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Journal:	*ACS Nano*
Manuscript ID	nn-2021-006296.R3
Manuscript Type:	Review
Date Submitted by the Author:	02-Apr-2021
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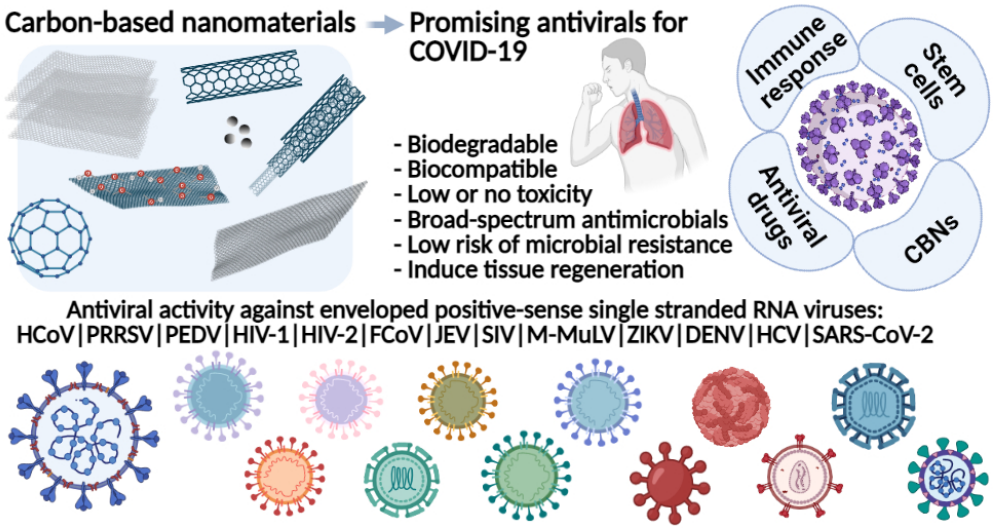

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Carbon-Based Nanomaterials: Promising Antiviral Agents to Combat COVID-19 in the Microbial Resistant Era

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Abstract

Therapeutic options for the highly pathogenic human Severe Acute Respiratory Syndrome-Coronavirus 2 (SARS-CoV-2) causing the current pandemic Coronavirus disease (COVID-19) are urgently needed. COVID-19 is associated with viral pneumonia and acute respiratory distress syndrome causing significant morbidity and mortality. The proposed treatments for COVID-19 have shown little or no effect in the clinic so far. Additionally, bacterial and fungal pathogens contribute to the SARS-CoV-2 mediated pneumonia disease complex. The antibiotic resistance in pneumonia treatment is increasing at an alarming rate. Therefore, carbon-based nanomaterials (CBNs), such as fullerene, carbon dots, graphene, and their derivatives constitute a promising alternative due to their wide-spectrum antimicrobial activity, biocompatibility, biodegradability, and capacity to induce tissue regeneration. Furthermore, the antimicrobial mode of action is mainly physical (*e.g.*, membrane distortion), characterized by a low risk of antimicrobial resistance. In this review, we evaluated the literature on the antiviral activity and broad-spectrum antimicrobial properties of CBNs. CBNs had antiviral activity against 13 enveloped positive-sense single-stranded RNA viruses, including SARS-CoV-2. CBNs with low or no toxicity to humans are promising therapeutics against COVID-19 pneumonia complex with other viruses, bacteria, and fungi, including those that are multidrug-resistant.

Keywords: COVID-19, SARS-CoV-2, carbon-based nanomaterials, fullerene, carbon dots, graphene, antiviral properties, pneumonia, tissue regeneration

History has repeatedly manifested that pathogens cause disastrous effects on human beings. Thus, the recent outbreak of the recent Severe Acute Respiratory Syndrome-Coronavirus 2 (SARS-CoV-2), which causes Coronavirus disease 2019 (COVID-19), spread to more than 200 countries, is a clear example. The current confirmed global COVID-19 cases and deaths have reached more than 90 million and more than 2 million, respectively.[1] Nevertheless, experts have suggested that many more undetected or asymptomatic cases exist,[2] especially in underdeveloped countries. COVID-19 continues to spread globally, threatening to collapse the health system of many developed countries such as United Kingdom and France that have been forced to go to a third lockdown. SARS-CoV-2 is an enveloped positive-sense, single-stranded RNA virus.[3–5] Its origin to this date remain enigmatic, however multiple hypotheses have been postulated so far.[6] However, the host tropism/adaptation pattern raised questions concerning the origin of SARS-CoV-2.[7] SARS-CoV-2 is the seventh coronavirus known to infect humans easily[8,9] and only the third one causing severe pneumonia.[10] Viral pneumonias may be complicated by secondary microbial infections.[11] Thus, co-infection can be caused by viruses in the setting of community-acquired bacterial pneumonia.[12–14] Co-infection of COVID-19 patients is seen with the most common type of bacterial pneumonia caused by *Streptococcus pneumoniae*.[15] There is a great concern about the rapid spread of pathogens, such as SARS-CoV-2 that can coexist with a broad range of other types of clinically relevant microorganisms, including those which are multidrug-resistant. Therefore, the co-infection of SARS-CoV-2 with other viruses, bacteria, or fungi constitutes a real life-threatening to humans during the approaching cold season. In this regard, several medications have been proposed which include remdesivir, hydroxychloroquine, lopinavir/ritonavir, interferon β-1a**,** tocilizumab, favipiravir, plitidepsin, convalescent plasma infusions and monoclonal antibodies, among many others.[16] However, presently, there is no effective treatment for COVID-19.[17,18] Furthermore, antibiotic resistance in bacterial pneumonia treatment is a wide-spread problem.[19–21] Therefore, in the quest to finding therapeutics for COVID-19, carbon-based nanomaterials (CBNs) are emerging as promising options that have shown potent antiviral activity against a broad range of enveloped positive-sense single-stranded RNA viruses,[22] including SARS-CoV-2, and showed low to no toxicity in humans.[23–28] Besides, they exert an effective biocidal action against a broad spectrum of bacteria, viruses and fungi, including multidrug-resistant strains.[29–31] These CBNs are mainly composed of carbon, an essential element in the human body,[32] are thus biodegradable, biocompatible and can induce tissue regeneration.[33–37] Moreover, the development of CBNs as antiviral agents is possible because they possess a high surface area that allows its functionalization or interaction with biocompatible polymers which further enhance their biocompatibility and therapeutic efficacy. The epithelial response to viral challenge is well-documented to involve autophagic and apoptotic cell death.[38] Indeed, the primary receptor for SARS-CoV-2, the carboxypeptidase ACE-2, is a protective "survival factor" for human lung alveolar epithelial cells (AECs),[39] but is significantly reduced by SARS-CoV2 infection.[40] The cells which express the most ACE-2 in the human lung alveoli, the type II AECs, normally serve as stem cells for regeneration of lost alveolar epithelia,[41] but are killed by viral challenge. For these reasons, any potential therapeutic that exerts both antiviral activity and the capacity to stimulate tissue regeneration would be expected to promote lung tissue repair in the face of ongoing viral-induced cell death. Research in this area is still in its early stage, though it is predicted to grow exponentially due to the grave consequences caused by the current pandemic. The increasing number of multidrug-resistant pathogens announced by the World Health Organization constitutes another real threat to humanity in this microbial resistance era.[42] Therefore, there is an

