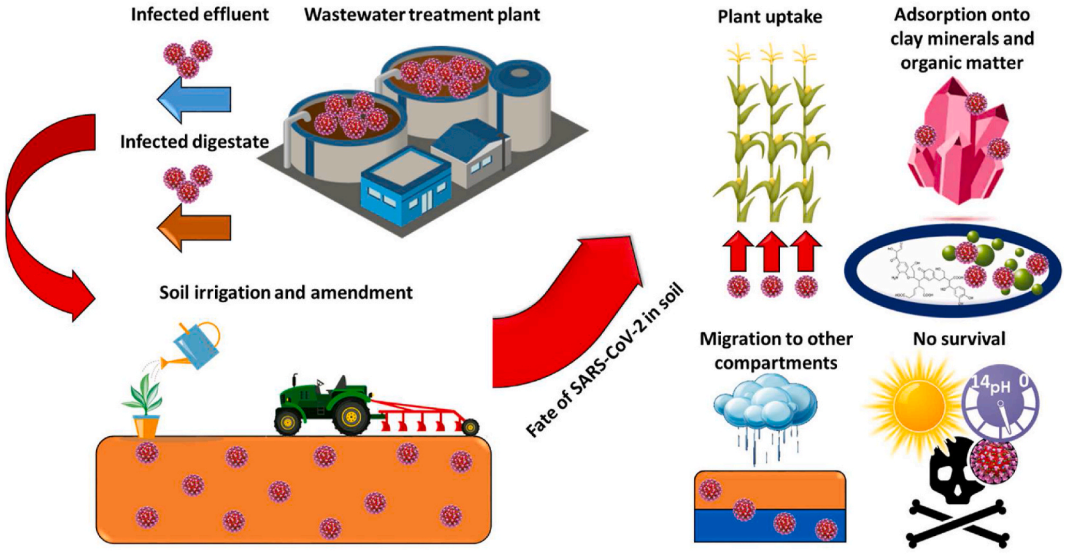

Fig. 2. Possible scenarios regarding the fate of SARS-CoV-2 in soil. SARS-CoV-2 can arrive in soil due to the discharge of infected effluent and digestate after an improper wastewater treatment. Afterwards, the virus can be taken by plants requiring the phosphorus, adsorbed onto clay minerals and organic substances due to electrostatic and hydrophobic interactions, respectively. Otherwise, SARS-CoV-2 can migrate from soil to other environmental compartments due to the reduction of ionic strength during rains. The virus survival can be limited by sunlight radiation, high temperature, acidic pH and the presence of pollutants.

properties (e.g. OM content, pH, ionic strength) (Betancourt et al., 2019; Bianco et al., 2021). Ye et al. (2016) recently simulated the SARS and MERS-CoV partitioning in wastewater, obtaining virus adsorption of 26% by the solid phase. The adsorption of enveloped viruses (e.g. SARS-CoV-2) can occur through hydrophobic interactions onto negatively charged soil particles with a sufficient equilibrium time generating hetero-aggregates (Katz et al., 2018). The occurrence of clay minerals can positively affect the enveloped virus adsorption onto soil particles with the increase of cation exchange capacity (CEC) (Kimura et al., 2008; Nasser, 2002). In addition, the presence of aluminum oxides in the soil can enhance the virus adsorption onto soil particles due to the combination of stronger electrostatic attraction and hydrophobic interactions (Attinti et al., 2010).

