Antiviral surfaces and coatings and their mechanisms of action

Paulina D. Rakowska¹,²✉, Mariavitalia Tiddia¹, Nilofar Faruqui¹, Claire Bankier¹, Yiwen Pei¹, Andrew J. Pollard¹, Junting Zhang¹ & Ian S. Gilmore¹

Viral infections are a serious health challenge, and the COVID-19 pandemic has increased the demand for antiviral measures and treatments for clean surfaces, especially in public places. Here, we review a range of natural and synthetic surface materials and coatings with antiviral properties, including metals, polymers and biopolymers, graphene and antimicrobial peptides, and their underpinning antiviral mechanisms. We also discuss the physico-chemical properties of surfaces which influence virus attachment and persistence on surfaces. Finally, an overview is given of the current practices and applications of antiviral and virucidal materials and coatings in consumer products, personal protective equipment, healthcare and public settings.

In today’s global society, disease outbreaks can spread rapidly and easily cross borders and continents, having a catastrophic impact on health and the global economy. There is no single solution to prevent the spread of viral infections. Different modes of infection transmissions such as through aerosol, droplets or fomites (everyday use surfaces), add to the problem. Therefore, multiple-barrier protection is often required and, next to high hygiene standards and vaccination programmes, a number of control measures including adequate personal protective equipment (PPE) or antiviral surfaces in public facilities, e.g., schools, health centres or airports are important to reduce transmission of viruses. A vast amount of research, concerned with antimicrobial properties of different surface materials and coatings, has been published. However, the antiviral or viricidal properties of materials are less well known. While bacteria are single-celled living organisms, viruses are not considered to be ‘alive’ due to reliance on a host to reproduce and survive. Many materials exert both antiviral and antimicrobial properties, but there are also distinct differences in responses to preventative measures and treatments often dictated by the differences between bacterial and viral structures and behaviours. We provide an overview of surface materials and coatings that exhibit antiviral and (or) virucidal properties, providing, where possible, their mechanism of action. A brief introduction to viruses, their persistence and methods used in virology research is also given to context the findings. The focus of this review is to gain a better understanding on the state of knowledge of antiviral properties of different types of surface materials and on their practical and potential applications. Box 1 introduces many of the key terms and phrases used in this review.

Virus

Viruses are biological entities built of DNA or RNA core wrapped in a protective protein coat. They are one of the most diverse groups of microorganisms with 6590 species recognised by the International Committee on Taxonomy of Viruses. Viruses infectious to humans typically range in size from 20 to 260 nm, although a few classes of viruses can grow much larger. Virus life cycles all comprise attachment and entry, replication and assembly and egress. Attachment and entry is via the virus capsid or envelope proteins. In mammalian cells, virus replication can be
Box 1 | Key terms

| Term                                      | Definition                                                                 |
|-------------------------------------------|---------------------------------------------------------------------------|
| Antiviral agents                          | Chemical agents, most often drugs, inhibiting the proliferation of viruses|
| Defective viral particle                  | Spontaneously generated virus mutants, thought to be caused by replication errors and believed to play a role in immune system priming and modulation of disease severity |
| Infectivity                               | Ability of the virus to cause infection by transmission between biotic and/or abiotic ecosystem constituents; |
| Persistence                               | Ability of the virus to survive in the environment in sufficient numbers to act as a source of infection (per time interval) |
| Viability                                 | Ability of the virus to replicate and infect                               |
| Viral titre                               | Number of infectious particles in a volume of fluid;                       |
| Viron                                     | Complete, infectious form of a virus                                       |
| Virucidal agents                          | Physical or chemical agents, e.g., disinfectants, that deactivates or destroys viruses |

Fig. 1 Schematic of a virus structure based on coronavirus. The nucleic acid for replication is protected by a protein capsid which is contained within a protective envelope consisting of envelope proteins, glycoproteins and is decorated with a spike protein to bind to cells.

completed within several hours producing upwards of $10^3$ virions per cell. Viruses rely on the host cell’s machinery to reproduce, amplify and release from the cell. Some, e.g., retroviruses, can carry their own enzyme (polymerase or reverse transcriptase) for replication but still cannot reproduce and amplify outside a host cell. Virion assembly occurs once enough nucleic acids and proteins are present. The structure of a virus (Fig. 1) is crucial to understanding the properties it might possess, such as infectivity potential, susceptibility to disinfectants or mode of replication. Typically, a virus will be surrounded by a protective protein layer—capsid. Capsids are formed in a highly specific manner and their function is to protect the viral genome. There are three main types of capsid shape: icosahedral, helical and complex (such as the pox virus capsids). Prokaryotic viruses (bacteriophages) have an elongated icosahedron shape termed a prolate. Capsid shapes are used to aid the identification and can provide insight into the life cycle of a virus. Some viruses, such as influenza or coronavirus, have an additional protective lipid bilayer—envelope. The envelope is often derived from the host’s plasma membrane during their release from the cell in a process called budding. Additional proteins (glycoproteins) are then incorporated into this envelope in the form of spikes. These spikes facilitate entry into host cells and coupled with the envelope can have multiple roles in virus-host interactions. The envelope offers the virus many advantages such as playing a protective role in the process of budding out from the cell. It also aids structure flexibility and helps to mask capsid spike antigens from antibodies in the host’s immune system. This ability to evade the host immune response could present an important factor in viral infection outbreaks. Even though the non-enveloped viruses are more likely to establish human-to-human transmission, the majority of the recent viral years viral epidemics such as Ebola, measles, Zika, avian influenza, SARS, MERS, and the ongoing COVID-19, have been caused by enveloped viruses. Although the lipid bilayer in enveloped viruses can offer additional protection to the virus, it can also be detrimental to their survival outside of the host cell as the lipid bilayer can degrade upon harsh physical or chemical conditions. Non-enveloped viruses tend to be more environmentally stable and resistant to detergents and heat. Non-enveloped viruses are more commonly found in extreme environments than enveloped viruses which demonstrates resistance to environmental stress. However, whether they can survive harsher conditions due to their lack of a lipid bilayer or whether they evolved a more resistant capsid layer in these extreme environments is yet to be fully understood.

Viral activity measurements and detection methods

There are a variety of methods to detect viruses and measure their activity and international standards to assess virucidal activity (Table 1). The most common assays used to test the presence and the persistence of viruses are summarised in Table 2. The methods can be divided into three general groups: (1) cell-based assays to measure viral infectivity, (2) gene or protein expression assays to detect virus presence and (3) direct count of viral particles.

Viral persistence on surfaces. Several factors influence the spread of viral infections throughout a population. These include age, structure, availability of a new host and route of transmission. Some viruses can be infectious to different species and cross-species transmission is also possible. For example, coronavirus are known to affect a wide range of birds and mammals, including household animals such as cats and dogs and the recent emergence of human CoVs capable of causing respiratory failure, such as SARS-CoV, MERS-CoV and COVID-19 has had the origins traced back to animals such as bats. Transmission of the viral particles between humans is often caused by respiratory, skin or other bodily fluid secretions from an infected individual. The virus can be spread directly through aerosols and droplets or indirectly through contact with contaminated surfaces (fomites). Viral contamination on fomites could cause multiple infections within a community. For example, norovirus (a non-enveloped virus) is well known to persist for several weeks on many different types of surface causing large outbreaks in healthcare facilities. Aerosols and droplets are a common route of transmission for many respiratory viruses including influenza and SARS-CoV. Aerosol transmission is often considered the most dangerous as it can spread infectious particles in high titres with limited infection control strategies available to prevent the spread. Therefore, understanding of how long viruses can survive on a surface can help inform infection control strategies and control potential outbreaks.

There are four main properties, which can affect the persistence of viruses on surfaces (Fig. 2). These include: (i) material form such as porosity; (ii) physical, abiotic factors such as relative humidity, temperature, exposure to light and type of surface, e.g., roughness; (iii) biological, like the structure of the virus or presence of other microorganisms, e.g., microbial biofilm.
Environmental conditions, including but not limited to, temperature, humidity (typical for indoor environments), the virus persisted for more than 24 h. Therefore, an outer fluid-absorbing layer in the design PPE garments and medical devices may be advantageous. A more recent study\(^{40}\) shows that, at 21–23 °C and 40% relative humidity, both the SARS-CoV-1 and SARS-CoV-2 coronavirus appeared to be more persistent on cardboard as compared to non-absorbent materials like plastic and stainless steel.

**Effect of surface physical properties on viral persistence**

Since the discovery of the first virus in 1892\(^{42}\), considerable efforts have been made to understand viral survival in different environments and to estimate the impact of surface properties on their viability. Major contributing factors with regards to surface properties include, among others\(^{33}\), porosity\(^{29,34–40}\), absorption\(^{41,42}\) and surface hydrophobicity\(^{43,44}\) (Fig. 2). Each type of virus interacts with a surface in its own unique way, and therefore the design of effective antiviral surfaces may need to be tailored for a specific type of virus. Viral persistence can also be influenced by other environmental conditions, including but not limited to, temperature, relative humidity or how viruses absorb onto various surfaces\(^{39}\). These additional factors should be taken into consideration when designing an antiviral surface\(^{39,32,33,37,38,41–44}\).

**Porosity.** The porosity of inanimate surfaces plays a significant role in viral survival and several studies have compared the persistence and time of complete decay of different types of viruses on porous and non-porous surfaces\(^{27,29,34–36,39}\). It is found that the majority of viruses exhibit greater persistence on non-porous materials than porous surfaces\(^{29,35}\), although there are exceptions\(^{45,46}\). In the case of influenza A virus, at 35–40% humidity (typical for indoor environments), the virus persisted for more than 24–48 h on stainless steel and plastic surfaces but the number of infectious viral particles dropped to undetectable levels after 8–12 h on porous surfaces such as cloth or paper\(^{27,36}\). It is thought that one of the contributing factors to poor virus persistence on porous surfaces could be complete drying of the viruses on the surfaces or insufficient elution of the viruses from these materials\(^{35}\). Both the SARS-CoV-1 and SARS-CoV-2 coronavirus demonstrated similar persistence in terms of surface porosity\(^{27,36}\). The reports show that these viruses persisted longer (period of days) on the surfaces with low porosity such as plastic and stainless steel than on the relatively porous surfaces such as cardboard (period of hours)\(^{27,36}\). It has been reported that the infectious SARS-CoV-2 virus remains on the outer layer of a surgical mask after seven days, whereas no viable virus was detectable on treated smooth surfaces of stainless steel and plastic after seven days\(^{34,38}\).

**Absorption.** Several studies suggested that the antiviral properties of surfaces can be associated with the ability to absorb a virus\(^{41,42}\). Due to the nature of liquid absorption, in contrast to fluid-repelling surfaces, absorbent materials (such as cotton and cardboard) can offer more protection against virus-containing respiratory droplets\(^{47,48}\). Lai and co-workers\(^{42}\) compared the survival of SARS-CoV\(^{2}\) between two different hospital PPE gowns, a fluid-repelling disposable type and a cotton gown. They found that highly concentrated droplets of virus culture were immediately absorbed by the cotton material and no viable virus was detectable after 1 h, whereas most of the virus remained viable on disposable gowns for up to 24 h. Therefore, an outer fluid-absorbing layer in the design PPE garments and medical devices may be advantageous. A more recent study\(^{40}\) shows that, at 21–23 °C and 40% relative humidity, both the SARS-CoV-1 and SARS-CoV-2 coronavirus appeared to be more persistent on cardboard as compared to non-absorbent materials like plastic and stainless steel.

**Surface hydrophobicity.** Viral persistence can be also affected by surface hydrophobicity\(^{43,44}\). Specifically, the hydrophobicity of the outer layers of proteins in virus capsids can affect its interactions with solid surfaces and the environment\(^{49}\). Building a fundamental understanding of the interactions between viruses, i.e., the outer surface of proteins, and solid surfaces is therefore vital for controlling environmental transmission and designing efficient antiviral strategies. Both experimental and computational analysis have been used to determine the hydrophobicity of different viruses\(^{30,31}\). Chattopadhyay et al.\(^{44}\) demonstrated that the sorption of hydrophobic viruses, i.e. viruses carrying hydrophobic proteins outer layers, was favoured by surfaces coated with hydrophobic sorbents, whilst the sorption of hydrophilic viruses was favoured by hydrophilic surfaces\(^{44}\.)