urgent need to find alternative antimicrobial strategies to curb the drug-resistance menace, thus providing a long-lasting treatment for the COVID-19 disease. This review addresses the application of these broad-spectrum CBNs as antimicrobial agents by analyzing a large number of antiviral studies performed so far with CBNs against enveloped positive-sense single-stranded RNA viruses, looking at their toxicity and biodegradability, and deciphering how they could defeat microbial resistance. This vision could surpass substantially any technological paradigms that currently exist or are under development a part of advanced therapeutics to treat COVID-19.

Next Generation of Antimicrobials: Carbon-Based Nanomaterials

Antiviral activity of viral infection compounds can be classified into two different subgroups: (1) virus-inhibiting compounds (usually cellular); or (2) compound that augments host defense improving or altering virus infections (immunomodulating agents).[43] Antiviral mechanisms usually require an inhibitory effect of a particular viral cell cycle essential in viral replication. As viral replication mainly depends on the host cell's metabolism as viruses are obligate intercellular parasites, valuable antivirals can disrupt viral-specific functions or at least obstruct virus-directed functions as opposed to the host cell. Therefore, the spectrum of antivirals generally is restricted. Many existing antiviral agents block host cell surface attachment followed by conformational changes of the viral capsid, leading to the uncoating of the viral genome, preventing the virus from penetrating the target cell. Others, such as nucleoside or nucleotide analogs, are antiviral drugs that provide an impairment of nucleic virus synthesis as their mode of action. An essential factor for antiviral activity is the inhibition of virus attachment to a cellular receipt or viral entry, inhibiting the viral from entering the cells. Many viruses involve proteolytic cleavage of polypeptides precursors utilizing protease inhibitors as antivirals to active, vital viral proteins. Some of the antivirals inhibit the integration of the viral genome with the host cells' genome. Other antivirals inhibit the uncoating (viral disassembly) and therefore distributing the viral life cycle. Finally, the inhibition of the virus release from the host cells through the prevention of viral clumping and thus inhibition cell-to-cell movement of viruses through enzymatic inhibition.[44–46]

To control the ever-increasing range of infections caused by multidrug-resistant microorganisms and viruses,[42] the use of antibiotics or alternative antimicrobial agents such as the ionic and/or oxidized form of metals,[47] quaternary ammonium compounds,[48] peptoids,[49] α-peptides[50] and β-peptides[51] present diverse problems including drug resistance after long-term utilization. In this regard, alternative materials such as CBNs with intrinsic broad-spectrum antimicrobial activity[52–63] represent a promising option that would probably overcome the microbial resistance problem due to their differential antimicrobial mechanisms. Thus, the antimicrobial action of CBNs, such as graphene (G) is often attributed to a combination of several physical and chemical mechanisms: directly on the microbial particle such as (peptidoglycan membrane structure disruption, entrapment of microorganisms, transfer of electrons) and and/or indirect through induction of oxidative stress by reactive oxygen species (ROS).[64,65] Other nanomaterials such as those based on silver, copper, titanium or zinc nanoparticles, for example, have shown strong broad-spectrum antimicrobial properties.[47,66–69] However, the existence of microbial resistance to these nanomaterials,[70–76] and their high toxicity to mammalian cells[77–81] render these materials less promising for long-term therapeutics. Therefore, CBNs are increasingly proposed as the next generation of antimicrobials against multidrug-resistant infections. They possess unique properties that include very high surface area, excellent electrical and thermal conductivity, biocompatibility, and the