Therefore, the migration of coronaviruses from soil to other environmental compartments should be limited (Kumar et al., 2020a), but their survival in soil could be guaranteed due to moisture content and low temperatures (section 2.4) (Mohan et al., 2021). However, the occurrence of acidic soil, i.e. with a pH lower than 7, can inactivate enveloped viral particles (Fig. 2) (Gutiérrez and Buchy, 2012). In addition, sunlight radiation can allow the inactivation of the SARS-CoV-2 virus (Fig. 2) by decreasing the T_{90} to a value lower than 10 min in the function of different parameters (i.e. latitude, season and hour) (Herman et al., 2020). Likewise, the presence of high concentrations of contaminants in environmental matrices (e.g. soil) due to the improper discharge of polluted wastewaters (Papirio et al., 2014) could significantly affect the T_{90} of SARS-CoV-2 (Foladori et al., 2020). For example, Touret et al. (2020) recently identified 15 drugs as inhibitors for SARS-CoV-2 in vitro replication. On the other hand, rainfall provides a higher flow rate in the soil column, thus increasing the viral migration via saturated soil pores, probably due to the decrease of soil ionic strength (Fig. 2) (Kumar et al., 2020c; Lama et al., 2020). Also, the migration of enveloped viruses (e.g. SARS-CoV-2) to other compartments can be enhanced by excessive soil tillage, which was reported to decrease OM content in soil of approximately 40% (Zhang et al., 2020).

3. Direct and indirect impact on the soil before and after pandemic

The advent of the COVID-19 in the world determined the closure of several secondary and tertiary sector activities (e.g. non-essential factories, tourism) as well as the reduction of traffic mobility due to the lockdown measures enforced by national policies, thus improving the air quality and reducing the dispersion of pollutants in soil or sediments (Table 2) (Berman and Ebisu, 2020; He et al., 2020; Tobías et al., 2020). The quarantine actions also allowed the cleaning of recreation areas (e.g. parks, seaside) due to the decrease of human activities in the mentioned spaces (Table 2) (SanJuan-Reyes et al., 2021). In addition, a reduction of the generated amount of solid waste (i.e. 20–30%) was observed during the global pandemic (Klemeš et al., 2020; Ragazzi et al., 2020). All this results in a beneficial effect on soil health status (Table 2), which is also supported by the reduction of SARS-CoV-2 abundance to zero, evaluated both in the middle and low-risk periods in soils sampled after the adopted stringent measures (Zhao et al., 2020).

In addition, the production of medical wastes (i.e. infected and uninfected) significantly raised after the pandemic started (e.g. up to 370% in Hubei province), probably increasing the potential transmission risk of SARS-CoV-2 (Adelodun et al., 2020; Klemeš et al., 2020) due to the improper hospital waste handling taking place in some cases (Table 2) (Yu et al., 2020). Therefore, several nations replaced the policies for recycling with waste landfilling or incineration to prevent the COVID-19 spread through the environment (Zambrano-Monserrate et al., 2020). The temperatures involved in a waste incinerator (i.e. >850 °C) as well as the thermophilic conditions which can occur during aerobic landfilling (i.e. up to 75 °C) can be considered as sanitization strategies to tackle the SARS-CoV-2 in wastes (Di Maria et al., 2020). Chlorine-based disinfection can be alternatively applied for the treatment of infected wastes before proper waste handling (Pandey et al., 2021), and is being extensively applied in WWTPs in disinfection tanks before the discharge of the treated wastewaters to a receiving water body (section 2.5). The major purpose of this way to proceed is the removal of residual pathogenic microorganisms in the waste for secure disposal (Cots et al., 2020). However, the increase of hospital or medical wastes led to the delay of municipal waste management activities (Table 2), which could negatively affect the soil status posing risks for the environment and human health (Kulkarni and Anantharama, 2020; Rahman et al., 2020).

Alongside the hospital and medical waste issue, personal protective equipment is being extensively used by citizens to cope with SARS-CoV-2 spread, such as face masks (e.g. surgical, KN95) (Feng et al., 2020), hand sanitizers (e.g. polyethylene bottles) and single-use plastic gloves (SanJuan-Reyes et al., 2021; Zand and Heir, 2020a), which are made of

Table 2
Impact of COVID-19 on the soil and other environmental compartment status, human and other living organism health, waste management and economy.