### Table 1 Overview of international standards to assess virucidal activity on surfaces and the effect of virucidal agents.

| Standard | Title |
|----------|-------|
| ISO Standards | Measurement of antiviral activity on plastics and other non-porous surfaces. Textiles - Determination of antiviral activity of textile products. |
| ISO 21702:2019(en) | Standard practice to assess the activity of microbicides against viruses in suspension. Test method to assess virucidal activity of chemicals intended for disinfection of inanimate, nonporous environmental surfaces. |
| ISO 18184 | Test method for neutralization of virucidal agents in virucidal efficacy evaluations |
| ASTM Standards | Test method for determining the virus-eliminating effectiveness of hygienic handwash and handrub agents using the fingerpads of adults |
| E1052 | Quantitative disk carrier test method for determining bactericidal, virucidal, fungicidal, mycobactericidal, and sporidical activities of chemicals |
| E1053 | Terminology relating to antimicrobial and antiviral agents |
| E1482 |  |
| E1838 |  |
| E2197 |  |
| E2756 |  |
| Federal Standards |  |
| 21 CFR Code of Federal Regulations (CFR), Food and Drug Administration, Part 58 | Laboratory practice for nonclinical laboratory studies |
| 40 CFR Code of Federal Regulations (CFR), Environmental Protection Agency, Part 160, | Good laboratory practice standard |
| European Standards | Chemical disinfectants and antiseptics—Quantitative suspension test for the evaluation of virucidal activity in the medical area—Test method and requirements (Phase 2/Step 1) |
| EN 14476:2013 + A1:2015 |  |
**Table 2 Overview of methods to detect viruses and measure their activity used in virology.**

| Assay Group 1 |  |
|---------------|------------------|
| **End point dilution assay (TCID<sub>50</sub>)** | Method: The end point dilution assay (also known as the tissue culture infective dose (TCID) assay), involves titrating a known viral titre in a cell line to estimate where 50% of cells in a cell culture die. The Spearman-Karber or Reed-Muench formula is commonly used to calculate the 50% point using serial dilutions. Modifications of this assay include using colorimetric methods (coloured tetrazole) to improve reliability. Results are determined by observing the microscopic cytopathic effects (CPE) caused by the virus in the monolayer of the cells. Results are often difficult to interpret and subjective. This assay is commonly used in the biotech industry to understand how effective viral clearance procedures are at removing viruses when developing blood/tissue-derived products, biologics and monoclonal antibodies. |
| **Plaque assay** | Method: Plaque assays are a common method to determine the virus titre by determining the number of plaque forming units (PFU). Similar to the TCID<sub>50</sub> assay, a serial dilution of a virus is inoculated into a plate containing a cell line. The virus lyses the cell and spreads from cell to cell. Cell lysis can be viewed either by eye or microscope. The method assumes that one PFU is a single infectious virus. Cells can be fixed and stained with crystal violet to help count PFU’s. Results: Plaque assays are regarded as one of the most quantitative methods used to assess antiviral substances and infectious virions. However, it provides the measure of only live (not defective) viruses. Plaque assays can only be used on viruses that cause cell lysis (influenza, herpesvirus). The assay often utilises an overlay method (such as using agarose or cellulose) to immobilise the cells and restrict the spread of the viruses. Modifications of the assay can produce variable results. |
| **Focus forming assay** | Method: The Focus Forming assay is a type of plaque assay that uses immunostaining techniques to detect PFU’s. Fluorescent antibodies can be used to detect specific viral antigens in infected host cells. Overlay methods similar to the traditional plaque assay are used and fluorescent microscopy can be used to detect the infected cells. Results: The focus forming assay is known to be as accurate as the TCID<sub>50</sub> method. However, expensive reagents are required and often specialist software is used to detect the foci. This method is not as commonly used as other plaque assays. |

**Assay Group 2**

| qPCR | Method: Real-time PCR (qPCR) is a molecular method that can be used to quantify and detect viruses. DNA or RNA from the virus can be extracted from a sample and specific primers and probes are selected dependent on the target virus. The reactions are then set up and amplified on a real-time PCR Thermal Cycler. If a virus is present, it can be quantified by the increase in cycle threshold which is proportional to the amount of nucleic acid in the sample. Results: This method can yield accurate and fast results and is often used in clinical settings to detect amount of viruses in patient samples. It is a quantitative method but can yield false negatives or positives and kit contamination can be an issue. |
| ELISA | Method: ELISA (Enzyme-linked immunosorbent assay) is often used in clinical settings to detect blood-borne viruses in particular. The assay uses enzymes linked to antibodies that attach to matching viral antigens. The enzyme substrate is then added (most common is horseradish peroxidase) which produces a signal if detected (most often a colour change). This colour change can be quantified by optical density (OD) using a spectrophotometer. Results: ELISA is a quantitative detection method but is often considered less quantitative than qPCR. ELISA can use standard equipment (spectrophotometer) and commercial kits are common. |

**Assay Group 3**

| Flow cytometry (flow virometry) | Method: Flow cytometry is used to analyse and sort cells. Recently, flow cytometer resolutions have improved and now have the capability to detect virus particles from background noise. Traditional forward light scatter has been improved to block light and noise at specific angles. More powerful lasers and detectors have also been developed which are more effective at differentiating between nanoparticles and cells. Results: Flow cytometry can be a very useful tool to detect the amount of viruses in a sample. It provides a complete count of virus particles including live and defective viruses. However, standard flow cytometers will have difficulty distinguishing the virus from particulate matter in samples or reagents and therefore significant investment is required for a high spec machine. However, flow cytometry can be highly accurate and can be used to characterise and track viruses through generations which is useful when trying to understand the origins and evolution of viruses. |
| Electron microscopy (EM) | Method: Electron microscopy is a well-established imaging method for detecting and characterising viruses. The method requires the sample to be preserved, followed by negative staining. Negative staining creates contrast between the sample and the background with an electron opaque (usually heavy metal based) stain, allowing the structures of the virions to be visible. Results: Electron microscopy is distinguished from other diagnostic methods, such as ELISA or qPCR as it can characterise interactions with cells and image new species that are undetected by antibodies. It is a technique that is more often used when imaging can be advantageous over other assays, for example to study novel or unknown viral infections. Electron microscopy provides quantification of all viral particles (defective and live). The defective particles are thought to be caused by replication errors and may play a role in immune system priming. |

**Antiviral materials and their proposed mechanisms of action**

**Metals and metal surfaces.** The biocidal action of heavy metals, occurring even at their low concentration, sometimes referred to as an oligodynamic effect was first described, in modern times by Karl Nägeli in 1893. More than 30 heavy metals are potentially able to interact with microorganisms and these include silver (Ag), gold (Au), bismuth (Bi), cobalt (Co), copper (Cu), iron (Fe), mercury (Hg), manganese (Mn), nickel (Ni), lead (Pb), platinum (Pt), antimony (Sb), tin (Sn), titanium (Ti), and zinc (Zn). It has been known for centuries that some heavy metals possess anti-infective properties. However, the mechanisms behind those properties are not yet entirely understood and several factors...
seem to be contributing. It is crucial to note that metal ions are integral to some viral proteins and processes. Therefore, an imbalance in metal concentration or proximity or presence of other reactive metal ions can have an effect on virus viability. Chaturvedi et al.\textsuperscript{53} state that metal ions play an important role in a range of processes including, for example, regulation of transcription and translation, reverse transcription, nucleic acid chaperoning and folding and unfolding, catalysis, minus- and plus-strand transfer, nucleic acid cleavage, inhibition of membrane current. Metals can also have a structural role. Zinc, magnesium and copper are the most common metals that bind with viral proteins.\textsuperscript{53–55} Knowledge of metal-binding/interaction mechanisms could prove useful in the design and development of antiviral materials and viral inhibitors. Bactericidal properties have been assessed and well documented for a range of metals, especially copper and silver. These have also been shown to possess potent virucidal properties as described below.

**Copper.** The virucidal properties of copper and copper alloy surfaces have been tested in several studies. Sagripanti et al.\textsuperscript{56} demonstrated the virucidal activity of Cu(II), in comparison to Fe (III) ions, on five viruses, representing different classes including enveloped and non-enveloped, single- and double-stranded DNA or RNA viruses. The virucidal effect of these metals was further enhanced by the addition of peroxide, particularly for Cu(II). They showed that mixtures of Cu(II) ions and peroxide were more efficient, under the applied test conditions, than glutaraldehyde in inactivating phi X174, T7, phi 6, Junin, and herpes simplex viruses (HSV's). This study suggested that such substances/mixtures have the potential to inactive most, if not all, viruses that have been found contaminating medical devices.\textsuperscript{56} Warnes and Keevil\textsuperscript{57} observed the effects of Cu on infectivity of murine norovirus, MNV-1. MNV-1 is a non-enveloped, linear, single-stranded, positive RNA virus and a close surrogate for the human norovirus, causing gastroenteritis. The viability of the virus was lost upon exposure to copper and copper alloys but retained on stainless steel. The researchers conducted two types of experiments simulating (i) slow drying vomitus (wet inoculum) and (ii) hand-to-hand/object transfer (dry inoculum). In the first study it was not possible to recover infectious virus after as little as 30 min of exposure to 100% Cu surface and 60 min to copper (89%)-nickel alloy. In the dry inoculum method, the inactivation was found to be 5 min for both surfaces. The study showed also that the contact of the virus with copper surfaces destroyed the viral RNA genome. In a subsequent study\textsuperscript{58} by the same group, it was observed that viral capsid integrity was compromised upon contact with a range of copper alloys and brasses. It was also revealed that only a small change in the percentage of copper, of 10%, had a significant effect on the virucidal properties of different copper alloys and brasses. A very similar study\textsuperscript{59} was conducted by the same group on a Human Coronavirus 229E, used as a surrogate for the more virulent coronaviruses responsible for SARS, MERS and now COVID-19. The research confirmed a rapid inactivation of the human coronavirus on brass and copper-nickel surfaces, using metal surfaces that did not contain copper (stainless steel, zinc, and nickel) as controls. With the wet inoculum method, the virus was inactivated in <40 min on brass and 120 min on copper-nickels containing <70% copper. Comparison was made with stainless steel and nickel surfaces, which did not demonstrate any virucidal activity, although mild antiviral activity was observed on zinc. The dried inoculum method was found to have inactivated the virus approximately eight times faster.

The virucidal properties of copper rely, primarily, on the release of copper ions. Both ionic species, Cu(II) and Cu(I), contribute to virus inactivation but the effect of Cu(I) seems to be longer term\textsuperscript{59}. This was demonstrated by Warnes et al.\textsuperscript{59}, who inoculated HuCoV-229E onto 100% copper and brass (70% Cu) surfaces in the presence of ethylenediaminetetraacetic acid (EDTA) and bathocuproine disulfonate (BCS), chelators of Cu (II) and Cu(I), respectively. Both chelators initially protected the virus from inactivation for up to 2 h with the BCS being still protective on brass after 2 h. There is also evidence for the generation of reactive oxygen species (ROS) contributing to virus inactivation.\textsuperscript{58,59} Carubelli et al.\textsuperscript{60} suggests that the cytotoxicity may be caused by free radicals interacting with protein components of a virus, through the generation of hydroxyl radicals of transition metal ions bound to the proteins. Secondly, oxidised nucleic acids may undergo degradation (in cells).\textsuperscript{61} Under oxidative stress, copper is well known to generate ROS, that damage nucleic acids, through Fenton-like and other reactions.\textsuperscript{60–63} The effect of ROS was also tested by Warnes et al.\textsuperscript{59}, HuCoV 229E was inoculated on copper and brass surfaces, in the presence of D-mannitol and Tirno (4,5-dihydroxy-1,3-benzene disulfonic acid) to quench hydroxyl radicals and superoxide anions, respectively, in order to determine whether these moieties were involved in the virus inactivation mechanism. Tirno protected the virus for the first hour of contact, suggesting the importance of superoxide generation. D-mannitol was only minimally protective on copper but protected the virus for the duration of the test on brass. Increasing the concentration of D-mannitol did not prolong the survival of the virus on copper. The researchers concluded that though the rapid inactivation of the virus is primarily due to copper ion release with minimal effect from ROS, the role of ROS becomes more significant with decreasing percentage of copper in the alloy.\textsuperscript{59} It has been also documented that the oxidation and oxidative stress such as the addition of peroxide or ascorbate

---

**Fig. 2 Surface properties influencing the persistence of viruses.** These include physical properties (including light exposure, temperature and humidity), chemical properties (such as pH or antiviral coatings) and biological properties (depending on the virus vulnerabilities, e.g., an envelope) as well as the type of surface such as the porosity or topography.
could enhance the biocidal properties of copper. Cu ions can also interact with different viral components, e.g., can cause the inactivation of viral metalloproteins by replacing the respective metal with copper. Proteins tend to select divalent metal ions from the environment, following the Irving-Williams order of stability, Mg$^{2+}$ < Mn$^{2+}$ < Fe$^{2+}$ < Co$^{2+}$ < Ni$^{2+}$ < Cu$^{2+}$ < Zn$^{2+}$. Copper ions are at the top in the series, with the ability to replace the weaker binding ions.