possibility to be combined with engineered polymers to develop advanced antimicrobial biomaterial composites.[29,82–86] Furthermore, a recent study about the paramount concern of this proposal regarding to the interaction of CBNs with the respiratory system showed that a single exposure of several CBNs (at ~0.3 and 1 μg/cm^2) did not manifest any adverse effects under acute exposure scenarios after 24 h.[87] Regarding the biodegradability of CBNs, it has been demonstrated that the human myeloperoxidase, a peroxide enzyme released by neutrophils, degraded graphene and its derivatives[88,89] leading to biodegradation of graphene-based nanomaterials (GBNs) in the blood after 14 days.[90] Some signals of *in vivo* degradation of graphene were reported in the lungs, liver, kidneys and spleen upon 90 days.[91] Oxidized forms of GBNs have shown higher susceptibility to degradation than the reduced forms.[92] The degradation products of graphene oxide (GO) with different dimensions exhibited no genotoxicity to human lung cells.[93] *In vivo* studies have shown that a concentration of 1.0 mg/kg of small GO particles (148-160 nm) tends to accumulate mainly in the liver with a lower amount in the spleen and lungs, and remain longer in circulation than large ones (556-780 nm),[94] which were mostly present in lungs. However, when the concentration of injected GO, increased 10-folds, the smaller GO particles accumulated in the lungs instead of the liver thereby potentially increasing its efficacy for treating pulmonary infections. GO and reduced GO (rGO) of different lateral dimensions (10-800 nm) in a concentration of 1 mg/kg exhibited long blood circulation times of 14 days after intravenous administration in mice and low uptake by the endothelial reticulum. No pathological variations were detected in the analyzed organs.[86] Biodegradation and cytotoxicity towards different cell lines depended on concentration, exposure time, oxidation degree, lateral size, and cell type[95–98] and could be modified by functionalization.[95,99–102] Smaller and more oxidized GBNs seem to be more cytocompatible than non-oxidized and larger particles.[100] Proteins, such as bovine serum albumin, adsorbed on the GBNs' surfaces, seem to have a protective effect on the hemolytic potential.[103–106] The biocompatibility of CBNs depends on their concentration, oxidation degree, lateral size and dispersibility.[107–115] The inflammation, and other effects on cells and blood components are minimal when a lower concentration of CBNs is applied. Oxidized and smaller GBNs are more biocompatible and accessible to biodegradation in the body. CBNs have shown the potential to support the growth, proliferation, and differentiation of stem cells into different tissue lineages.[35,116] These features potentiate the use of CBNs in combination with stem cell therapies for tissue regeneration as well as for COVID-19 patients. SARS-CoV-2 infection is accompanied with many physiopathological changes and could reach all human organs harnessing several pathways,[117] during this it reaches and damages the vascular endothelial cells. CBNs can induce the production of angiogenic factors which initiate a series of signaling pathways to induce angiogenesis through promoting the proliferation and differentiation of endothelial cells or mesenchymal stem cells (MSCs).[118–120] In addition to the MSCs are not infected with SARS-CoV-2, of note that the stem cells (specially the mesenchymal stem) represent one of promising strategy for COVID-19 therapy, specifically those in moderate and/or severe infection.[121–124] Besides, MSCs downregulate Th1 and Th17 inflammation immunity and upregulate the anti-inflammation immunity of Th2 and Treg cells, it enhances the recruitment and proliferation of many productive cells and supportive materials (collage and extracellular matrix). Because of its potential, up to 21.02.2021, nine (out of ninety-one) clinical trial has been completed.[125] MSC are capable to self-replication and differentiation into specialized functions to replacing the disrupted and/or dysfunctions cells/tissues.[126] SARS-CoV-2 induces its pathological changes *via* exacerbate cytokines production which leading to many complications may reach to severe in some cases. Stem cells have specific cytokines[127] that tightly derive

immunomodulation (Figure 1) which may not only be helpful in control the SARS-CoV-2 infection severity,[126,128–130] but extend to beyond patient recovery specifically from severe infection.[131]