COVID-19 impact	Soil status	Other environmental compartment status	Human health	Other living organism health	Waste handling	Economy
Closure of work activities	++	+++	+	+	+	+++
Reduction of traffic mobility	+++	++	++	++	+	+++
Lockdown measures	++	++	+++	++	++	+++
Medical and hospital wastes	+++	++	++	+	+++	+
Personal protective equipment	+++	++	+++	+++	+++	+++

+ Less intensive; ++ Moderately intensive; +++ Very intensive.

recalcitrant and durable materials, thus posing a threat for the soil status. For example, the average generation of health protection wastes considerably raised from 3.64 to 27.32 kg d^{-1} every 1000 inhabitants in Wuhan during the pandemic. This emerging type of waste would increase the amount of plastics yearly estimated to go in the sea and ocean (i.e. up to 12 million tons) (Patrício Silva et al., 2021). Moreover, face masks can be considered as potential microplastic sources in the environment, which could be easily swallowed by living organisms, thus entering the food chain and affecting human health (Aragaw, 2020). Hence, national policies could consider to provide health care waste collection bins in islands to cope with this issue, by raising awareness of the people via social media in order to perform a proper waste delivery (Zand and Heir, 2020b).

4. Methods for quantification of viruses and SARS-CoV-2 monitoring in soil

Soil chemical processes are of crucial importance for long-term sustainability of soils and the overall environment. With that in mind and knowing that it affects to both living organisms and all other components in that environmental compartment, this section mainly focuses on methods for virus elution during the extraction process, virus concentration techniques, detection, and quantification (Fig. 3 and Table 3).

4.1. Methods for virus elution

The first step to be followed for the quantification of viruses in soil matrices is the elution protocol (Fig. 3). Inhibitory compounds that can essentially affect the molecular biology-based techniques employed for virus detection are naturally existing in environmental samples such as soil (Guzmán et al., 2007). Thus, the elution method is a relatively low-cost approach based on the use of inorganic chemicals to minimize the abovementioned issue (Katayama et al., 2002).

Albert and Schwartzbrod (1991) developed a protocol for the elution process of virus in 3% beef extract media at alkaline pH solution (Table 3). Grabow et al. (1991) reported an elution technique using glycine and saline followed by a Freon insert of beef extract media. Furthermore, Alouini and Sobsey (1995) developed the virus elution protocol with 7% beef extract media. Using these protocols, Monpoeho et al. (2001) obtained up to 35% recovery of enteroviruses in bio-solid waste (Table 3). Likewise, Jofre et al. (1989) reported a recovery of enteroviruses up to 28%. Alouini and Sobsey (1995) reported poliovirus