Copper ions have been shown to be able to directly inhibit virus’s proteases by reacting with surface cysteine and to inflict damage to the viral genome in HIV (single-stranded, positive-sense, enveloped RNA viruses) and HSV (enveloped, double-stranded, linear DNA) 

Silver. The biocidal properties of silver are well documented. The attractiveness of using silver as a biocidal agent lies in its low toxicity and biocidal effect at low concentrations. Since ancient times silver was used in water containers to prevent the putrefaction of liquids and foods. Silver was also mentioned in the Roman pharmacopoeia of 69 B.C. Nowadays, there are numerous examples of silver incorporated in everyday objects to provide antiviral activity. It can be found in water purification systems, food packaging, toys and infant pacifiers, synthetic fabrics and consumer products, e.g., washing machines or telephone handles. It has also been widely employed in healthcare and medical devices. Both the Environmental Protection Agency and the World Health Organization (WHO) regard silver as safe.

Silver can cause argyria (irreversible skin discoloration) but only by the ingestion of gram quantities of silver over several years or by the administration of high concentrations to ill individuals. Even though silver is commonly used biocide there have been very few studies on its use as a virucide. Silver ions have been described as having the highest level of antimicrobial activity of all the heavy metals and many mechanisms for silver’s bactericidal activity have been proposed and some interactions of viruses with silver would be similar to that of bacteria. However, as opposed to bacteria, viruses do not possess the metabolic activity and rely on host cell processes, therefore it is suggested that the inactivation mechanism is the one that does not require a metabolic process, e.g. the immobilization of the virus to a surface, the blocking or destruction of host-cell receptors, or the inactivation of the nucleic acid within the viral capsid. It has been shown that viruses that contain sulphhydril termini in their molecular structure, may bind silver. This can further affect their replication cycle. In addition, similarly to copper, silver is believed to be subject to site-specific Fenton mechanism, in which it binds to a biological molecule and undergoes reduction by superoxide radicals or other reductants and then reoxidized by hydrogen peroxide. These cyclic redox reactions result in radicals caused damage to the neighbouring target site of the molecule.

There are only a few reports on antiviral properties of silver and silver-containing compounds on specific viruses. For example, silver, as Ag$_2$O$_3$ in concentration of 1–20 ppm, was shown to be effective against HIV-1. Kadar et al. reported the antiviral effect of silver-peroxide solutions (concentrations 0.025–0.1%) on Papovavirus SV-40, Herpes simplex type 1, Vaccinia virus, Adenovirus and Poliovirus. Silver sulfadiazine was shown to cause direct inactivation of herpes simplex and vesicular stomatitis viruses (SVSs). The virucidal effects of silver depend heavily on chemistry, concentration and synergistic effects with other compounds in the surrounding environment. Also, the effects will differ between viruses including different types of the same virus. In terms of silver containing antiviral surfaces or materials, the reports are even fewer. Han et al. tested metal catalysts: Ag/Al$_2$O$_3$ and Cu/Al$_2$O$_3$, pressed into wafers, which have shown inactivating properties against SARS-coronavirus and baculovirus. After exposure of virus solutions to the Ag/Al$_2$O$_3$ and Cu/Al$_2$O$_3$ surfaces for 5 and 20 min, the infectivity of both viruses dropped down to a very low and undetectable level, as measured by TCID$_{50}$ assay. The research states that contact with metals destroys the replication and propagation abilities of the viruses and the inactivation of viruses is dependent on the ability of the metal catalysts to interact with oxygen in the air.

Bright et al. reported antiviral properties of zeolite (sodium aluminosilicate) powders containing Ag and Cu ions in varied ratios. First, they tested the zeolite as a suspension in phosphate-buffered saline (PBS), showing that the 3.5% Ag/6.5% Cu ion combination was the most efficacious against human coronavirus 229E and feline infectious peritonitis virus (FIPV). Next, they incorporated the most effective Ag/Cu-zeolite formulation into plastic at either 5 or 10% (wt/wt). After 24 h of exposure, they observed a significant reduction in coronavirus 229E (tested by TCID$_{50}$ assay), with a 1.84-log$_{10}$ and a 1.77-log$_{10}$ reduction for the 5 and 10% (wt/wt) concentrations, respectively. The reductions for the FIPV were even greater, of 3.84-log$_{10}$ on the 5% and 5.05-log$_{10}$ reduction on the 10% Ag/Cu-zeolite plastics.

Polymers and biopolymers

The antiviral properties of surfaces could be directly linked to the mechanisms and chemistries viruses use to attach and enter a host cell. Viral entry often occurs via the attachment of the viral spike protein to a receptor on a host cell surface, followed by low pH fusion and replication. As an example, HIV uses a complex series of steps to deliver its genome into the host cell. Firstly, it binds to a target cell. This attachment can be mediated through various cell attachment factors such as relatively nonspecific negatively charged cell-surface heparan sulfate proteoglycans, or through interactions with more specific cell-surface integrins. These binding steps bring the HIV envelope into close proximity with the CD4 receptor at the cell surface. The cell infection depends then on the interaction of the viral envelope glycoprotein gp120 with the CD4. Polyanionic substances, such as polysulfates and polyphosphate, have been shown to inhibit this process. Examples of such polyanionic substances including dextran sulfate, polyvinyl alcohol sulfate and naphthalene sulfonate are given by De Clercq. The types of polymers presenting antiviral and antibacterial properties are varied and range from natural biopolymers with intrinsic antiviral/virucidal properties to engineered thermoplastic elastomers. Antimicrobial polymers, polysaccharides like glutamine and glucosan, contain a sulfonic functional group, which works effectively against viral adhesion. Hydrogels are often deemed biocompatible and can be used to incorporate antiviral inorganic materials or could be combined with synthetic polymers. Synthetic polymers, like linear macromolecules such as polyethyleneimine (PEI), poly-L-lysine (PLL), diethyleniminoethyl-dextran (DEAE-dextran), and branched polymers such as poly(amidoamine) (PAMAM) dendrimers and polyphenol ether can be used as coatings and
their antiviral mechanism of action relies on an ionic chain. In the development of antiviral strategies, nature itself can be inspirational and naturally derived biopolymers could be considered as an attractive option as a parallel to the discovery of naturally occurring antibiotics such as antimicrobial peptides (AMPs). Natural products could provide versatile and tuneable designs against a wide spectrum of viruses. Some natural bacteriostatic materials exhibit also antiviral properties and can be indirectly used as antiviral/virucidal drug carriers93–96.

**Synthetic polymers and coatings**

**Polyethylenimines (PEI).** Haldar et al.90,97 conducted studies of the bactericidal and virucidal properties of several branched and linear N,N-dodecyl methyl-polyethylenimines and other hydrophobic PEI derivatives, prepared as painted coatings on glass slides. The materials were challenged with two common pathogenic bacteria, Staphylococcus aureus and Escherichia coli and two different strains of influenza A virus. The study showed that all of the PEI derivatives presented biocidal properties, with some variances in virucidal effectiveness dependent on size (molecular-weight) of PEIs90,98. Differently charged derivatives of linear PEI (zwitter-ionic, anionic and neutral) were also tested90. The neutral PEI showed no virucidal properties, while zwitter-ionic PEI was as effective as a corresponding cationic polymer and able to inactivate 100% of the virus within minutes. In contrast, the anionic polymer was only partially virucidal in a 30 min experiment, however, longer exposure times of 2 h showed a steady yet substantial increase from 66% to nearly 90%. They postulated that the virucidal mechanism of action is destroying the virus lipid envelope through “tentacle” fragments of the polyanionic chains90. Their results suggested that both positively and negatively charged sites of the PEI are able to attack the viral membrane, with the negative charge having a predominant effect. The study was expanded to other viruses including human and avian influenza viruses with wild-type strains and drug-resistant ones. They showed that the N,N-dodecyl, methyl-PEI painted glass slides exhibit a 100% virucidal efficiency against influenza A virus, regardless of the strain99. The practical application of the N,N-dodecyl, methyl-PEI in a prophylactic modality (as a coating on latex condoms) was demonstrated by Larson et al.100, who showed its effectiveness to inactivate two strains of HSV. Klibanov et al. have developed a series of water-insoluble, hydrophobic polycations PEI-based polymeric material coatings, with biocidal properties98,101,102. Surfaces can be either covalently derivatised102 with the polymer or simply painted98 with it.

**Sulfonated block copolymers—TESET.** Low pH conditions can have a sterilizing effect. Therefore, materials able to create an acidic environment can present biocidal properties. Peddinti et al.103 demonstrated this for a class of charged hydrophilic multiblock polymers containing a midblock functionalised with sulfonic acid groups. The polymer was a thermoplastic elastomer poly[tertbutylstyrene- b-(ethylene-alt-propylene)-b-(styrnesulfonate)-b-(ethylene-alt-propylene)-b-tert-butylystyrene] (TESET). They examined TESETs with a different midblock sulfonation degree, in a range from 26 to 52 mol%, showing that the polymers are able to kill Gram-positive bacteria (MRSA) and Gram-negative bacteria (A. baumannii, K. pneumoniae and E. coli), including some drug-resistant strains in <5 min. Irrespective of the degree of sulfonation, the TESETS were shown to be effective in the inactivation of enveloped viruses—VSV and Influenza A. However, in the case of a non-enveloped human adenovirus-5 (HAd-5) the 26% sulfonated TESET was completely ineffective and the 56% sulfonated TESET showed antiviral properties but less than for enveloped species. They postulate that the TESET mechanism of action is the ability to rapidly and significantly lower the pH of pathogen suspensions leading to degradation of the protective outer membrane, protein denaturation and inactivation. The biocidal properties are activated by the simple presence of water and their effectiveness reduces upon repeated exposure to the suspensions. However, it was demonstrated, the materials can be fully rejuvenated by exposure to acid and removal of cations complexed with sulfonic acid groups.

**Photodynamic polymers.** Peddinti et al.104 investigated, also, polymeric materials with photoactivated biocidal properties. They studied a semi crystalline thermoplastic elastomer—multiblock copolymer composed of high-density polyethylene blocks and poly(ethylene-co-a-octene) blocks (OBC26), to which they incorporated ~1 wt% of zinc-tetra(4-N-methylpyridyl)porphine (ZnTMPyP4+). The ZnTMPyP4+ moiety acts as a photosensitiser and, when activated by photon energy, it exchanges energy with molecular oxygen generating high-energy single oxygen or ROS. Since the ROS mechanism of action is non-specific, the approach could be applicable to a wide range of pathogens. The researchers104 examined six Gram-positive and Gram-negative bacteria (S. aureus, vancomycin-resistant E. faecium, E. coli, A. baumannii and K. pneumonia) and two viruses, including VSV, and HAD-5 and demonstrated the photoreactive polymer being highly effective against those pathogens. OBC26 without ZnTMPyP4+ moiety was used as a control and showed no biocidal activity.

**UV-enhanced antiviral polycations.** Wang et al.92 investigated a series of poly(phenylene ethynylene)- based cationic conjugated polyelectrolytes (CPE) and oligo-phenylene ethynlenes (OPE), shown previously to exert significant photoinducible antimicrobial activity105–107. They tested antiviral properties of these materials on two model non-enveloped bacteriophages—T4 and MS2. Most of the employed compounds exhibited high inactivation ability against MS2 phage and moderate inactivation against T4 in dark conditions. This ability is believed to be attributed to the cationic CPEs and OPEs binding to the slightly negatively charged surfaces of both bacteriophages through electrostatic interactions leading to the destruction of viral structures and/or inhibition of infection pathways. Under irradiation with UV light significant antiviral ability was observed for all the tested compounds against both bacteriophages. In some cases, especially with T4, the virucidal activity was significantly enhanced in comparison to the experiments in the dark. Exposure to UV light results in the generation of ROS causing photochemically induced damage of the phage capsid protein.