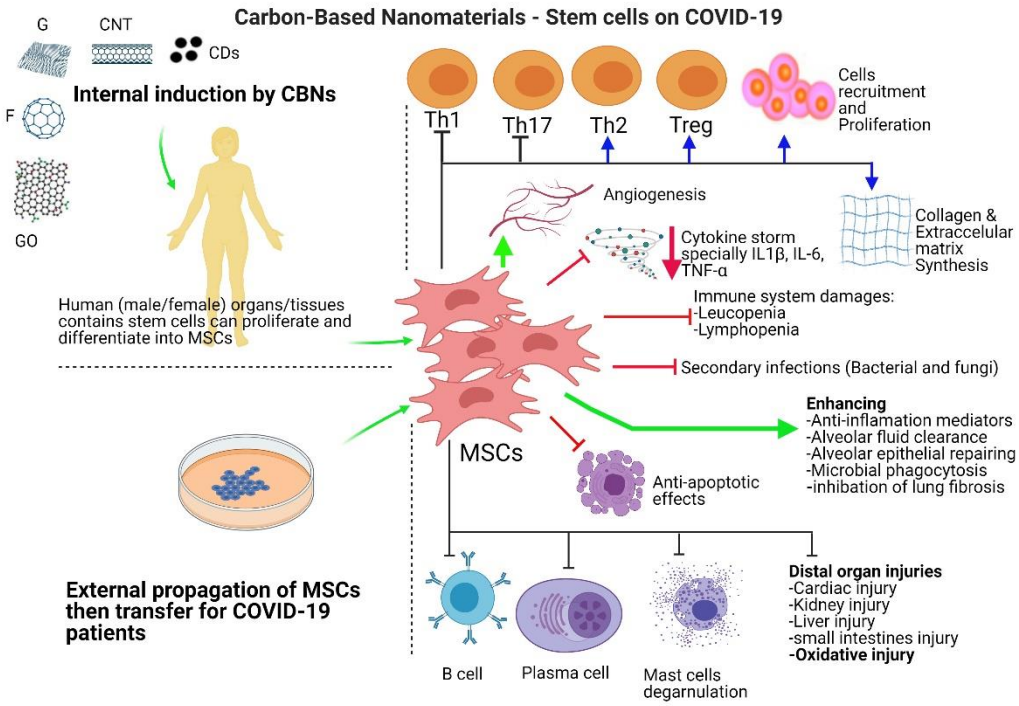

Figure 1. General panorama for external propagated and differentiated mesenchymal stem cells (MSCs) or internal induced of many tissues contains MSCs by CBNs (G: graphene, GO: graphene oxide, F: fullerene, CDs: carbon dots or CNT: carbon nanotubes). MSCs have various roles in COVID-19 and/or recovered patients through secretion and modulation of physiological and immunological networks. SARS-CoV-2 infection causes many pathophysiological changes such as tissue inflammation, immune system damages (leukopenia, lymphopenia), respiratory microstructures and distal organs injury and secondary infections, and microvascular system. CBNs in combination with MSCs have the potential to target these pathophysiological events, acting as an alternative strategy for treating COVID-19 patients.

Antiviral Properties of Carbon-Based Materials

This section analyzes the antiviral properties of CBNs with different carbon-based structures (Figure 2), such as fullerene, carbon dots, graphene, and derivatives against 13 enveloped positive-sense single-stranded RNA viruses, such as SARS-CoV-2.

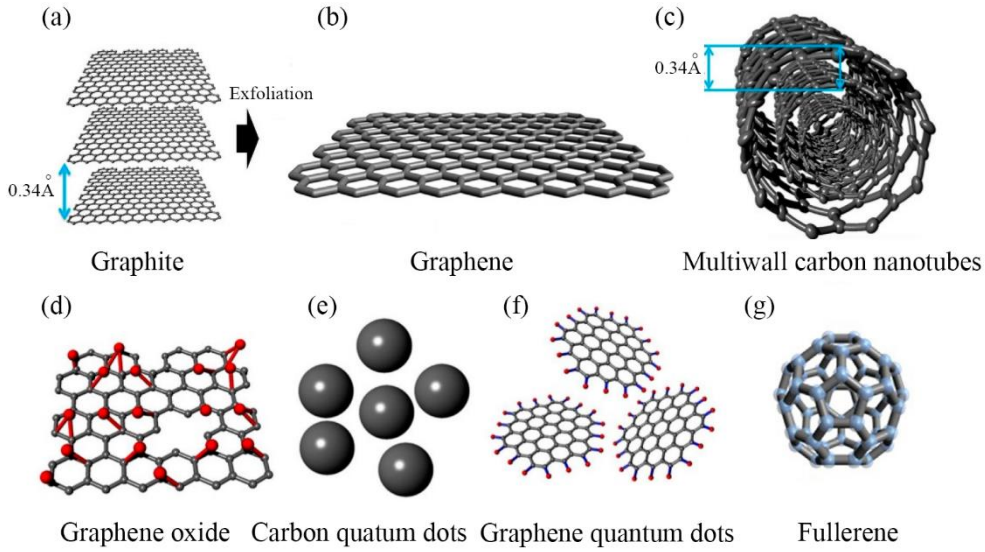

Figure 2. Main carbon-based structures studied against enveloped positive-sense single-stranded RNA viruses: (a) Graphite. Reprinted in part with permission under a Creative Commons CC BY 4.0 License from ref 133. Copyright 2019 MDPI; (b) Graphene. Reprinted in part with permission from ref 132. Copyright 2019 Elsevier; (c) Multiwall carbon nanotubes. Reprinted in part with permission under a Creative Commons CC BY 4.0 License from ref 133. Copyright 2019 MDPI; (d) Graphene oxide. Reprinted in part with permission under a Creative Commons CC BY 4.0 License from ref 133. Copyright 2019 MDPI; (e) Carbon quantum dots. Reprinted in part with permission under a Creative Commons CC BY 4.0 License from ref 133. Copyright 2019 MDPI; (f) Graphene quantum dots. Reprinted in part with permission under a Creative Commons CC BY 4.0 License from ref 133. Copyright 2019 MDPI; (g) Fullerene. Reprinted in part with permission from ref 132. Copyright 2019 Elsevier.

Fullerene and Derivatives

Fullerene is a zero-dimensional allotrope of CBNs with antiradical and antioxidant properties.[134,135] Due to the high hydrophobicity character of pristine fullerene, antiviral fullerene derivatives can be synthesized to produce hydrophilic drugs that can be easily dispersed in aqueous media.[30] Studies on fullerenes as antiviral agents started in 1993 on human immunodeficiency virus type 1 (HIV-1) infections.[136] In that study, compound 1 showed effective *in vitro* antiviral activity. Another study showed that a bis(monosuccinimide) derivative of p,p'-bis(2-aminoethyl)diphenyl-C_{60} was actively inhibiting the HIV-1 and also the type 2 (HIV-2) in acutely or chronically infected human lymphocytes and against 3'-azido-3'-deoxythymidine-resistant HIV-1. Another subsequent study showed a water-soluble fullerene-peptide conjugate capable of interacting, albeit weakly, with the HIV-1 protease.[137] In 1996, nine functional derivatives of C_{60}-fullerene compounds displayed antiviral capacity at low micromolar concentrations.[138] Furthermore, three of these compounds exhibited antiviral activity at lower concentrations than any fullerene derivative reported to that date. In 1997, nonderivatized fullerene (buckminsterfullerene) showed *in vitro* antiviral activity against another 2 enveloped positive-sense single-stranded RNA viruses similar to SARS-CoV-2: the Moloney murine leukemia virus (M-MuLV) and the simian immunodeficiency virus (SIV).[139] However, a study performed in 2003 tested a series of fullerene derivatives