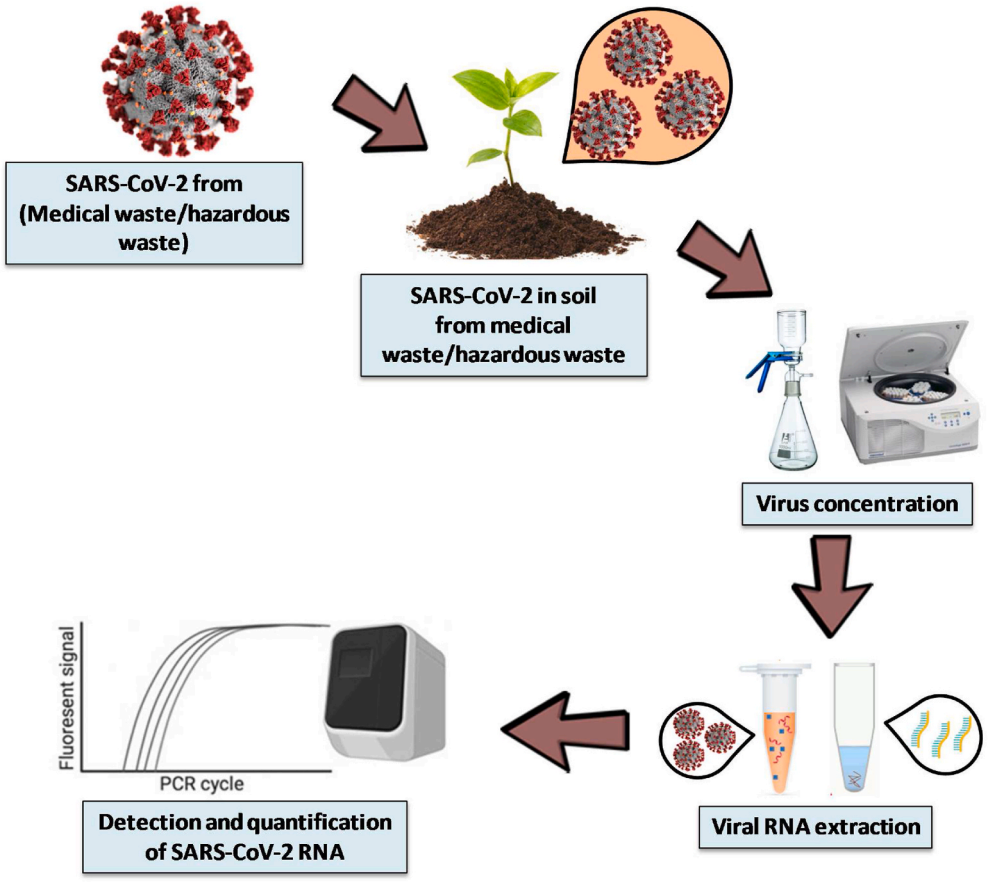

Fig. 3. A sequence of operations for SARS-CoV-2 detection and quantification in soil.

Table 3
Summary of studies reporting methods for detection of viruses in soils, sediments, biosolids, sewage sludges and waters.

S. no.	Sample type	Virus type	Extraction method	Detection method	References
1	Soil, sewage, biosolids	Bacteriophages	Elution method	Calculation of plaque forming units	Guzmán et al. (2007)
2	Coastal seawater	Poliovirus	Adsorption and elution with negatively charged membrane	Cell culture RT-PCR and direct RT-PCR	Katayama et al. (2002)
3	Sewage sludge	HAD (Human adenovirus), HAV (Hepatitis A virus), PV (Poliovirus), RV (Rotavirus)	Elution and organic-based extraction protocol (Phenol/ trizol/Silica based)	PCR, RT-PCR and nested PCR	Schlindwein et al. (2009)
4	Sludge	Enterovirus	Elution-concentration procedures	Agar overlay plaque formation technique	Albert and Schwartzbrod (1991)
5	*Mercenaria Mercenaria* (Hardshell clams)	Poliovirus and HAV	Extraction-concentration method	–	Alouini and Sobsey (1995)
6	Sludge	Enterovirus	Viral-Elution method	Cell culture and RT-PCR	Monpoeho et al. (2001)
7	Coastal sediment	Enteroviruses and Rotavirus	Elution and concentration	BGM cell cultures and indirect immunofluorescence	Jofre et al. (1989)
8	Agricultural soils	Bacteriophages	Elution method	VLP count by epifluorescence microscopy (EFM), and direct counts by transmission electron microscopy (TEM)	Williamson et al. (2003)
9	Sewage	Human astroviruses (HAstV)	Adsorption-elution method	RT-PCR and quantitative real-time (qPCR)	Guimarães et al. (2008)
10	Freshwater, oligotrophic mountain lake, oligomesotrophic lake, eutrophic lake, domestic sewage	Virioplankton	Polyethylene glycol (PEG) and ultracentrifugation	TEM and EFM	Colombet et al. (2007)
11	Shellfish: oysters (Saccostrea forskali), cockles (Anadara nodifera) and mussels (Perna viridis),	Rotavirus	Adsorption-twice elution-extraction method	RT-nested PCR	Kittigul et al. (2015)
12	Potable water	Poliovirus I	Adsorption-elution method	–	Katzenelson et al. (1976)
13	Freshwater	Human rotavirus, Simian rotavirus and Poliovirus	PEG 6000 precipitation method	–	Lewis and Metcalf (1988)
14	Sewage	Group A rotaviruses	Adsorption- elution/ ultrafiltration method	Ultracentrifugation-based method	Fumian et al. (2010)
15	Soil	SARS-CoV-2	RNA extraction (NaCl and PEG 6000)	RT-qPCR	Zhang et al. (2020)
16	Soil	Bacteriophages	Negatively charged HA membranes, PEG and ultracentrifugation (UF)	Random amplified polymorphic DNA (RAPD), TEM	Dias et al. (2020)
17	Freshwater beaches	Human adenovirus (HAdV), Human enterovirus (HEnV), and Human norovirus genogroups I/II	Cation-coated filtration method	qPCR	Lee et al. (2014)
18	Stool	Enterovirus, Adenovirus	Cell culture	PCR and hydridization	Allard et al. (1992)
19	Sediment	Poliovirus and Adenovirus	Elution method	PCR and qPCR	Miura et al. (2009)
20	Groundwater	Enteroviruses, reoviruses, HAV and Norwalk virus	Celite elution procedure	RT-PCR	Fout et al. (2003)

