UNIVERSITÀ DEGLI STUDI DI NAPOLI
FEDERICO II

ENVIRONMENTAL, ECOLOGICAL AND PUBLIC HEALTH CONSIDERATIONS REGARDING CORONAVIRUSES, OTHER VIRUSES, AND OTHER MICROORGANISMS POTENTIALLY CAUSING PANDEMIC DISEASES

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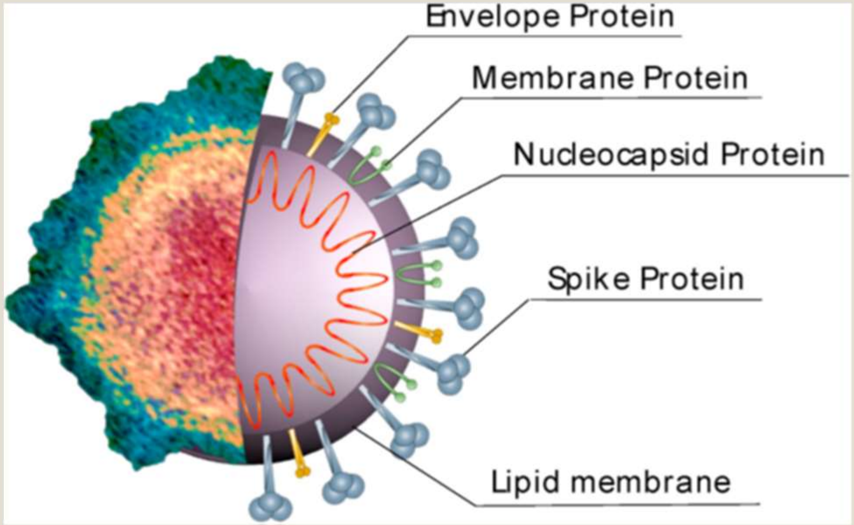
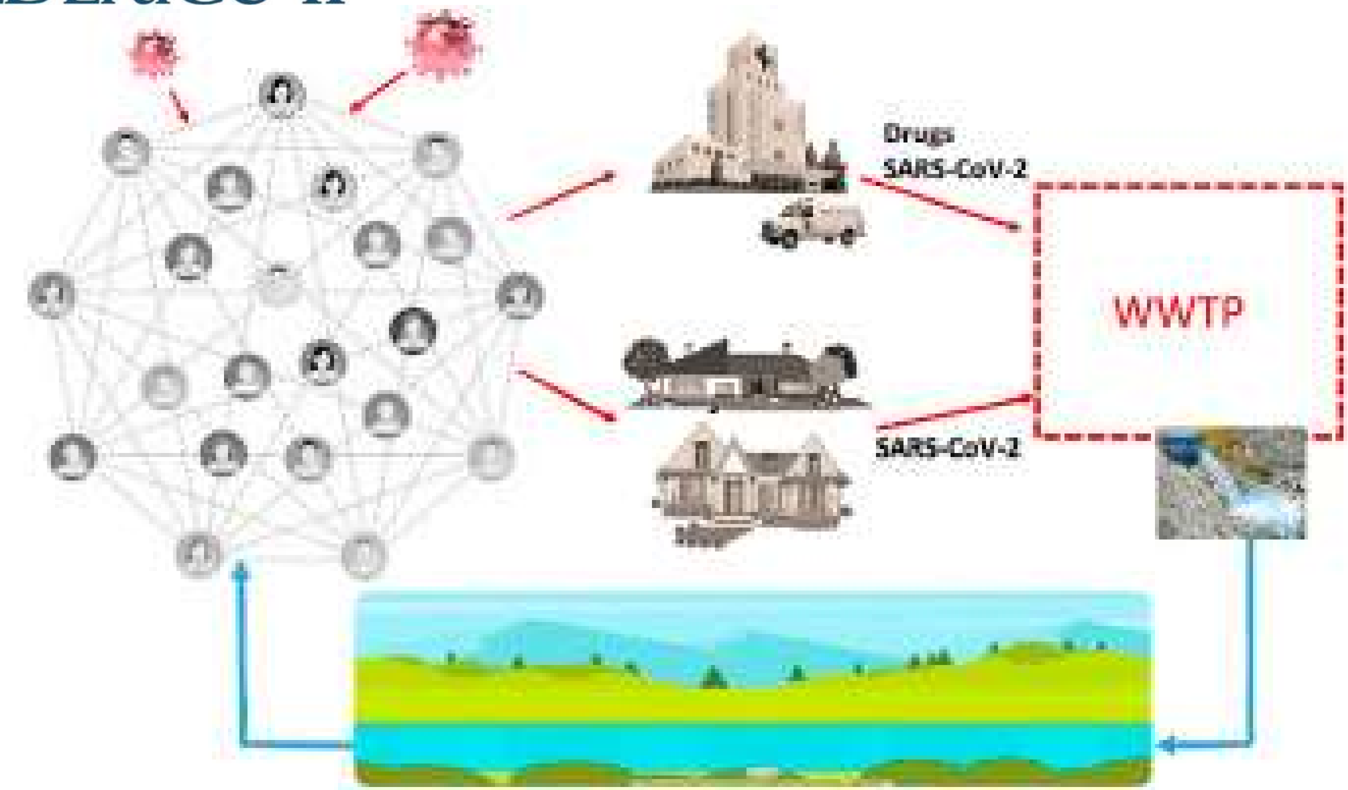