(13 in total, compounds 1-13) against HIV-1 and HIV-2. The results of that study showed that some of these CBNs (6 (*trans-2*), 7 (*trans-3*) 8 (*trans-4*), 9 (*equatorial*), 12 (*trans-2*)) exhibited potent antiviral activity against HIV-1 but not HIV-2 in the low micromolar concentration range.[140] Nonetheless, cationic fullerene derivatives showed broader anti-HIV properties.[141] In 2007, chlorofullerene was developed as a precursor for the straightforward synthesis of antiviral fullerene derivatives against HIV with high solubility in water.[142] A later study in 2011 showed that derivatives of C_{70}-fullerene (*i.e.*, the fullerene molecule consisting of 70 carbon atoms) exhibited high water-solubility and virucidal activity against HIV and influenza virus.[26] A series of fullerene derivatives (Figure 3) showed potent viral inhibition of hepatitis C virus (HCV).[143]

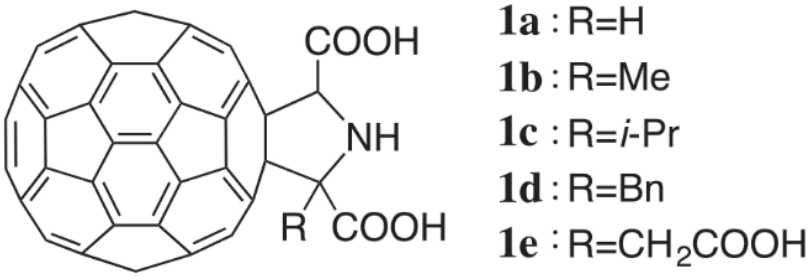

Figure 3. Chemical structure of fullerene derivatives 1a-1e. Reprinted with permission from ref 143. Copyright 2016 Elsevier.

Recently, tridecafullerenes appended with up to 360 1,2- mannobiose showed outstanding antiviral activity against Zika virus (ZIKV) and Dengue virus (DENV).[24] Examples of antiviral studies performed with fullerenes and their derivatives are summarized in Table 1.

Table 1. Studies analyzing the antiviral properties of carbon-based nanomaterials (fullerene, carbon dots, graphene and derivatives) against 13 enveloped positive-sense single-stranded RNA viruses. Source and manufacture of the CBNs, 50% cytotoxic concentration (CC50), half maximal effective antiviral concentration (EC50), tested viruses, tested cell line/inhibition, year and references are indicated for each study.