**Hydrogels.** Hydrogels are three-dimensional networks of hydrophilic polymers, with the ability to take up a large amount of water while maintaining the 3D structure108. They can be created from a range of different polymers but the crosslinking of water-soluble polymers such as poly(ethylene glycol), polyacrylamide, poly(vinyl alcohol), etc. seems most convenient and attractive108–110. Some natural polymers such as agar or gelatine have been also utilised in hydrogels111,112. Due to their non-toxicity and biocompatibility hydrogels are already used in many biomedical applications113 including contact lenses, wound dressings, coatings on medical devices or hygiene products. The use of hydrogels goes above the biomedical applications as they are widely present in different everyday products including, e.g., food packaging or paints. In some cases, the same polymers that constitute hydrogel coating, can also exert antimicrobial/antiviral properties97 or can be engineered to incorporate antimicrobial/antiviral moieties such as nanoparticles57,77, surfactants or drugs. As an example,
monocarpin is a highly efficient antiviral monoglyceride, shown to be effective against enveloped viruses, including VSV, HSV, visna virus and HIV. Thorgeirsdottir et al. tested the antiviral activity on a range of monocarpin-containing, polyacrylic acid based, hydrogels. Formulations with different ratios of propylene glycol (co-solvent) in either 5 or 7.5% concentration with 0.2 or 0.4% polysorbate 40 (surfactant), were assessed. In all cases, the gels showed up to 1 log10 greater reduction in viral titer (6.0 log10) of HSV than the corresponding solutions of the same co-solvent and surfactant ratios (5.0 log10). The researchers postulate that the hydrogel structure aids the antiviral properties of monocarpin by forming bonds with the viral envelope leading to increased contact between the virucide and the virus.

Meng et al. presented an interesting approach of incorporating catechol, a natural adhesive moiety found in mussel proteins into polyacrylamide-based, containing 10% dopamine methacrylamide, hydrogel. Upon rehydration in physiological pH catechol undergoes autooxidation, generating hydrogen peroxide (H2O2), thus exerting biocidal properties. It is suggested that the biocidal mechanism of H2O2 comes from its oxidizing properties and reactivity with bacterial or viral biomolecules such as proteins, lipids, nucleic acids, etc. affecting their structure/function. The researchers achieved a continuous peroxide generation of 1-5 mM of H2O2, over a period 4 days, This concentration proved virucidal, which was demonstrated on non-enveloped porcine parvovirus (PPV) and enveloped bovine viral diarrhea virus (BVDV). After 12 h of incubation with the viruses, the hydrogels reduced the viral titer of PPV by nearly log10. For the enveloped BVDV the infectivity reduction was even greater of 4-5 log10.

Hydrogels incorporating “non-drugs” molecules, e.g., nanoparticles find widespread biomedical applications. Ag or Au being most commonly used and are well known for their high biocidal activity. Nanoparticles of metal-oxides, particularly TiO2, ZnO, and CeO2, have been also found to display biocidal properties.

**Biopolymers**

Biopolymers are a class of naturally derived materials. Due to their biocompatibility and intrinsic antimicrobial properties several biopolymers have been used, for example, as an attractive material in food packaging. While their antimicrobial properties have been documented, little attention has been given to their antiviral properties.

**Chitosans.** Chitosan (poly-β-1,4-glucosamine) is an aminated polysaccharide, formed by acid alkylation of crustaceans chitin (poly-β-1,4)-N-acetyl-d-glucosamine. Chitosan has been extensively studied for its antimicrobial properties and potential uses in the pharmaceutical, cosmetics, agricultural and food industries. Chitosan has been extensively studied for its antimicrobial properties and potential uses in the pharmaceutical, cosmetics, agricultural and food industries. There are also reports of antiviral properties of chitosan and its modified forms, which have been examined on a range of animal and plant viruses. For example, in the report by Ait et al. chitosan extracted from the larvae of *Musca domestica* exhibited inhibition of infectivity of two baculoviruses, *graphica californica* multiple nucleopolyhedrovirus (AcMNPV) and bomyx mori nucleopolyhedrovirus (BmNPV). In another study by Davis et al. 53 kDa chitosan showed a reduction in the growth of surrogate enteric viruses, *feline calicivirus FCV-F9* and murine norovirus MNV-1. derived 3,6-O-sulfated chitosan sulfated chitosan was shown, by Gao et al., to exert broad anti-human papillomavirus (HPV) activities in vitro, by targeting the viral capsid protein and by disrupting P13K/Akt/mTOR pathway, known to play a role in viral replication inside a host cell. Li et al. showed that sialyllactose (SL)-incorporated chitosan-based materials are able to inhibit viral adhesion of influenza virus to a host cell. The SL-chitosan binds to hemagglutinin protein on the surface of an influenza particle, which is responsible for viral attachment to the host cell surface via binding with surface glycoligands such as SL. Guo et al. reported on antiviral mechanism of a combined biological agent consisting of chitosan oligosaccharide and cytosine phosphorothioate (CytPM-COS). CytPM-COS effectively inhibited tobacco mosaic virus (TMV), suppressed viral RNA and capsid protein accumulation and triggered production of ROS and induced upregulation of various defence responsive genes in the tobacco plants. Davydova et al. reported that low molecular weight derivatives of chitosan especially with decreasing degree of polymerisation showed antiviral property against TMV by inhibiting the development of necrosis caused by the virus.

**Fucoidans.** Fucoidan is an anionic sulfated polysaccharide extracted from brown marine algae. It was recently demonstrated that sulfated polysaccharides exhibit antiviral activities both in vivo and in vitro and, since they have low cytotoxicity in comparison to other antiviral drugs, there is interest in their use in drug and gene delivery systems and wound and burn healing formulations. As reported by Ponce et al., fucoidans extracted from *Adenocystis utricularis* show inhibitory activity against the replication of several enveloped viruses including human immunodeficiency and human cytomegalovirus.

**SP-303.** Phenolic biopolymer SP-303 is a 2 kDa plant flavonoid, found to exhibit antiviral activity against two strains of HSV type 1 and type 2. As postulated by Bernard et al., the antiviral mechanism of SP-303 comes from the inhibition of virus penetration into cells by binding directly to viral or cell membranes or to both. It was also tested by Wyde et al., in vivo in cotton rats (*Sigmodon hispidus*) for antiviral activity against respiratory syncytial (RSV) and parainfluenza type 3 (PIV3) viruses.

**Glycopolymers.** Hidari et al. synthesized and characterised glycopolymers with terminal 2,6-sialic acid on lactosamine repeats, which they tested on a range of viruses. They showed that the glycopolymers exerted viral inhibition properties against human and swine influenza viruses. The effectiveness of the inhibition was dependant on and growing with the number of lactosamine repeats on the glycopolymers. Similar to the SL-chitosan described earlier, the lactosamine-glycopolymers bind to haemagglutinin protein acting as competitive inhibitors that prevent virus entry to the host cell, which seem to be their predominant antiviral mechanism. Avian and equine viruses were also tested, yet no inhibition was observed by any of the lactosamine-glycopolymers. Sequence analysis and molecular modelling highlighted that amino acid substitution of haemagglutinin along with core carbohydrate and sialyl linkages on the receptor glycoconjugate have an impact on its antiviral properties.

**Mucins.** Mucins are highly glycosylated polymeric proteins. They are the main constituents of mucus—a porous biopolymer matrix produced by epithelial tissues in most animals, which serves as the first line of defence against many pathogens, including viruses. Lieleig et al. showed the ability of isolated porcine gastric mucin to block human pathogenic viruses like human papillomavirus HPV-16, Merkel cell polyomavirus (MCV), and a strain of influenza A. In this study, a mucin layer was applied on three different cell lines, which were next incubated with the viruses. It was demonstrated that the mucin isolates were highly effective in blocking the viruses from infecting the cells. The researchers speculated that the antiviral mechanism of the mucins lies in the...
trapping of virus particles in the mucin matrix, caused by multiple low-affinity bonds between the mucin sugar groups and the virus capsids. The efficiency of mucin shielding layer towards viral infection was also shown to be dependent on the mucin concentration, increasing with increased mucin content.

**Antimicrobial peptides.** AMPs are short, usually positively charged oligopeptides with diverse structures and functions. They play a fundamental role in the innate immune system response to injuries and infections. AMPs are expressed in a wide variety of tissues including skin, eyes, oral cavity, ears, airway, lung, female reproductive tract, cervical-vaginal fluid, intestines, and urinary tract.

Defensins and cathelicidin LL37, derived from humans have been extensively studied for its antibacterial effects but more recently anti-viral properties are being elucidated. The Anti-microbial Peptide Database (APD) contains more than 3000 AMPs, among which are 189 AMPs with antiviral activities. There are reports of incorporating AMPs into other materials, which have been assessed for their antimicrobial properties. However, methods to graft or adsorb AMPs on the surface to fabricate, for example, medically useful products are required to fully realise their potential.

Yu et al. studied the effect of human cathelicidin LL-37 and engineered LL-37 AMPs on a recombinant virus, pseudotyped with Ebola virus glycoprotein, and on a wild-type Ebola virus. The peptides inhibited the infection by targeting the endosomal cell-entry step by impairing cathepsin B-mediated processing of Ebola virus glycoprotein. AMPs synthesized by incorporating D-amino acids cannot be degraded by intracellular enzymes and exhibit a higher degree of activity in the early stages of viral infection rather than during its replication stage. In different reports, Barlow et al. showed antiviral properties of the cathelicidin LL-37 against influenza virus and Currie et al. against the respiratory syncytial virus. Cathelicidin-derived peptides (GF-17 and BMAP-18) have also shown efficacy against Zika virus and inhibition is through direct inactivation of the virus by the interruption and damage to the viral membrane but also, indirectly, via the inhibition of interferon signalling pathway. Defensins are small cysteine-rich peptides, well known for their host defence and immune signalling activities. They have been also recognised for their antiviral properties, reported in a number of papers. In one example, neutrophil α-defensins have been shown, by Daher et al., to have a direct effect on several enveloped viruses including influenza A, herpes simplex viruses 1 and 2, VSV and cytomegalovirus. Non-enveloped viruses, echovirus type 11 and reovirus type 3 have been also tested but seemed unaffected by the action of defensins. The difference in susceptibility to inactivation between enveloped and naked viruses suggest the lipid envelope as an interaction site between the peptide and the virus. It was also noted that the effect of defensins on viral infection appears to be specific to the type of defensin peptide, virus and a target host cell. In addition to the direct effect on the virus and the cell, defensins act as immune modulators that may play a role in viral transmission.

Lactoferrin is an iron-binding glycoprotein, which has exhibited antiviral activity against a wide range of human and animal viruses (both DNA and RNA), including hepatitis C (HCV) through the direct binding of the peptide to the HCV particles, HSV through inhibition of virus-host interactions, or through inhibition of human immunodeficiency virus (HIV) replication in host cell. To other reported lactoferrin-affected viruses belong poliovirus, calicivirus, rotavirus and SARS coronavirus. Lactoferrin prevents entry of virus in the host cell and its antiviral properties vary among viruses, where it may bind either directly to the virus particle or to the host cell receptor or coreceptor.

Hepcidin, a 25-amino-acid antimicrobial peptide (LEAP-1), is predominately expressed by liver hepatocytes and is the principal regulator of iron absorption. The effects of hepcidin on the pathogenesis of viral infections is not well understood; however, expression of hepcidin has been reported following a number of human and murine viral infections.

Wang et al. reported elevated hepcidin expression in chronic HBV and HCV patients, in contrast to acute hepatitis infections. It was postulated that hepcidin expression may be upregulated at later stages of infection when iron levels are elevated. Increased hepcidin levels in HIV-1 positive plasma donors of both acute as well as transitioning to chronic infections were reported. HIV replication is iron-dependent and the hepcidin-induced sequestration of iron in cells such as lymphocytes and macrophages is a highly favourable condition for HIV pathogenesis. Rajanbabu et al. studied the antiviral effects of tilapia hepcidin (TH1)-1, against infectious pancreatic necrosis virus (IPNV) in Chinook salmon embryo (CHSE-214) cells. The presence of TH1-1 decreased the number of plaques formed by the cytopathic effect of IPNV in the CHSE-214 cells. In addition, the presence of hepcidine showed, further, modulation of interleukin, annexin and other viral-responsive gene expressions and, hence, demonstrated immunomodulatory functions.