and Human Hepatitis A virus (HAV) recoveries of about 53% and 22%, respectively (Table 3).

Other studies used four different elution buffers for phages in soil samples, specifically 1% potassium citrate, 250 mM glycine buffer, 10% beef extract, and 10 mM sodium pyrophosphate, and found 29% viable phages recovery using glycine and beef extract buffers (Table 3) (Williamson et al., 2003). However, Williamson et al. (2003) found a higher rate of recovery of viruses, reaching up to 65% (Table 3).

US Environmental Protection Agency (US EPA) proposed a protocol for elution to determine viral particles from soil samples, by using an adsorption process in an $AlCl_3$ synthetic solution with acid pH. Researchers that used the US EPA protocol for the determination of different viruses, obtained a recovery of rotavirus, adenovirus, and both hepatitis A virus and Poliovirus of 90, 20 and 100%, respectively (Table 3) (Schlindwein et al., 2009).

Hence, several elution methods are available in the literature with various recovery efficiencies in the function of the physical-chemical properties of each investigated virus (e.g. specific density, morphology) and membrane attachment patterns (Table 3) (Lewis and Metcalf, 1988). On the other hand, the ultracentrifugation (UF)-based method can result in a greater virus recovery compared to the adsorption-elution method (Table 3) (Fumian et al., 2010). The filters utilized in the adsorption-elution approach can be obstructed by the high amount of detritus present in a soil sample, thus decreasing the recovery rate (Table 3) (Guimarães et al., 2008).

4.2. Virus concentration techniques

A concentration technique is generally employed before the quantification of viral particles in soil samples (Fig. 3). Colombet and Sime-Ngando (2012) determined virus particles from infected samples of humans, animals, and plants using different concentration methods. The UF method is commonly used to concentrate viruses (Table 3) (Ammersbach and Bienzle, 2011; Fumian et al., 2010; Nordgren et al., 2009), but the poly-ethylene glycol (PEG) method can be utilized for the determination of virus particles by inducing interaction of crystallinity between DNA during the precipitating and concentrating process (Table 3) (Colombet et al., 2007; Kittigul et al., 2015; Shieh et al., 1999).

Katzenelson et al. (1976) reported a recovery of 75% of poliovirus from soil and sediment samples using the PEG virus concentrated method (Table 3). Similarly, 70% of recovery of rotavirus can be obtained employing the PEG precipitation method at neutral pH, 15% concentration of PEG, and 3% beef extract-sodium nitrate eluent (Table 3) (Lewis and Metcalf, 1988). Both methods were extensively discussed in detail by Peyret (2015). However, Dias et al. (2020) proposed a modified protocol design for concentrating viral particles through UF and PEG techniques in soil samples (Table 3). Briefly, viral particles are taken after reinitializing and subsequently filtered through the diafiltration system. Afterwards, this retentate is re-concentrated through UF and PEG precipitation. Thus, in a UF method, viral pellets are re-concentrated and incubated under agitation with 3 h duration and SM buffer mixtures. In the case of the PEG method, a mixture solution (i.e. PEG 8000 and NaCl) is added to viral ultra-filtered retentates to final concentrations. However, Yamamoto et al. (1970) also reported that a molecular weight of PEG of 6000 can be effective in precipitating macromolecules. Indeed, Zhang et al. (2020) recently investigated the presence of SARS-CoV-2 in soil samples by precipitation with NaCl (0.3 mol L^{-1}) and PEG 6000 (10%) overnight (Table 3).