Fullerene and Derivatives	**Source and Manufacture**	**Toxicity (CC50)**	**Antiviral (EC50)**	**Tested viruses**	**Tested cell line/inhibition**	**Year**	**Ref.**
Fullerene derivatives (Compound 1)	Synthesis of bis(phenethylamino-succinate)-C_{60}	None (compound 1)	7 μM	HIV-1	PBMC/HIV-1 protease	1993	136
Memethanofullerene (2c)	Synthesis of diamido diacid diphenyl fulleroid derivative	Not tested	Effective at 1 mg/ml	HIV-1 and HIV-2	HIV- 1 and HIV-2 protease and reverse transcriptase	1993	144
Derivatized C_{60} Fullerene	Synthesis of bis(monosuccinimide) derivative of p,p'-bis(2-aminoethyl)diphenyl-C_{60} (compound 1)	>100 μM (compound 1) >640 μM (phosphonoformate) >100 μM (3'-Azido-3'-deoxythymidine)	HIV-1 10.8 μM, HIV-2 5.5 μM HIV-1 not tested, HIV-2 0.44 μM HIV-1 >100 μM, HIV-2 0.003 μM	HIV-1 and HIV-2	PBM, H9, Vero, and CEM /HIV-1 protease	1993	145
Bioactive fullerene peptide	Synthesis of synthon 1,2-dihydro-l,2-methanofullerene [60]-61-carboxylic acid covalently linked to the α-amino group of the hydrophilic 4-8 sequence of peptide T	Not tested	6 nM	HIV-1	HIV-1 protease	1994	137
Functional derivatives of C_{60}-fullerene	Synthesis of fullerene derivatives (9 active compounds)	>100 μM (compound 1) >100 μM (compound 2) >100 μM (compound 3)	7.3 μM 2.5 μM 0.9 μM	HIV-1	PBMC and Vero	1996	138
Nonderivatized fullerene (buckminsterfullerene)	C_{60} of Gold Grade purity (Hoechst AG, FrankfuM, Germany)	Not tested	3 μM	SIV and M-MuLV	MT-2 (for SIV) and M-MuLV reverse transcriptase inhibition	1997	139
Bis-functionalized fullerene derivatives bearing two or more solubilizing chains	Synthesis of fullerene derivatives (13 compounds tested)	4.79 μM (trans-2) 3.02 μM (trans-3) 13.2 μM (trans-4) 6.59 μM (equatorial)	0.40 μM 0.96 μM 2.60 μM 1.60 μM	HIV-1 and HIV-2 (effective against HIV-1, but not HIV-2)	CEM	2003	140
Cationic fullerene derivatives	Synthesis of a series of regioisomeric bis-fulleropyrrolidines bearing two ammonium groups (compounds 3-7)	2.93 μM (compound 3) 9.04 μM (compound 4) 12.5 μM (compound 5)	HIV-1 0.21 μM, HIV-2 0.2 μM HIV-1 0.35 μM, HIV-2 0.70 μM HIV-1 1.08 μM, HIV-2 2.50 μM	HIV-1 and HIV-2	CEM	2005	141
Polycarboxylic fullerene derivatives using chlorofullerene as a precursor	Friedel–Crafts arylation of $C_{60}C_{l6}$ with methyl esters of phenylacetic and benzylmalonic acids	>63 μM (compound 4a) 2.9 μM (compound 7) 9.0 μM (compound 8)	HIV-1 1.2 μM, HIV-2 4.4 μM HIV-1 0.21 μM, HIV-2 0.2 μM HIV-1 0.35 μM, HIV-2 0.7 μM	HIV-1 and HIV-2	CEM	2007	142
Polycarboxylic derivatives of C_{70}-fullerene	Synthesis of $C_{70}[p\text{-}C_6H_4(CH_2)_nCOOH]_8$ (n = 2, 3) starting from chlorinated [70]fullerene precursors $C_{70}Cl_8$ and $C_{70}Cl_{10}$	>43 μM (compound 2aK) >86 μM (compound 2bK)	HIV-1 1.8 μM, HIV-2 23 μM HIV-1 3.3 μM, HIV-2 17 μM	HIV-1 and HIV-2	CEM and MDCK	2011	26
Fulllerene derivatives (1a, 1b, 1c, 1d, 1e)	Synthesis of proline-type fullerene derivatives	Not tested (compound cis-1a) Not tested (compound trans-1a)	NS5B 0.29 μM, NS3/4A 0.15 μM NS5B 0.23 μM, NS3/4A 0.85 μM	HCV	NS5B polymerase and HCV NS3/4A protease	2016	143
Tridecafullerenes appended with up to 360 1,2-mannobiosides	Synthesis of multivalent disaccharide/[60]fullerene nanoballs	>10 μM (compound 32)	ZIKV 67 pM, DENV 35 pM	ZIKV and DENV	Jurkat	2019	24
Carbon dots and Derivatives		**Toxicity (CC50)**	**Antiviral (EC50)**	**Tested viruses**	**Tested cell line/inhibition**	**Year**	**Ref.**
Carbon dots	Synthesis from PEG-diamine and ascorbic acid by a process of grounding, autoclaving and purification	>0.250 mg/mL	effective at 0.125 mg/ml only	PRRSV	PK-15 and MARC-145	2016	63
Boronic acid-attributed carbon quantum dots	Synthesis by calcination of citric acid anhydrous and reaction with 4-carboxy-3- chlorobenzeneboronic acid, 4-dimethylaminopyrid and 1-ethyl-3-(3- dimethylaminopropyl) carbodiimide	>600 μg/ml	4.69-9.37 μg/ml	HIV-1	MT-4/HIV-1 and MOLT-4	2016	61
Functional carbon quantum dots	Synthesis of CQDs-5 and CQDs-6 according to the authors' protocol[146]	>100 μg/ml (CQDs-5) >100 μg/ml (CQDs-6)	10-20 μg/ml 2-5 μg/ml	HCoV	Huh-7	2019	22
Benzoxazine monomer derived carbon dots	Synthesis from benzoxazine monomers and NaOH by an process of autoclaving and purification	>75 μg/ml	JEV 18.63 μg/ml, ZIKV 3.715 μg/ml, DENV 37.49 μg/ml	JEV, ZIKV and DENV	BHK-21 (for JEV) and Vero (for ZIKV and DENV)	2019	147
Glycyrrhizic-acid-based carbon dots	Synthesis from glycyrrhizic acid of Chinese herbal medicine by a hydrothermal method	>0.90 mg/ml	0.30 mg/ml	PEDV, PRRSV	Vero (for PEDV), MARC-145 (for PRRSV)	2020	54
Graphene and Derivatives		**Toxicity (CC50)**	**Antiviral (EC50)**	**Tested viruses**	**Tested cell line/inhibition**	**Year**	**Ref.**
Graphene	Single layered graphene modeled with the Avogadro software	Not tested	Not tested	HIV-1	Essential target proteins (HIVVpr, Nef and Gag)	2014	148
Multi-walled carbon nanotubes: Pristine MWCNT (*p*-MWCNT), ox-MWCNT; CHI360, CHI415	*p*-MWCNT produced by catalytic chemical vapor deposition from isobutane on a Fe/Al_2O_3 catalyst and purified (>95%). ox-MWCNT prepared by oxidation of *p*-MWCNT with nitric acid/sulfuric acid (1:3)	21.22 μg/ml (*p*-MWCNT) 11.43 μg/ml (ox-MWCNT) 5.48 μg/ml (CHI360) 75.73 μg/ml (CHI415)	>69.35 μg/ml 9.04 μg/ml 0.01 μg/ml 0.42 μg/ml	HIV-1	MT-4	2015	149
Graphene-based materials (GO, rGO, GO/PVP, GO/PDDA, Gt, GtO)	GO from Nanjing XFNANO Materials Tech. Gt from Sigma-Aldrich. GtO from Gt *via* the modified Hummers method. Prepared GO-PDDA, GO-PVP[150], and rGO[151].	>50 μg/ml	PRV effective at 6 mg/ml PEDV effective at 1.5 mg/ml No (GO/PDDA and Gt)	PRV, PEDV	Vero	2015	28
Graphene oxide with silver nanoparticles	GO *via* Hummers' method and dispersion in a solution with $AgNO_3$ and ethylene glycol in microwave oven	12.5-25 mg/ml	FCoV effective at 0.1 mg/ml IBDV effective at 0.0625 mg/ml	FCoV, IBDV	fcwf-4	2016	152
Graphene oxide with silver nanoparticles	GO from Chengdu Organic Chemicals and AgNO3 from Sigma-Aldrich were self-assembled *via* interfacial electrostatic force	8.0 μg/ml	PRRSV effective at 4.0 μg/ml PEDV effective at 2.0 μg/ml	PRRSV, PEDV	MARC-145(for PRRSV) and Vero (for PEDV)	2018	153
Water-soluble GQD and drug-conjugated GQD	Water-soluble GQD prepared by prolonged acidic oxidation of *p*-MWCNT[154]. The anchorage of the antiretroviral drugs to the GQD surface by coupling reactions between the nucleophilic amino groups of the drugs and the carboxylic functionalities of the GQD	62.82 μg/ml (CHI499) 2.71 μg/ml(CDF119) 23.9 μg/ml (GQD-CHI499)	0.12 μg/ml 0.64 μg/ml 0.066 μg/ml	HIV-1	MT-4	2018	155
Graphene platforms with precise dual sulfate/alkyl functionalities	The surface of graphene is functionalized with polyglycerol sulfate (PGS) and aliphatic chains of different length (C6, C9, C10, C11, C12)	>1000 μg/ml (G-PGS-C9) 63.4 μg/ml (G-PGS-C10) 68.9 μg/ml (G-PGS-C11) 100.1 9 μg/ml (G-PGS-C12)	FCoV 749.4 μM, SARS-CoV-2 339.7 μM FCoV 9.8 μM, SARS-CoV-2 29.1 μM FCoV 6.3 μM, SARS-CoV-2 0.8 μM FCoV 82.2 μM, SARS-CoV-2 22.9 μM	FCoV, SARS-CoV-2	A549, HBE, Vero		156