There is a requirement to improve the delivery and stability of these peptides whilst retaining their functionality and a few strategies have been proposed. As an example, in the study by Zhang et al., amphiphilic α-helical peptide p41, a positively charged analogue of C5a peptide derived from the HCV protein, was incorporated into anionic poly (amino acid)-based block copolymers via electrostatic coupling. In vitro antiviral activity of this nanoformulation against both HCV and HIV were retained. In vivo incorporation was also able to decrease the viral load in mice transplanted with human lymphocytes and HIV-1-infected. A subsequent study of using the same nanoformulation in a galactosylated form, was tested as liver-specific delivery system. It exhibited prominent advantages to prevent HCV association with lipid droplets and was able to suppress the intracellular expression of HCV proteins in an in vitro assessment. In addition, in vivo assessment indicated preferential accumulation in the liver of the tested animals. Another strategy to prevent protease degradation is to substitute L amino acids with D-amino acids. Jackman et al. engineered an α-helical peptide AH-D (D-enantiomer amino acid), that effectively targeted the highly curved membrane of small enveloped viruses such as Zika by rupturing the virus' lipid envelope much quicker than the L-isomer equivalent, it was also effective against other neurotropic viruses in vitro such as Dengue and Chikungunya which have a similar structure to Zika. The peptide showed promise in tackling Zika virus-infected mice, reducing viral load and inflammation in the brain. Subsequent study by Camargoes et al. showed that AH-D peptide, when administered to mice during the early stages of pregnancy, prevented Zika replication and the death of offspring.

**Graphene**

Graphene is a two-dimensional layer of sp²-hybridised carbon atoms, arranged in a hexagonal lattice, that has demonstrated extraordinary properties, such as high electrical and thermal conductivity. It is being used in commercial products, like nanocomposites and electronics. Graphene can be produced in different forms, classically as a single-layer sheet over a substrate, or as few-layer flakes with finite lateral dimensions, typically found in a powder/liquid dispersion form. This latter nanoscale particulate...
Silver nanoparticles (AgNPs) are known for their excellent antiviral properties. However, differently to silver in its metal bulk form, using free AgNPs, due to their size and properties, poses several drawbacks such as particle aggregation, cytotoxicity or genotoxic damage by inhalation or ingestion into human body in larger quantities. Park et al.189 found that by decorating silica with ≈30 nm AgNPs an efficient hybrid system can be developed, which showed clear dose-dependent effects against influenza A virus. They suggested that the major antiviral mechanism of this material was its interaction with the components present on the viral membrane. Martinez-Abad et al.189 studied the incorporation of 0.001–1.0 wt.% silver ions into polylactide (PLA) films by a solvent casting technique, with the aim to create a potential antimicrobial food packaging material. The PLA-silver films showed antibacterial and antiviral activity in vitro, with increasing effects at higher silver concentrations when tested against Salmonella and feline calcivirus. Mori et al.191 studied the antiviral effect of silver nanoparticles (AgNPs)-chitosan composite against influenza A virus. The antiviral activity was evaluated from the decreased TCID50 ratio of viral suspensions between composite-treated samples and controls. Composites with different sizes of AgNPs were tested and stronger antiviral activity was observed for smaller particles for comparable concentrations of incorporated AgNPs. For all the tested particle sizes, the antiviral activity increased with increasing concentration of the particles in the composite. In another study with AgNPs composite192, chitin nanofibers sheets (CNFs) with immobilized AgNPs also showed antiviral efficacy against H1N1 influenza A virus.

Tyo et al.195 tested the antiviral properties of a multi-layered nanoparticle (NP)-electrospun fibre (EF) with Gristhins (GRFT) composite. The composites were fabricated from polycaprolactone (PCL) fibres surrounding polylethylene oxide (PEO) fibres that incorporated methoxy poly(ethylene glycol)-b-poly(lactide-co-glycolide) (mPEG-PLGA) GRFT NPs. The efficacy of GRFT NP-EFs was assessed using human immunodeficiency virus (HIV-1) pseudovirus assays, demonstrating complete in vitro protection against HIV-1 infection. In addition, sustained-release NP-EFs, administered 24 h prior to infection, prevented the lethal effect of herpes simplex virus 2 (HSV-2) in a murine model. Mommatapoj et al.194 studied TiO2-modified hydroxyapatite composite and showed its strong activity against influenza A virus. They speculated this composite as a promising material for use in antimicrobial filtration materials in e.g. medical face masks. In another study with titanium dioxide, Amirkan et al.195 created nano-biocomposite material consisted of titanium dioxide nanoparticles covered with polylysine (PL) and DNA/peptide nucleic acid (PNA) and showed its inhibitory effects on influenza A virus. In a study by Grover et al.196, perhydrolase (AcT) was immobilized onto multi-walled carbon nanotubes (MWNTs) and then incorporated into latex-based paints to form catalytic coatings. The material showed >4 log10 reduction in the titer of influenza virus (X-31). Even though the test lacked testing on a broad virus spectrum, it shows the potential of perhydrolase acyl donor substrates, such as propylene glycol diacetate (PGD), glyceryl triacetate or ethyl acetate, to engineer antiviral materials with potential use in antiviral paints in hospital to reduce the propagation of infection.

Overall, the proposed mechanisms of action can be categorised into six basic types: (a) ionic surfaces that degrade the RNA/DNA such as metals and polylethlenimines; (b) Photosensitizing materials producing ROS; (c) adsorbing surfaces causing membrane disruption through dehydration; (d) sharp nanostructured surfaces causing membrane disruption through puncture such as graphene; (e) controlled release of virucides including AMPs in hydrogel and (f) inactivating surfaces such as biopolymers binding to the membrane or capsid proteins. In Table 3, we provide a summary of materials with antiviral properties with, where reported, the mechanisms of action and persistence.

**Composites**

Composites offer an attractive strategy in fabricating useful materials with antiviral properties. Combining different classes of materials or incorporating antiviral moieties, e.g., metals, into easily moulded materials such as polymers allows tailoring the required properties of materials.

---

**References**

1. Park et al. 189
2. Martinez-Abad et al. 189
3. Mori et al. 191
4. Tyo et al. 195
5. Mommatapoj et al. 194
6. Amirkan et al. 195
7. Grover et al. 196
8. ---
Table 3 Summary of literature on materials with antiviral properties with reported mechanism of action and persistence.

| Material/surface                  | Proposed mechanism of action                                                                 | Target viruses                        | Persistence | Ref. |
|-----------------------------------|---------------------------------------------------------------------------------------------|---------------------------------------|-------------|------|
| Metals                            |                                              |                                       |             |      |
| Cu                               | Possibly viral replication inhibited by copper-linked (cross linking) RNA damage             | Influenza A                           | 7 min       | 56   |
| Cu(II) (metal ions dissolved in solution with the addition of peroxide) | Nucleic acid damage by crosslinking                                                        | Bacteriophage ɸX174, bacteriophage T7, bacteriophages ɸ6, Junin herpes simplex | <10 min, 22 min, 33 min, 8 min | 56   |
| Fe(III) (metal ions dissolved in solution with the addition of peroxide) | Nucleic acid damage by crosslinking                                                        | Bacteriophage ɸX174, bacteriophage T7, bacteriophages ɸ6, Junin herpes simplex | >60 min, <10 min, 24 min, >60 min | 56   |
| Cu                               | Damage to the viral capsid, RNA degradation                                                 | Murine norovirus MNV-1                | <5 min (dry) | 57,58 |
| Cu(89%)-Ni alloy                  | Damage to the viral capsid, RNA degradation                                                 | Human coronavirus 229E                | <5 min (dry) | 59   |
| Brass (70% Cu, 30% Zn)            | Damage to the viral capsid, RNA degradation                                                 | Human norovirus MNV-1                 | <10 min     | 57,58 |
| Stainless steel                   |                                              |                                       | ≥120 min (wet) | 59   |
| Cu/Al₂O₃                          | Charged “tentacle” fragments of polyanionic chains interacting and destroying viral membrane| Human and avian influenza A           | 5-120 min   | 90,97,99 |
| Ag/Al₂O₃                          |                                              | Herpes simplex virus                  | <15 min     | 100  |
| Ag/Cu-zeolite plastics            |                                              | Coronavirus 229E feline infectious peritonitis virus (FIPV) | <24 h       | 82   |
| Polymers                          |                                              |                                       |             |      |
| Polyethylenimines (PEI)           | Sulfonic acid promoting local acidic environment, leading to stress on the outer membrane | Vesicular stomatitis virus (VSV) human adenovirus 5 (HAd-5) | <5 min (TESET26 and TESET56) | 103  |
| TESET (Midblock-sulfonated block co-polymer) | Generation of high-energy singlet oxygen (\*O₂), or other reactive radical species, inducing inactivation by non-specific reactions with viral components | Vesicular stomatitis virus (VSV) human adenovirus 5 (HAd-5) | <60 min | 104  |
| Multiblock co-polymer OBC26 with ZnTMPyP₄⁺ | Generation of ROS causing photochemically induced damage of the phage capsid protein | Bacteriophage T4, bacteriophage MS2 | <5 min | 92 |
| Pol(phenylene ethynylene)-based cationic polyelectrolytes (CPE) and oligo-phenylene ethynlenes (OPE) Polycrylic hydrogel containing monacarpin | Inhibition of viral replication | Herpes simplex virus | - | 115 |
| Catechol containing polyacrylamide-based hydrogel, with 10% dopamine methacrylamide Biopolymers Chitosan | Peroxide releasing, oxidative stress, reactivity with viral structural compounds | Porcine parvovirus (PPV) bovine viral diarhoea virus (BVDV) | <12 h | 116 |
| Biopolymers                       |                                              |                                       |             |      |
| Chitosan                          |                                              |                                       |             |      |
| 3,6-O-sulfated chitosan (36 s)    | Blocking of adsorption to host cell via direct binding to the viral capsid proteins         | Autographa californica multiple nucleopolyhedrovirus (AcMNPV) bombyx mori nucleopolyhedrovirus (BmNPV) feline calicivirus (FCV-F9), murine norovirus (MNV-1) Human papillomavirus (HPV) | | 133,134,135,136 |
| SL-chitosan                       | Blocking of adsorption to host cell via direct binding to capsid protein (hemmaglutin)     | Influenza                             |             | 136  |
| Cytosinpeptidemycin- chitosan oligosaccharide (CyTPM-COS) | Induced generation of ROS; duced upregulation of various defence responsive genes | Human norovirus MNV-1                 | <10 min     | 137  |
| Low molecular weight (LM) chitosans |                                              | Tobacco mosaic virus (TMV)            |             | 138  |
| Material/surface                      | Proposed mechanism of action                                                                 | Target viruses                                                                 | Persistence | Ref. |
|--------------------------------------|---------------------------------------------------------------------------------------------|--------------------------------------------------------------------------------|-------------|------|
| Fucoidan                             | Inhibition of viral replication                                                              | Human immunodeficiency virus (HIV)                                              |             | 139  |
| SP-303                               | Inhibition of virus penetration into cells by binding directly to virus or cell membranes or both | Herpes simplex viruses type 1 (including a thymidine-kinase-deficient strain) and type 2 respiratory syncytial virus (RSV) parainfluenza type 3 viruses (PIV3) |             | 142  |
| Glycopolymers with sialic acid on lactosamine repeats | Block adsorption to host cell via direct binding to capsid protein (hemmaglutinin)            | Human influenza viruses Swine influenza virus                                    |             | 144  |
| Mucin                                | Entrapment of viral particles in mucin polymeric matrix                                        | Human papillomavirus HPV-16 Merkel cell polyomavirus (MCV) influenza A          |             | 145  |
| *Antimicrobial peptides*              |                                              |                                                                              |             |      |
| Cathelicidin LL-37                   | Preventing endosomal cell-entry by impairing cathepsin B-mediated processing of viral glycoprotein | West Africa Ebola virus (EBOV) Respiratory syncytial virus Zika virus (ZIKV)      |             | 152  |
| Cathelicidin GF-17 / cathelicidin BMAP-18 | Damage and interruption of viral membrane integrity; inhibition of interferon signalling pathway | Influenza A herpes simplex virus 1 & 2 Vesicular stomatitis virus cytomegalovirus Including hepatitis C (HCV), poliovirus feline calicivirus rotavirus136 SARS coronavirus.137 |             | 153  |
| Defensin                             | Direct interactions with the viral envelope; immunomodulatory roles                           | Influenza A herpess simplex virus 1 & 2                               |             | 154  |
| Lactoferrin                          | Direct interactions with the viral envelope; competitive binding to the host cell receptor or coreceptor | Influenza A Vesicular stomatitis virus cytomegalovirus Including hepatitis C (HCV), poliovirus feline calicivirus rotavirus136 SARS coronavirus.137 |             | 155  |
| Hepcidin                             | Sequestering iron from pathogens                                                             | Hepatitis B Hepatitis C HIV Infectious pancreatic necrosis virus (IPNV)         |             | 156  |
| *Graphene*                           | Negative charge leads to electrostatic interaction with virions so that nanoscale sharp edges destroy and inactivate virus | Pseudorabies virus porcine epidemic diarrhoea virus |             | 157  |
| Graphene oxide (GO) reduced graphene oxide (rGO) | Particles bind enveloped viruses during the adhesion process and inhibit virus by preventing viral adhesion to the host cell | African swine fever virus |             | 158  |
| Reduced graphene oxide functionalised with either dendritic polyglycerol or linear polyglycerol azide functional groups | Sulfonated moieties capture virions allowing virus destruction using photothermal effect through irradiation with near-infrared light | Herpes simplex virus 1 |             | 159  |
| Graphene flakes with polyglycerol sulfate and fatty amine functional groups | Electrostatic interactions trigger binding of graphene to virus, whilst secondary hydrophobic interactions induce antiviral activity | Herpes simplex virus 1 |             | 160  |
| Composites                           | Interaction with components present on viral membrane                                          | Influenza A |             | 161  |
| Silica/30 nm silver particles        |                                              | Feline calicivirus influenza A influenza A                                        |             | 162  |
| Silver-infused polylactide films     |                                              | Human immunodeficiency virus 1 herpes simplex virus 2                            |             | 163  |
| silver nanoparticles/chitosan sheets (CNFSs) |                                              | Influenza A influenza A influenza A                                            |             | 164  |
| Griffithsin nanoparticle/fibre composites |                                              | Human immunodeficiency virus influenza A influenza A influenza A influenza A influenza A influenza A |             | 165  |
| TiO$_2$-modified hydroxyapatite      |                                              | Influenza X-31                                                                  |             | 166  |
| TiO$_2$-polylysine nanoparticles/DNA/ peptide nucleic acid |                                              |                                                                              |             | 167  |
| Perhydrolase/multi-walled carbon nanotubes |                                              |                                                                              |             | 168  |
Applications of antiviral materials and coatings