Transmission electron microscope image (left side) and structural schematic representation (right side) of SARS-CoV-2.

- The SARS-CoV-2 (coranavirus that causes Covid-19) is a viral strain from a wide viruses group identified as coronaviruses (Woo et al., 2010).
- The group includes strains such as MERSCoV and SARS-CoV, which have been responsible for outbreak in the last 20 years



- Studies conducted national and globally indicate that genes specific to SARS-CoV-2 can be detected in wastewater
- The number of studies concerning the fate of enveloped viruses in aquatic compartments, instead, is quite limited, as enveloped viruses are predisposed to deactivate in waters

Table 1

Information related to experimental conditions, investigated viruses, concentration/detection methods, and main results on virus persistence in wastewaters reported in literature studies.

Experimental	Virus	Concentration method	Detection method	Virus persistence main results	Reference
21 stool and urine samples collected from Xiao Tang Shan Hospital and 309th Hospital; sewage samples collected for 7 d before disinfection (2500 ml) and after disinfection (25,000-50000 ml)	SARS-CoV	Positively charged filter media particles	RT-PCR assay	• No presence of infectious SARS-CoV • SARS-CoV RNA detection in stool samples (7 on 11) of symptomatic patients • No RNA detection in urine samples and in stool of recovered patients • RNA detection in sewage samples before disinfection and RNA detection in sewage after disinfection only in 3 d	Wang et al. (2005a)
Sewage samples collected before disinfection (2500 ml) and after disinfection (25,000-50000 ml) from Xiao Tang Shan Hospital, 309th Hospital and housing estate	Bacteriophage f_2 (as coronavirus model) and SARS-CoV	Positively charged filter media particles	RT-PCR assay	• No presence of infectious SARS-CoV • SARS-CoV RNA detection in sewage samples before the disinfection • RNA detection in sewage after disinfection only in 3 d • Average f_2 recovery from hospitals samples of 79.2 and 85.8% before disinfection and 61.2 and 85.5% after disinfection	Wang et al. (2005b)
Samples of stool (3) and urine (2) from Xiao Tang Shan Hospital; wastewater samples from 309th Hospital; sewage samples from housing estate; disinfection tests on wastewater with different chlorine (by dissolution of sodium hypochlorite) or chlorine dioxide concentration and disinfection time	Bacteriophage f_2 (as coronavirus model) and SARS-CoV	–	RT-PCR assay	• Persistence of SARS-CoV in wastewater and sewage samples for 2 d at 20 °C and 14 d at 4 °C • Persistence of 3 d in stool and 17 d in urine at 20 °C and persistence > 17 d at 4 °C • Complete SARS-CoV inactivation with 10 ppm of chlorine after 10 min • Reduced effectiveness in presence of chlorine dioxide • Total inactivation with 20 ppm of chlorine in 1 min and with 40 ppm of chlorine dioxide in 5 min	Wang et al. (2005c)
Wastewater samples collected from wastewater treatment plant and pasteurized; comparison with reagent-grade and lake water; tests on temperature effect carried out at 23–25 °C and 4 °C	TGEV and MHV (as surrogates coronaviruses)	–	–	• 99% decrease in infectious titer equal to 22 d for TGEV and 17 d for MHV at 25 °C • No significant decrease over 49 d at 4 °C in reagent-grade water and 99% decrease in infectious titer equal to 13 d for TGEV and 10 d for MHV at 25 °C • $1log_{10}$ decline for TGEV and no decline for MHV after 14 d at 4 °C in lake water • 99% decrease in infectious titer equal to 9 d for TGEV and 7 d for MHV at 25 °C in pasteurized settled water • 99% decrease in infectious titer equal to 49 for TGEV and 70 d for MHV at 4 °C in pasteurized settled water	Casanova et al. (2009)
Samples of unfiltered tap water tested at 23 °C and filtered tap water tested at 23 and 4 °C; samples of filtered and unfiltered primary effluent tested at 23 °C; samples of unfiltered secondary (activated sludge) effluent tested at 23 °C	Feline infectious peritonitis virus (FIPV), Human coronavirus 229 E (HCoV) and Poliovirus 1 LSc-2ab (PV-1)	–	Plaque assay or $TCID_{50}$	• 99% virus titer decrease of 6.76 d (for HCoV and FIPV) and 43.3 d (for PV-1) in filtered tap water at 23 °C • Higher persistence in unfiltered tap water at 23 °C equal to 8.09 d for HCoV, 8.32 d for FIPV, and 47.5 d for PV-1 • Persistence in filtered tap water at 4 °C equal to 392 d for HCoV, 87 d for FIPV, and 135 d for PV-1 • 99% virus titer decrease of 1.57 d for HCoV, 1.60 d for FIPV, and	Gundy et al. (2009)