Humic acid (HA) negatively charged membrane is another method for concentrating viral particles (Table 3) (Lee et al., 2014). In this method, the viral particle is concentrated by adsorption phenomena, where $MgCl_2$ is used as a key agent in the viral suspension sample. Due to adsorption phenomena, the virus gets into the membrane and then the concentrated viral particles are collected for further analysis.

Dias et al. (2020) also determined viruses in soil samples through HA membranes (Table 3). Firstly, 50 mL of suspension sample is added with $MgCl_2$ 25 mM in a HA membrane having 47 mm diameter, and 0.45 μm pore size for viral concentration. Afterwards, the final viral suspension is filtered and centrifuged through a DNA concentrator to get the total volume of 500 μL. In the UF method, the morphology found by transmission electron microscopy (TEM) images showed different viral morphotypes containing larger viral particles. Dias et al. (2020) concluded from TEM images, that a greater number of viral particles can be observed in the HA membrane as compared to the other above-mentioned methods. Indeed, Dias et al. (2020) showed in a metagenome sequencing employing these methods, 24 genera by using UF and HA membrane methods, and in the case of the PEG method, no genus was detected. Williamson et al. (2017) also observed virus particles from TEM images, which were *Podoviridae*, *Myoviridae*, *Siphoviridae* and other non-tailed filamentous structure virus particles.

In addition, Williamson et al. (2005) found different morphologies of viral particles in different soils such as wetland, agricultural, and forested. An amount of 56 and 80% of viral spherical particle structures and tailed phages was reported in agricultural, and other soils, respectively (Williamson et al., 2005). Similar viruses were observed by different researchers in an aquatic environment (Ackermann, 2001; Weinbauer, 2004; Wommack and Colwell, 2000). Swanson et al. (2009) found 70 and 5% viral spherical particle, and tailed phages, respectively, in an agricultural soil sample (Table 3). To be noted that, from TEM images, these researchers reported several non-tailed in addition to the tailed phages.

Hence, the HA membrane method represents a promising approach for obtaining metagenomic data of virus particles from soil samples compared to UF and PEG. Thus, the HA membrane can be considered the most suitable concentration method due to the low required background material in the electron micrographs, and also for recovering various morphotypes and viruses.

4.3. Virus detection

In general, after elution and concentration of viral particles, virus detection methods (e.g. cell culture, molecular techniques) are used for quantification of infectious viruses (Table 3). This procedure evaluates metagenomics after extracting the entire DNA or RNA from soil samples (Fig. 3) using for instance a soil kit (i.e. RNeasy® PowerSoil®), as previously reported by Zhang et al. (2020). Comparing to the isolation method, the latter frequently focuses only on a single virus. The DNA is subsequently broken up into several minute pieces and sequenced. The resulting sequence is examined to build the virus genomes of the sample (Trubl et al., 2020).

Fong and Lipp (2005) described virus detection by cell culture method, and samples were initially inoculated with different cell lines such as Madin-Darby bovine, pig, and buffalo green monkey kidney, CaCo-2, RD, A549, MA104, and FRhK-4. Staggemeier et al. (2011) reviewed different techniques for the detection of viral communities in soils and sediments. The virus detection (Table 3) and quantification techniques include most-probable number assays (MPNs), plaque assays (PAs), Polymerase Chain Reaction (PCR), TEM, epifluorescence microscopy (EfM), and flow cytometry (FC).