Antiviral materials and coatings have very broad applications, from antiviral food packaging and food contact surfaces for controlling human enteric viruses\(^{197}\), to health products for prevention of sexually transmitted infections to PPE in the healthcare sector. The applications dictate the different properties of materials. For example, in food production and retail, the antiviral materials cannot be toxic\(^{197}\). Materials used in public transport need to be stable, durable and non-flammable. Here, we give an overview of practical applications of antiviral materials and coatings which are summarised in Table 4. Example applications in the context of a hospital setting are illustrated in Fig. 3.

PPE and soft surfaces. PPE is clearly a prime area of importance, to protect people from the risk of contact infection. As PPE includes facial masks or visors, protective suits, spill gowns, gloves, boot covers, goggles, etc., it requires compatibility with a wide range of materials from woven fabrics, used in masks, to disposable aprons. However, generally the materials should be non-toxic and skin friendly.

Masks and respirators are one of main application of antiviral materials since the primary transmission carriers for respiratory viruses are droplets and aerosols, which are released through a cough, sneeze or when speaking and even breathing. During the SARS-CoV-2 pandemic, face masks and respirators are often mandated in public areas to limit the spread of infections\(^{198}\). The conventional type of face masks and respirators composed of woven or non-woven fabric block the virus by filtration of aerosols and droplets. However, the virus persists on the surface\(^{199}\) which leads to a risk due to incorrect wearing of masks or their reuse. Research has focused on developing self-cleaning masks by using different materials and technologies\(^{200}\), especially nano-based materials and techniques, some of which are already available or close to the market. One popular strategy is incorporating antiviral nanoparticles into fibrous membranes.

Table 4 Examples of published antiviral materials and coatings with the potential use in PPE, public areas and other applications.

| Application type               | Material                                                                 | Note                                                                                                                                 |
|-------------------------------|--------------------------------------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------|
| Masks and PPE                 | Rechargeable antibacterial and antiviral nanofibrous membranes (RNMs)    | The photoactive RNMs can store biocidal activity and under light irradiation and readily release ROS to provide biocidal functions under dim light or dark conditions\(^{223}\) |
| Masks and PPE                 | Silver nanoparticles absorbed on a chitin sheet                          | Development of microbial/antiviral material, using Ag NPs absorbed on a chitin sheet with a nanoscale fibre-like surface structure is under development\(^{192,204,224}\) |
| Face masks                    | Graphene                                                                 | Few-layer graphene deposited onto low-melting temperature nonwoven masks. The masks could be reusable after sunlight sterilization\(^{202}\) |
| Face masks                    | Copper oxide                                                             | Impregnation of copper oxide into respiratory protective face masks endows them with potent anti-influenza biocidal properties without altering their physical barrier properties\(^{201}\) |
| Face masks                    | TiO\(_2\)-modified hydroxyapatite composite                               | Hydroxyapatite modified with anatase titanium dioxide composite shows antiviral activity against H1N1 Influenza A virus, which is promising to apply to masks\(^{194}\) |
| Face masks                    | Nanoporous membrane                                                      | A flexible nanoporous Si-based template on a silicon-on-insulator wafer is used as a hard mask during a reactive ion etching process to transfer the patterns onto a flexible and lightweight polymeric membrane. Pores with sizes down to 5 nm were achieved. The mask is antifouling and self-cleaning due to intrinsically hydrophobic properties of membrane\(^{203}\) |
| Face masks, PPE and wipes     | Catechin polyphenol                                                      | Chemical modification of the surface of non-woven cellulose fibre filters (Kimwipes\(^{®}\)) by fixing polyphenol could confer antiviral properties, which could be potentially used as filters in masks and wipes for the cleaning of surfaces and as protective clothing\(^{225}\). Sealing covers used in public transport system could be treated with silver through the in situ photoreduction of a silver solution\(^{21}\) |
| Public Transport              | Silver                                                                   | The surfaces could be simply coated with this polycation’s butanol solution, which exerts antimicrobial and antiviral properties\(^{97}\). |
| Various surfaces              | Polycation N,N-dodecyl, methyl-polyethylenimine (PEI)                    | The surfaces for cellulosic fibre filters could confer antiviral properties, which could be potentially used as filters in masks and wipes for the cleaning of surfaces and as protective clothing\(^{225}\) |
| Various surfaces              | Silica nanoparticles (SNPs) coated with a quaternary ammonium cationic surfactant, didodecyldimethylammonium bromide (DDAB). | SNPs coated with a quaternary ammonium cationic surfactant are very effective biocide and can be reused several times without significantly losing their biocidal activity. SNPs can be potentially attached to any substrate, by a chemical adhesive formed by polydopamine\(^{226}\) |
| Door frames, window panels, and hospital and medical equipment | Nanostructured surfaces | Nanostructures randomly aligned as ridges on aluminium 6063 alloy surfaces, providing antiviral properties\(^{212}\) |
of the mask or depositing an ultrathin nanoparticle layer on the respirators. Examples include copper oxide and silver nanoparticles incorporated into nanofiber membranes or the fabric of masks\(^{[191,201]}\). Another way is to make masks superhydrophobic, so that virus-laden droplets would not be able to remain on the masks. A possible approach is depositing a few layers of graphene onto commercial surgical masks\(^{[202]}\). It is proposed that an advantage of graphene coating comes from the possibility of its self-sterilisation triggered either by electrical charge induction or by exposing it to the sunlight for 40–100 s\(^{[202]}\). Other superhydrophobic materials, such as fluorinated polymers and metallic nanowires, have also the potential to be used in respiratory masks\(^{[203]}\).

Antiviral compounds incorporated to other types of PPE, such as apron or gloves, would further reduce the risk of infection for health care workers. Many antiviral materials, already used in masks and respirators, e.g., Zn or Ag nanoparticles\(^{[191,204]}\) or graphene\(^{[202]}\), with high antiviral efficiency and low-toxicity to humans, could be potentially used. Some drawbacks of utilising metal ions or metallic nanoparticles in PPE or other everyday-use surfaces should be considered when designing, manufacturing and implementing the products. Firstly, ion leaching can occur leading, eventually, to the loss of antiviral and antimicrobial properties of the material\(^{[205,206]}\). Secondly, there is a potential risk to the environment that metallic nanoparticles can exert after being released from products with, for example, domestic or industrial wastewater\(^{[207,208]}\). Photo-induced antimicrobial/antiviral materials are promising candidates for application in PPE as apart from high biocidal efficiency and long-term stability they can be “green” materials with low environmental concern\(^{[209,210]}\). By incorporating daylight-active chemicals into rechargeable nanofibrous membranes (RNMs), photoactive RNMs could act by releasing ROS to provide biocidal functions under dim light or dark conditions and store the biocidal activity under light irradiation, which could be potentially incorporated as a surface protective layer of PPE. There have been many advances to improve the antibacterial properties of medical devices especially those that go into the wet or humid environment of the body (e.g. catheters, tracheal and laryngeal tubes). Biopolymers and biocompatible polymer coatings could also be used to reduce viral transmission.

Large surfaces and infrastructure. Another application of antiviral materials is the use of antiviral surfaces or surface coatings in public settings such as healthcare facilities or public transport system, to slow down the spread of virus through fomites. This application requires durable materials with long-term (weeks or even months) antiviral efficiency. Similar to PPE, metal ions such as silver and copper are popular candidates for use in public areas. Deposition of silver clusters, through the photoreduction of a silver salt, directly on the surface could be implemented, e.g., on the natural leather seats in the public transport system\(^{[211]}\). The coatings based on polymers, incorporating metallic nanoparticles or metal ions, could protect the metal from oxidation and corrosion and could also be engineered for slow release of metal ions, providing long-lasting antiviral properties. A non-release approach, enhancing the durability of antiviral coatings, was proposed by Haldar et al.\(^{[97]}\), based on coating surfaces with biocidal PEI polymers, as described earlier. Some of the materials discussed in this review can be applied as a paint and these could be applied to a wide variety of surfaces from walls, doors and cabinets to equipment and other hard surfaces.

Hard surfaces. The ubiquitous touch screen is found everywhere, from personal mobile phones to medical equipment, and is notorious for harbouring bacteria and viruses. Many of the materials, such as copper, discussed in this review could be potentially used in thin-film coatings on glass surfaces. Door handles, taps and other frequently touched hard surfaces could similarly benefit from metal coatings such as copper. An alternative way is to generate nanostructured topography on industrially relevant surfaces that can physically inactivate viruses. Recently, Hasan et al.\(^{[212]}\) fabricated 23 nm wide nanostructures randomly aligned as ridges on aluminium 6063 alloy surfaces which could significantly reduce the viability of common respiratory viruses compared to the smooth surface\(^{[212]}\). With excellent durability, this strategy could be potentially used in hospitals and other public areas.

Outlook

The aim of this review is to provide an overview and better understanding of the current state of knowledge, research direction and practices in the area of antiviral materials and coatings. We focus on the reported mechanisms of action. The repertoire of materials with antiviral and antimicrobial properties is large and varied. Adding to that further possibilities of design and engineering of new chemistries, provides many options.
Antimicrobial properties of these materials are widely studied but reports on antiviral properties are much fewer and this is a gap that should be considered.

As can be seen, there is a significant resource of existing literature on viral persistence on different surfaces. However, a quantitative or even semi-quantitative analysis of the data is hampered by a lack of equivalence in the ways that persistence is measured or a consistent set of virus classes that are used to challenge the materials. There are existing ISO, ASTM, US Federal and EU standards on the measurement of antiviral activity but a lack of use in the literature. The most frequently used method is the end point dilution assay (TCID₅₀), though as noted earlier, the results are often difficult to interpret and are subjective. There appears to be a compelling need for systematic studies of different material types, challenged with specific virus strains of representative classes (enveloped, non-enveloped etc.) using quantitative approaches. This points to ‘high-throughput screening’ type experiments with material arrays. Furthermore, a reference standard surface, that could be used in an intercomparison study, would greatly benefit the community by providing a base-line of repeatability within a lab and replicability between laboratories. We believe that such studies would considerably increase the value and re-use of data created in future studies.