Carbon Dots and Derivatives

Carbon dots (CDs), also known as carbon quantum dots (CQDs) or Cdots, are other members of the CBNs family with small dimensions up to 10 nm in diameter, cost-effective, and environmentally inert.[23,157] It possesses an organic molecule's chemical functionality, shows a very high surface-to-volume ratio, and can be homogeneously dispersed in water. Many CDs are explored in several fields, such as chemical sensing, bioimaging, electrocatalysis, and other applications.[62,158] The antiviral activity of functionalized CDs produced from 4-aminophenylboronic acid against human coronavirus (HCoV) infections has been recently demonstrated in human Huh-7 liver cells[22] (see Table 1). Benzoxazine monomer-derived carbon dots (BZM-CDs) were effective against the Japanese encephalitis virus (JEV), ZIKV, and DENV.[147] The antiviral activity of highly biocompatible glycyrrhizic-acid-based carbon dots (Gly-CDs) synthesized from glycyrrhizic acid was demonstrated against large-enveloped RNA viruses by using the porcine epidemic diarrhea virus (PEDV) as a viral model of the coronavirus (CoVs).[54] This virus belongs to the *alphacoronavirus* genus and it cannot be transmitted to humans.[159]

Graphene and Related Carbon-Based Nanomaterials

Graphene and its oxidated form, GO, are 2D CBNs with excellent physical and biological properties[160–164] that can be used successfully to detect, and capture viruses[148,165] (see Table 1). The antiviral activity of carbon nanofibers (CNFs) incorporated into alginate was reported using a non-enveloped double-stranded viral model.[166] However, the antiviral capacity of CNFs has never been tested against an enveloped virus belonging to the same Baltimore group[167] such as SARS-CoV-2. Functionalized multiwall carbon nanotubes (MWCNT), another type of carbon-based filamentous nanomaterials, exhibited potent viral inhibition against HIV.[149] In this study, the effect of hydrophilicity and dispersibility of the nanomaterials showed to be the key to control the antiviral activity of MWCNT-based nanomaterials. Carboxylated MWCNT (ox-MWCNT sample) and drug-conjugated MWCNT (MWCNT-C-CHI36) showed high antiviral activity contrary to pristine MWCNT. GO has also shown potent antiviral activity with nonionic polyvinyl pyrrolidone (PVP) in contrast to its combination with the cationic poly(diallyl dimethyl ammonium chloride) (PDDA).[28] The combination of silver nanoparticles (AgNPs) with GO inhibited the infectivity of enveloped feline coronavirus (FCoV) by 25% compared to 16% for GO.[152] AgNPs are well-known alternative antiviral agents that interact with cell surface receptors and blocks of the virus entry into the host cells.[66] In the same research line, GO-AgNPs nanocomposites showed better viral inhibition capacity than AgNPs or GO using a PRRSV pattern on the replication of virus.[153] Water-soluble graphene quantum dots (GQD) synthesized from MWCNT through oxidation and exfoliation with and without conjugated antiretroviral agents exhibited efficient viral inhibition of the HIV.[155] Very recently, graphene derivatives with long aliphatic chains have shown inhibition capacity against FCoV and SARS-CoV-2 replication.[156] Therefore, these results confirm the potential utilization of CBNs in the fight against viruses such as SARS-CoV-2.

Carbon-Based Nanomaterials Against Viruses

This current report analyzes the antiviral potentials of CBNs against 13 viruses. The antiviral activities were detected in 40 out of 44 studies as depicted in Figure 4.