Infection with SARS-CoV-2 , is accompanied by the shedding of RNA from the virus in stool. Therefore, the quantification of SARS-CoV-2 in wastewater affords the ability to monitor the prevalence of infections amongst the population via wastewater-based epidemiology (WBE).

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Potential environmental impact of administered drugs

Toxic effects of drugs used for the COVID-19 disease treatment on selected models and biomarkers.

Compound	Organism	Species	Endpoint (exposure time)	EC50 (ppm)	Reference
Tocilizumab	Alga	*Desmodesmus subspicatus*	Growth rate inhibition (72 h)	> 100	Roche safety data sheet (2018)
	Alga	*Desmodesmus subspicatus*	Biomass inhibition (72 h)	> 100	
	Crustacean	*Daphnia magna*	Immobility (48 h)	> 100	
	Fish	*Danio rerio*	Embryotoxicity (96 h)	> 100	
Chloroquine	Bacteria	*Aliivibrio fischeri*	Bioluminescence Inhibition (24 h)	132.1	Zurita et al. (2005)
	Alga	*Chlorella vulgaris*	Growth Inhibition (24 h)	133.3	
	Crustacean	*Daphnia magna*	Immobility (24 h)	21.5	
	Topminnow	PLHC-1 cell line	Protein content (24 h)	158.3	
	Basket willow	Salix viminalis	Relative transpiration (NRT) (117 h) (pH from 6 to 8)	7–28	Rendal et al. (2011)
	Crustacean	Daphnia magna	Immobility (48 h) (pH from 7 to 9)	4–30	
Hydroxychloroquine	Alga	Raphidocelis subcapitata	Growth rate (72 h)	3.1	FASS safety data sheet (2019)
	Crustacean	Daphnia magna	Immobility (48 h)	14	

In the current pandemic, the existing drugs are being administered in much larger amount so representing an important threat to the quality of the receiving water bodies.
Most of these drugs are excreted as unchanged parent compounds with highly bioactive characteristics which are resistant to conventional treatments in wastewater treatment plants. Moreover, they can react with organic and inorganic constituents during wastewater treatment and can be transformed in additional molecules characterized by higher persistence