Copper is one of the most effective and simplest of the materials in this review and would appear to be easily integrated, e.g., as membrane-enveloped viruses with nanotechnology strategies: towards antiviral drug development and pandemic preparedness. ACS Nano 15, 125–148 (2021).

Received: 23 November 2020; Accepted: 6 April 2021; Published online: 28 May 2021

References

1. Walker, P. J. et al. Changes to virus taxonomy and the International Code of Virus Classification and Nomenclature ratified by the International Committee on Taxonomy of Viruses (2019). Archives Virol. 164, 2417–2429 (2019).
2. Colson, P., La Scola, B., Levasseur, A., Caetano-Anollés, G. & Raoult, D. Mimivirus: leading the way in the discovery of giant viruses of amoebae. Nat. Rev. Microbiol. 6, 243–254 (2007).
3. Ng, T. F. F. et al. Preservation of viral genomes in 700-y-old caribou feces from a subarctic ice patch. Proc. Natl Acad. Sci. USA 111, 16842–16847 (2014).
4. Woolhouse, M., Scott, F., Hudson, Z., Howey, R. & Chase-Topping, M. Human viruses: discovery and emergence. Philosophical Transactions of the Royal Society B: Biological Sciences 367, 2864–2871 (2012).
5. Chinchas, V. REPLICATION OF VIRUSES. Encyclopaedia of Virus. 1471–1478 (1999).
6. Moelling, K. & Broecker, F. Viruses and evolution - viruses first? A personal perspective. Front. Microbiol. 10, 523–523 (2019).
7. Retroviruses. In Overview of Reverse Transcription, (Eds Coffin, J. M., Hughes, S. H., Varmus, H. E.) (Cold Spring Harbor Laboratory Press, 1997).
8. Roos, W. H., Ivanovska, I. L., Evilevitch, A. & Wuite, G. J. L. Viral capsid: mechanical characteristics, genome packaging and delivery mechanisms. Cell. Mol. Life Sci. 64, 1484–1497 (2007).
9. Condit, R. C., Moussatche, N. & Traktman, P. In a nutshell: structure and assembly of the vaccinia virion. Adv. Virus Res. 66, 31–124 (2006).
10. Fokine, A. et al. Molecular architecture of the prolate head of bacteriophage T4. Proc. Natl Acad. Sci. USA 101, 6003–6008 (2004).
11. Sun, X. & Whitaker, G. R. Role for influenza virus envelope cholesterol in virus entry and infection. J. Virol. 77, 12543–12551 (2003).
12. Bosch, B. J., van der Zee, R., de Haan, C. A. M. & Rottier, P. J. M. The coronavirus spike protein is a class I virus fusion protein: structural and functional characterization of the fusion core complex. J. Virol. 77, 8801–8811 (2003).
13. Wiskichirken, K., Lucifora, J., Michler, T. & Poter, U. New pharmacological strategies to fight enveloped viruses. Trends Pharmacol. Sci. 35, 470–478 (2014).
14. Geoghegan, J. L., Senior, A. M., Di Giallonardo, F. & Holmes, E. C. Viral factors that increase the transmissibility of emerging human viruses. Proc. Natl Acad. Sci. USA 113, 4170–4175 (2016).
15. Yoon, B. K., Jeon, W.-Y., Sun, T. N., Cho, N.-I. & Jackman, J. A. Stopping material types, challenged with specific virus strains of representative classes (enveloped, non-enveloped etc.) using quantitative approaches. This points to ‘high-throughput screening’ type experiments with material arrays. Furthermore, a reference standard surface, that could be used in an intercomparison study, would greatly benefit the community by providing a base-line of repeatability within a lab and replicability between laboratories. We believe that such studies would considerably increase the value and re-use of data created in future studies.

Copper is one of the most effective and simplest of the materials in this review and would appear to be easily integrated, e.g., as membrane-enveloped viruses with nanotechnology strategies: towards antiviral drug development and pandemic preparedness. ACS Nano 15, 125–148 (2021).

Received: 23 November 2020; Accepted: 6 April 2021; Published online: 28 May 2021

References

1. Walker, P. J. et al. Changes to virus taxonomy and the International Code of Virus Classification and Nomenclature ratified by the International Committee on Taxonomy of Viruses (2019). Archives Virol. 164, 2417–2429 (2019).
2. Colson, P., La Scola, B., Levasseur, A., Caetano-Anollés, G. & Raoult, D. Mimivirus: leading the way in the discovery of giant viruses of amoebae. Nat. Rev. Microbiol. 6, 243–254 (2007).
3. Ng, T. F. F. et al. Preservation of viral genomes in 700-y-old caribou feces from a subarctic ice patch. Proc. Natl Acad. Sci. USA 111, 16842–16847 (2014).
4. Woolhouse, M., Scott, F., Hudson, Z., Howey, R. & Chase-Topping, M. Human viruses: discovery and emergence. Philosophical Transactions of the Royal Society B: Biological Sciences 367, 2864–2871 (2012).
5. Chinchas, V. REPLICATION OF VIRUSES. Encyclopaedia of Virus. 1471–1478 (1999).
6. Moelling, K. & Broecker, F. Viruses and evolution - viruses first? A personal perspective. Front. Microbiol. 10, 523–523 (2019).
7. Retroviruses. In Overview of Reverse Transcription, (Eds Coffin, J. M., Hughes, S. H., Varmus, H. E.) (Cold Spring Harbor Laboratory Press, 1997).
8. Roos, W. H., Ivanovska, I. L., Evilevitch, A. & Wuite, G. J. L. Viral capsid: mechanical characteristics, genome packaging and delivery mechanisms. Cell. Mol. Life Sci. 64, 1484–1497 (2007).
9. Condit, R. C., Moussatche, N. & Traktman, P. In a nutshell: structure and assembly of the vaccinia virion. Adv. Virus Res. 66, 31–124 (2006).
40. van Doremalen, N. et al. Aerosol and surface stability of SARS-CoV-2 as compared with SARS-CoV-1. *N. Engl. J. Med.* 382, 1564–1570 (2020).
41. Small, J., Yu, M. W. N. & Talbot, P. J. Survival of human coronaviruses 229E and OC43 in suspension and after drying onsurfaces: a possible source of ophthalmically-acquired infections. *J. Hosp. Infect.* 46, 55–60 (2000).
42. Lai, M. Y. Y., Cheng, P. K. C. & Lim, W. W. L. Survival of severe acute respiratory syndrome coronavirus. *Clin. Infect. Dis.* 41, e67–e71 (2005).
43. Zhang, L., Z. Q., Wu, M., Wang, L., C. P. Silver as a residual disinfectant to prevent biofilm formation in water distribution systems. *Appl. Environ. Microbiol.* 74, 1639–1641 (2008).
44. WHO. Silver in drinking-water. Background document for preparation of WHO Guidelines for drinking-water quality. *Geneva.* (2003).
45. EPA, U. S. E. P. A. U. S. 2018 Edition of the Drinking Water Standards and Health Advisories Table. *Environ. Sci.* 54, 403–435 (1992).
46. Antelman, M. S. In *Anti-pathogenic multivalent silver molecular semiconductors, PRECIOUS METALS–CONFERENCE*–141, (PERGAMON PRESS, 1980).
47. Kadar, M. et al. Antiviral Effect of New Disinfectant Containing A Silver Complex and Hydrogen peroxide as Active Agents. *(1993).*
48. Montes, L. F., Muchinik, G. & Fox, C. L. Jr Response of variella zoster virus and herpes zoster to silver sulfadiazine. *Catis.* 38, 363–365 (1986).
49. Han, J. et al. Efficient and quick inactivation of SARS coronavirus and other microorganisms exposed to the sunlight of some metal catalysts. *Biomed. Environ. Sci.* 176, 176–180 (2005). Inactivation of SARS-CoV, baclovirus and E. coli, via affecting their replication and propagation abilities, through exposure to the surfaces of Ag2O5 and Cu2O5.
50. Bright, K. R., Sicairos-Ruelas, E. E., Gundy, P. M. & Gerba, C. P. Assessment of the antiviral properties of silver-containing and copper/copper-containing zeolites (sodium aluminosilicate), tested on human coronavirus 229E, feline infectious peritonitis virus, and feline calicivirus F-9.
51. Ryu, W.-S. in *Molecular Virology of Human Pathogenic Viruses* (Ed. Ryu, W.-S.) 667–381 (Academic Press, 2017).
52. Wilen, C. B., Tilton, J. C. & Doms, R. W. HIV: cell binding and entry. *Cold Spring Harbor Perspect. Med.* 2, (2012).
53. De Clercq, E. Novel approaches towards anti-HIV chemotherapy. *Pharmacochem. Library* 24, 101–94 (1994).
54. Sezer, A. D. & Cerher, E. in *Active Implants and Scaffolds for Tissue Regeneration* (Ed Zberner, M.) 377–406 (Springer Berlin Heidelberg, 2011).
55. Malmsten, M. Antimicrobial and antiviral hydrogels. *Soft Matter 7*, 8725–8736 (2011).
56. Wahid, F., Zhong, C., Wang, H.-S., Hu, X.-H. & Chu, L.-Q. Recent advances in antimicrobial hydrogels containing metal ions and metals/metal oxide nanoparticles. *Polymers* 9, 636 (2017).
57. Spoden, G. A. et al. Polyethylenimine is a strong inhibitor of human papillomavirus and cytomegalovirus infection. *Antimicrob. Agents Chemotherapy* 56, 75–82 (2012).
58. Haldar, J., An, D., De Cienfuegos, L. Á., Chen, J. & Klibanov, A. M. Polymeric coatings that inactivate both influenza virus and pathogenic bacteria. *Proc. Natl Acad. Sci. USA* 103, 17667–17671 (2006). Shows how PEI painted (coated) surfaces, in addition to being extremely bactericidal, reduce the amount of influenza A and shows the relationship between the viridical activity and the coating polymer structure.
59. Mecke, A., Lee, D.-K., Ramamoorthy, A., Orr, B. G. & Holl, M. B. Synthetic and natural polycationic polymer nanoparticles interact selectively with fluid-membrane domains of DMPC bilayers. *Langmuir* 21, 8588–8590 (2005).
60. Wang, Y. et al. Cationic phenylene ethylene polymers and oligomers exhibit efficient antiviral activity. *ACS Appl. Mater. Interfaces* 3, 2209–2214 (2011).
61. Allam, A. N., Naggar, V. F. & El Gamal, S. S. Formulation and physicochemical characterization of chitosan/acyclovir co-crystals. *Pharm. Dev. Technol.* 18, 856–865 (2013).
62. Bajaj, H., Bish B., Yadav, M., Singh, V. & Singh, M. Design and development of nevirapine loaded surfactant free chitosan microemulsion. *Acta Pol. Pharm.* 68, 981–988 (2011).
63. Calderon, L. et al. Nano and microparticulate chitosan-based systems for antiviral topical delivery. *Eur. J. Pharm. Sci.* 48, 216–222 (2013).
64. Gopika, C. et al. Development of a drug delivery system based on chitosan nanoparticles for oral administration of interferon-alpha. *Biomacromolecules* 18, 3302–3309 (2017).
Yu, Y. et al. Engineered human cathelicidin antimicrobial peptides inhibit Ebola virus. Infection. Science 23, 100999 (2020).

Barlow, P. G. et al. Antiviral activity and increased host defense against influenza infection elicited by the human cathelicidin LL-37. PLoS ONE 6, e25333-e25333 (2011). Shows that cathelicidin LL-37 possess antiviral activity.

Currie, S. M. et al. Cathelicidins have direct antiviral activity against respiratory syncytial virus in vitro and protective function in vivo in mice and humans. J. Immunol. 196, 2699–2710 (2016).

He, M. et al. Cathelicidin-derived antimicrobial peptides inhibit Zika virus through direct inactivation and interferon pathway. J. Immunol. 9, 722 (2020).

Klotman, M. E. & Chang, T. L. Defensins in innate antiviral immunity. Nat. Rev. Immunol. 6, 447–456 (2006). Provides a comprehensive insight into the role of defensins in innate antiviral immunity.

Ding, J., Chou, Y. Y. & Chang, T. L. Defensins in viral infections. J. Innate Immunity 1, 413–420 (2009).