Allard et al. (1992) described viral detection through molecular techniques and Polymerase Chain Reaction (PCR), which has several advantages such as high detection limit and good accuracy in very small amounts and is much faster (Table 3) (Griffin et al., 2003; Lee and Jeong, 2004). However, depending on elution protocol and substances present in the soil samples, inhibition phenomena can occur (section 4.1). For example, PCR inhibition was reported in the elution process performed in the beef extract media (Lewis et al., 1985). Miura et al. (2009) reported that HA could hamper PCR detection if present on the soil samples (Table 3). Other researchers utilized hyphenated PCR methods such as Real-Time PCR (qPCR), Nested PCR, Multiplex PCR and Randomly Amplified Polymorphic DNA-PCR (RAPD-PCR) for detection of viral particles on soil samples. Rajtar et al. (2008) used the qPCR technique for detection of DNA of enteric viruses followed by quantification using fluorescence emission. Donaldson (2002) showed a greater detection sensitivity via qPCR than conventional PCR. These methods are commonly applied for the detection and quantification of SARS-CoV-2 in human-deriving samples (Table 4). Ehlers et al. (2005) reported good sensitivity through nested PCR due to its design of two pair primers. Le Guyader et al. (1994) detected rotavirus, and enterovirus in sediment samples using nested PCR. However, Katayama et al. (2002) reported a high probability of contamination through nested PCR (Table 3). Fout et al. (2003) developed a multiplex PCR for detection of enteric viruses in environmental samples and they concluded that multiplex PCR techniques need further investigation towards optimization and reaction conditions of PCR (Table 3). Winget and Wommack (2008) detected DNA fragments in viral-containing soil samples using random PCR amplification of polymorphic (RAPD-PCR) with a single decamer primer. This technique provides a banding pattern that can be used as a proxy fingerprint for the underlying complexity of the original DNA template. They found 105 and 104 viruses per reaction in the soil samples. Srinivasiah et al. (2013) developed RAPD-PCR, which has high accuracy for the detection of soil viral communities. They demonstrated RAPD-PCR fingerprinting is an inexpensive, high-throughput of viral community dynamics within environmental samples.

Suttle (2007) described the protocol of PA method for detection of infected viruses in environmental samples, whose form a clearing (plaque) on a lawn of host cells on solid media. Suttle (2007) also described the MPN method mainly on microplates, which counted viable virus. This method detects the infected virus by the number of dilutions with cell lysis in microplates or well plates. Børsheim et al. (1990) described another technique for virus particles in environmental samples through TEM analysis, which provides data on both the abundance and morphology of virus-like particles. Suttle et al. (1990) used the EfM technique to estimate the virus particles in sediment and marine water samples. This method determined virus particles with a 4', 6-diamidino-2-phenylindole fluorescent dye after virus concentration by different methods. Different fluorescent dyes, such as YO-PRO, SYBR Green and SYBR Gold were also used for the same techniques (Hennes and Suttle, 1995; Noble and Fuhrman, 1998; Winget et al., 2005). However, EfM has virus diversity limitations. Likewise, Brussaard

Table 4
Methods applied for the detection and quantification of SARS-CoV-2 in human-deriving samples.

Type of study	Assay method	Analyte	Concentration range	Tested samples	Reference
Research article	RT-LAMP	RNA	80–500 copies·mL^{-1}	Nasopharyngeal swab	Huang et al. (2020)
Rapid communication	RT-PCR assay	RNA	276 copies·reaction^{-1}	Nasopharyngeal swab	Pfefferle et al. (2020)
Research article	Antibody-based detection	RNA	–	Nasopharyngeal swab	Liu et al. (2020)
Protocol	CRISPR-based approach	RNA	10–100 copies·μL^{-1}	Nasopharyngeal or oropharyngeal swabs	Zhang et al. (2021)
Research letter	CT-SCAN	–	–	–	Fang et al. (2020)

(2004) described the FC method for further detection of virus diversity and quantify subpopulation of viral communities in environmental samples with a change in dye fluorescence frequency.