Holly, M. K., Diaz, K. & Smith, J. G. Defensins in viral infection and pathogenesis. Annu. Rev. Virol. 4, 369–391 (2017).

Daher, K. A., Selsted, M. E. & Lehrer, R. I. Direct inactivation of viruses by human granulocyte defensins. J. Virol. 60, 1068–1074 (1996).

Ikeda, M. et al. Characterization of antiviral activity of lactoferrin against hepatitis C virus infection in human cultured cells. Virus Res. 66, 51–63 (2000).

McCann, K. B., Lee, A., Wan, J., Roginski, H. & Coventry, M. J. The effect of bovine lactoferrin and lactoferrin B on the ability of feline calicivirus (a norovirus surrogate) and poliovirus to infect cell cultures. J. Appl. Microbiol. 95, 1026–1033 (2003).

Soomro, F., Ammori, G. M. V., Valenti, P. & Seganti, L. Antirotaviral activity of milk proteins: lactoferrin prevents rotavirus infection in the enterocyte-like cell line HT-29. Med. Microbiol. Immunol. 186, 83–91 (1997).

Lang, J. et al. Inhibition of SARS pseudovirus cell entry by lactoferrin binding to heparan sulfate proteoglycans. PLoS ONE 6, e25710 (2011). Shows the mechanism of action of lactoferrin binding proteoglycans in the inhibition of SARS pseudovirus.

van der Strate, B. W., Beljaars, L., Molema, G., Harmsen, M. C. & Meijer, D. K. He, M. et al. Cathelicidin-derived antimicrobial peptides inhibit Zika virus through direct inactivation and interferon pathway. J. Immunol. 9, 722 (2020).

Klotman, M. E. & Chang, T. L. Defensins in innate antiviral immunity. Nat. Rev. Immunol. 6, 447–456 (2006). Provides a comprehensive insight into the role of defensins in innate antiviral immunity.

Ding, J., Chou, Y. Y. & Chang, T. L. Defensins in viral infections. J. Innate Immunity 1, 413–420 (2009).

Holly, M. K., Diaz, K. & Smith, J. G. Defensins in viral infection and pathogenesis. Annu. Rev. Virol. 4, 369–391 (2017).

Daher, K. A., Selsted, M. E. & Lehrer, R. I. Direct inactivation of viruses by human granulocyte defensins. J. Virol. 60, 1068–1074 (1996).

Ikeda, M. et al. Characterization of antiviral activity of lactoferrin against hepatitis C virus infection in human cultured cells. Virus Res. 66, 51–63 (2000).

McCann, K. B., Lee, A., Wan, J., Roginski, H. & Coventry, M. J. The effect of bovine lactoferrin and lactoferrin B on the ability of feline calicivirus (a norovirus surrogate) and poliovirus to infect cell cultures. J. Appl. Microbiol. 95, 1026–1033 (2003).

Soomro, F., Ammori, G. M. V., Valenti, P. & Seganti, L. Antirotaviral activity of milk proteins: lactoferrin prevents rotavirus infection in the enterocyte-like cell line HT-29. Med. Microbiol. Immunol. 186, 83–91 (1997).

Lang, J. et al. Inhibition of SARS pseudovirus cell entry by lactoferrin binding to heparan sulfate proteoglycans. PLoS ONE 6, e25710 (2011). Shows the mechanism of action of lactoferrin binding proteoglycans in the inhibition of SARS pseudovirus.

van der Strate, B. W., Beljaars, L., Molema, G., Harmsen, M. C. & Meijer, D. K. Antiviral activities of lactoferrin. Rev. Immunol. 52, 225–239 (2001).

Walker, A. P., Partridge, J., Srai, S. K. & Dooley, J. S. Hepcidin: what every gastroenterologist should know. Gut 53, 624–627 (2004).

Armitage, A. E. et al. Distinct patterns of hepcidin and iron regulation during infection on hepcidin expression in hepatitis B patients. Ann. Clin. Lab. Sci. 11A, 1217–12192 (2014).

Wang, X. H., Cheng, P. P., Jiang, F. & Jiao, X. Y. The effect of hepatitis B virus infection on hepcidin expression in hepatitis B patients. Ann. Clin. Lab. Sci. 43, 126–134 (2013).

Rasimbabu, V. & Chen, J. Y. Antiviral function of tilapia hepcidin 1-5 and its modulation of immune-related gene expressions against infectious pancreatic necrosis virus (IPNV) in Chinook salmon embryo (CHSE-214) cells. Fish Shellfish Immunol. 30, 39–44 (2011).

Zhang, J. et al. Antiviral peptide nanocomplexes as a potential therapeutic modality for HIV/HCV co-infection. Biomaterials 34, 3866–3857 (2013).

Zhang, J., Garrigan, J., Polukhova, L. V., Bronich, T. K. & OsnSy, N. A. Liver-targeted antiviral peptide nanocomplexes as potential anti-HCV therapeutics. Biomaterials 70, 37–47 (2015).

Jackman, J. A. et al. Therapeutic treatment of Zika virus infection using a brain-penetrating antiviral peptide. Nat. Mater. 17, 971–977 (2018).

Camargos, V. N. et al. In-depth characterization of congenital Zika syndrome in immunocompetent mice: antibody-dependent enhancement and an antiviral peptide therapy. EbioloMedicine 44, 516–529 (2019).

Ferrari, A. C. et al. Science and technology roadmap for graphene, related two-dimensional crystals, and hybrid systems. Nanoscale 7, 4598–4810 (2015).

Legge, E. J. et al. Physicochemical characterisation of reduced graphene oxide for conductive thermal transfer. RSC Adv. 8, 37540–37549 (2018).

Ye, S. et al. Antiviral activity of graphene oxide: how sharp edged structure and charge matter. ACS Appl. Mater. Interfaces 7, 2517–25175 (2015). Shows how graphene oxide and reduced graphene oxide can be used as an antiviral agent against a DNA and RNA virus via structural destruction prior to viral entry.

Ziem, B. et al. Polyvalent 2D entry inhibitors for pseudorabies and African swine fever virus. Macromol. Biosci. 17, 1600499 (2017).

Elazzazy, A. M., Elbeshey, E. K. & Bethia, M. A. In vitro assessment of activity of graphene silver composite sheets against multidrug-resistant bacteria and Tomato Bushy Stunt Virus. Tropical J. Pharm. Res. 16, (2017).

Dokmar, A. R. et al. Graphene-based “hot plate” for the capture and destruction of the herpes simplex type virus 1. Bioconjugate Chem. 28, 1115–1122 (2017).
206. Zodrow, K. et al. Polysulphone ultrafiltration membranes impregnated with silver nanoparticles show improved biofouling resistance and virus removal. *Water Res.* 43, 715–723 (2009).

207. Durenkamp, M. et al. Nanoparticles within WWTP sludges have minimal impact on leachate quality and soil microbial community structure and function. *Environ. Pollut.* 211, 399–405 (2016).

208. Benn, T. M. & Westerhoff, P. Nanoparticle silver released into water from commercially available sock fabrics. *Environ. Sci. Technol.* 42, 7025–7026 (2008).

209. Velema, W. A. et al. Optical control of antibacterial activity. *Nat. Chem.* 5, 924–928 (2013).

210. Yeung, K. L., Leung, W. K., Yao, N. & Cao, S. Reactivity and antimicrobial properties of nanostructured titanium dioxide. *Catal. Today* 143, 218–224 (2009).

211. Pollini, M. et al. Antibacterial natural leather for application in the public transport system. *J. Coat. Technol. Res.* 10, 239–245 (2013).

212. Hasan, J. et al. Antiviral and antibacterial nanostructured surfaces with excellent mechanical properties for hospital applications. *ACS Biomater. Sci. Eng.* 6, 3608–3618 (2020). *Shows the combined antibacterial and antiviral property of the nanostructured surface with excellent nanomechanical properties which could be potentially significant for use in hospital environments to stop the spread of infections arising from physical surfaces.*

213. Ramakrishnan, M. A. Determination of 50% endpoint titer using a simple formula. *World J. Virol.* 5, 85–86 (2016).

214. Pourianfar, H. R., Javadi, A. & Grollo, L. A colorimetric-based accurate method for the determination of enterovirus 71 titer. *Indian J. Virol.* 23, 303–310 (2012).

215. Ryu, W.-S. in *Molecular Virology of Human Pathogenic Viruses* (Ed. Ryu, W.-S.) 47–62 (Academic Press, 2017).

216. Baer, A. & Kehn-Hall, K. Viral concentration determination through plaque assays: using traditional and novel overlay systems. *J. Visualized Exp.* e52065 (2014).

217. Kang, W. & Shin, E.-C. Colorimetric focus-forming assay with automated focus counting by image analysis for quantification of infectious hepatitis C virions. *PLoS ONE* 7, e43960 (2012).

218. Watzinger, F. et al. Real-time quantitative PCR assays for detection and monitoring of pathogenic human viruses in immunosuppressed pediatric patients. *J. Clin. Microbiol.* 42, 5189–5198 (2004).

219. Tahamtan, A. & Ardebili, A. Real-time RT-PCR in COVID-19 detection: issues affecting the results. *Expert Rev. Mol. Diagn.* 20, 453–454 (2020).

220. Qi, T. et al. Development of an antigen-capture ELISA for the quantitation of equine arteritis virus in culture supernatant. *Arch. Virol.* 163, 1469–1478 (2018).

221. Lippé, R. Flow virometry: a powerful tool to functionally characterize viruses. *J. Virol.* 92, e01765–17 (2018).

222. Richert-Pöggeler, K. R., Franzke, K., Hipp, K. & Kleesips, R. G. Electron microscopy methods for virus diagnosis and high resolution analysis of viruses. *Front. Microbiol.* 9, 3255 (2019).

223. Si, Y. et al. Daylight-driven rechargeable antibacterial and antiviral nanofibrous membranes for bioprotective applications. *Sci. Adv.* 4, eaar5931 (2018). *Shows that the green bioprotective nanofibrous membranes (RNMs) with rechargeable antibacterial and antiviral activities can be incorporated into protective equipment to achieve robust bio-protection against pathogens either in aerosol or in liquid forms.*

224. Ishihara, M., Nguyen, V.-Q., Mori, Y., Nakamura, S. & Hattori, H. Adsorption of silver nanoparticles onto different surface structures of chitin/chitosan and correlations with antimicrobial activities. *Int. J. Mol. Sci.* 16, 13973–13988 (2015).

225. Catel-Ferreira, M., Tnani, H., Hello, C., Cosette, P. & Lebrun, L. Antiviral effects of polyphenolic development of bio-based cleaning wipes and filters. *J. Virol. Methods* 212, 1–7 (2015).

226. Botequim, D. et al. Nanoparticles and surfaces presenting antifungal, antibacterial and antiviral properties. *Langmuir* 28, 7646–7656 (2012).

227. Yount, J. S., Gitlin, L., Moran, T. M. & López, C. B. MDA5 Participates in the detection of paramyxovirus infection and is essential for the early activation of dendritic cells in response to Sendai virus defective interfering particles. *J. Immunol.* 180, 4910–4918 (2008).

**Acknowledgements**

This work forms part of the 3D OrbiSIMS project in the Life-science and Health programme of the National Measurement System of the UK Department of Business, Energy and Industrial strategy.

**Author contributions**

All authors contributed to the writing of this review and approved the content. Specifically, P.D.R. introduction and metals section, C.B. viral activity and standards, Y.P. surface properties, M.T. polymers, N.F. biopolymers, antimicrobial peptides and composites, A.J.P. graphene and 2D materials, I.Z. and I.S.G. applications and I.S.G. wrote the outlook. I.S.G. and P.D.R. edited the review.

**Competing interests**

The authors declare no competing interests.

**Additional information**

**Supplementary information**

The online version contains supplementary material available at https://doi.org/10.1038/s43246-021-00153-y.

**Correspondence**

and requests for materials should be addressed to P.D.R. or I.S.G.

**Peer review information**

Primary handling editor: John Plummer

**Reprints and permission information**

is available at http://www.nature.com/reprints

**Publisher’s note**

Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

**Open Access** This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons license, and indicate if changes were made. The images or other third party material in this article are included in the article’s Creative Commons license, unless indicated otherwise in a credit line to the material. If material is not included in the article’s Creative Commons license and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this license, visit http://creativecommons.org/licenses/by/4.0/.