Some of the advanced technologies and soil management strategies currently available (and/or even other to be developed) are now required to restore soil quality due to the fact that the COVID-19 pandemic has advanced in an accelerated manner, but studies on that topic are still in a rough phase (see section 4.4). With this in mind, focusing specifically on virus detection/quantification, sequence-specific DNA probes designated with a fluorescent colorant could be produced to specifically target virome, with their copiousness analyzed by qPCR. This method can be suitable to quantify specific viruses (e.g. SARS-CoV-2) in the environment, but not quickly illustrative of soil native viruses (Trubl et al., 2020).

4.4. SARS-CoV-2 monitoring in soil and data management

The management of soil quality is undoubtedly required to reduce the urgent issue of environmentally spread diseases from pathogenic microorganisms (Qian et al., 2020). Although no works regarding the presence of SARS-CoV-2 in soil-related liquid samples have been performed (Conde-Cid et al., 2021), enveloped viruses could persist in soil matrices due to their structure and environmental conditions (Section 2.5). Therefore, SARS-CoV-2 should be rapidly monitored in the soil to prevent its transmission to humans by promoting effective detection strategies.

Conde-Cid et al. (2020) recently proposed both i) *in-situ* monitoring and ii) *in-situ* sampling for performing lab analyses. i) *In-situ* monitoring could be carried out by installing specific devices, which should be sterilized (e.g. chemicals, UV lights) before employment (Conde-Cid et al., 2020). Both flat and slope areas can be sampled through piezometers allowing the water vertical-flow from long distances, or otherwise tensiometers and vacuum pumps (Conde-Cid et al., 2020). Thus, the samples can be subsequently analyzed via rapid tests specifically for the detection of SARS-CoV-2 (Krüttgen et al., 2021), in order to evaluate the degree of mobilization of the virus onward the groundwater. ii) *in-situ* sampling for performing lab analyses can be conducted using sanitized stainless steel cores or Edelman-type probes to preserve or not the tridimensional structural integrity of the soil, respectively (Conde-Cid et al., 2020). Afterwards, all samples have to be stored on ice and conveyed to the laboratory for the extraction and quantification methods (see section 4.3) (Randazzo et al., 2020).

Data obtained from the monitoring of SARS-CoV-2 in soil samples could be stored in an online database for soil management. For example, smart web-based geospatial decision support systems (S-DSSs) can be used as a suitable tool for this purpose (Terribile et al., 2015). S-DSSs can combine soil databases with high-performance computing (HPC) to assess the spread of COVID-19 on a large scale with high-resolution detail (e.g. 20 m Sentinel) (Lal et al., 2020). In such context, several issues related to fragmentation of soil policy, detached soil administration for environmental and agricultural subjects, and rough and subdivided geospatial acquaintance regarding soil processes and characteristics, could be overcome.

5. Conclusions

In the current COVID-19 pandemic, different basic essential parameters, mostly focusing on public health, are generally assessed. However, soil health, which plays a major role in bacterial growth, should be carefully evaluated to maintain environmental equilibrium. Indeed, these bacteria populations can be affected by viruses that are transmitted from infected water bodies such as untreated sewage, sewage sludge, and irrigation systems. This could deeply affect soil health likely due to a variety of phenomena, including strong adsorption-desorption of viruses such as SARS-CoV-2. In such a context, the improper treatment of wastewater would pose a threat to human and animal health. The viral load of enveloped viruses such as CoV could be maintained for a prolonged time in the soil environment in the function of different parameters such as the temperature, moisture content, pH, OM, sunlight radiation, and occurrence of clays and nutrients. Moreover, the characteristics of enveloped viruses (e.g. SARS-CoV-2) can influence the virus's mobility in the soil environment. Thus, the presence of SARS-CoV-2 could be directly monitored in soil matrices *in-situ* or with soil sampling for performing lab analyses after extraction and quantification methods (e.g. elution protocol, PEG, PCR). Therefore, future studies about SARS-CoV-2 should be especially aimed at focusing soil characteristics in order to cope with the COVID-19 pandemic and long-term sustainability.

Credit author statement

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Declaration of competing interest

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

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