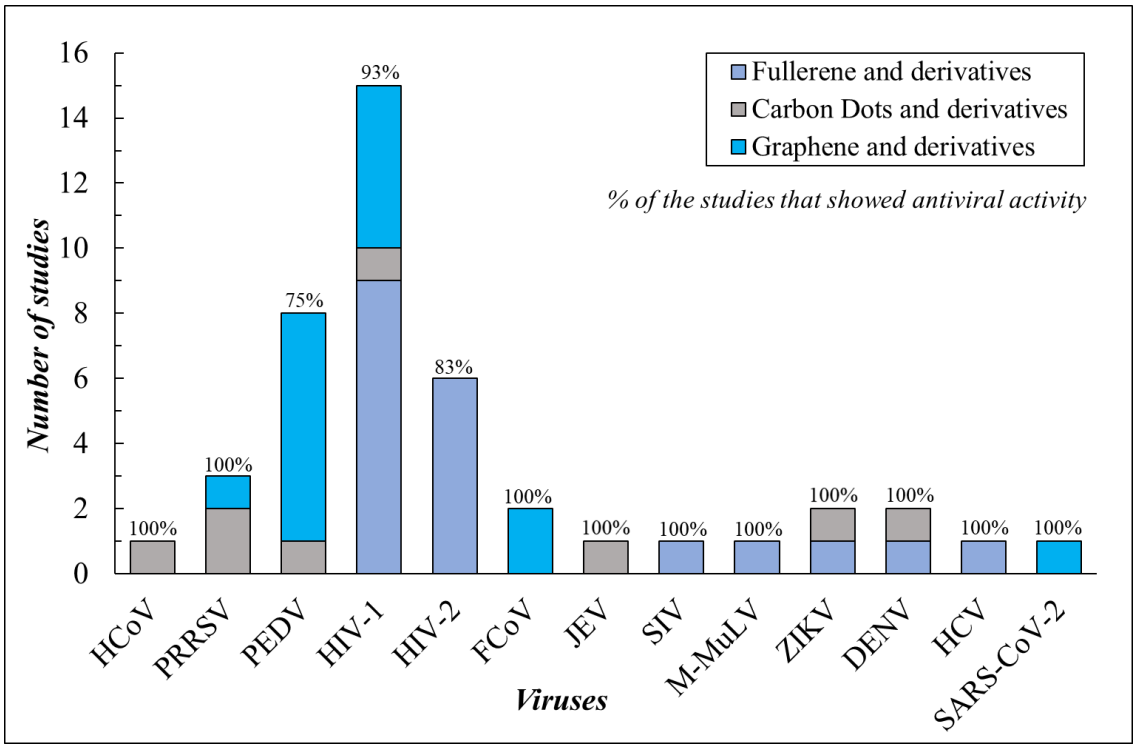

Figure. 4. Studies of carbon-based nanomaterials' antiviral activity indicating the percentage of studies that showed antiviral activity against 13 enveloped positive-sense single-stranded RNA viruses. The carbon-based nanomaterials were in the form of fullerenes, carbon dots, graphene and derivatives shown in Table 1.

The number of studies is greater than the 22 published papers shown in Table 1 because several CBNs and/or types of viruses were studied in some publications. To perform this count of studies, only CBNs with different chemical structural forms were considered. Thus, fullerene and its derivatives showed antiviral activity in nine studies against HIV-1,[26,136,138,140–142,144,145] five studies against HIV-2[26,140–142,144,145] out of six and against other viruses such as SIV,[139] M-MuLV,[139] HCV,[143] ZIKV[24] and DENV.[24] Carbon dots and their derivatives exhibited antiviral activity against HCoV,[22] HIV-1,[61] JEV,[147] ZIKV,[147] DENV,[147] PEDV[54] and two studies against PRRSV.[54,63] Graphene and related CBNs have shown antiviral activity against HIV-1 in four studies out of five,[148,149,155] five studies against PEDV out of seven,[28,153] two studies against FCoV[152,156] and one study against PRRSV[153] and SARS-CoV-2.[156] All these viruses are enveloped positive-sense single-stranded RNA viruses (IV Baltimore group[167]). Therefore, the antiviral properties of CBNs have been tested against a wide range of viruses similar to SARS-CoV-2, which suggests that CBNs are promising nanomaterials as alternative antiviral agents against this pathogen. The viruses tested against the CBNs are shown in Table 2 with all their characteristics such as name, abbreviation, genus, family, viral affection and disease/action and references.

Table 2. Information of the enveloped viruses tested to study the antiviral properties of CBNs belonging to the same Baltimore classification of SARS-CoV-2 (Group IV ((+)ssRNA[167]): single-stranded positive-sense RNA virus).

Virus name	Abbreviation	Genus	Family	Infects	Disease/action	References
Human coronavirus	HCoV	Alphacoronavirus	Coronaviridae	Humans	Common cold, pneumonia and bronchiolitis	22
Porcine reproductive and respiratory syndrome virus	PRRSV	Betaarterivirus	Arteriviridae	Pigs	Porcine reproductive and respiratory syndrome	54,63,153
Porcine epidemic diarrhea virus	PEDV	Aphacoronavirus	Coronaviridae	Pigs	Porcine diarrhea	28,54,153
Human immunodeficiency virus type 1	HIV-1	Lentivirus	Retroviridae	Humans	AIDS	26,61,136,140–142,148,149,155
Human immunodeficiency virus type 2	HIV-2	Lentivirus	Retroviridae	Humans	AIDS	26,140–142
Feline coronavirus	FCoV	Alphacoronavirus	Coronaviridae	Cats	Feline infectious peritonitis	152,156
Japanese encephalitis virus	JEV	Flavivirus	Flaviviridae	Humans through Culex mosquitoes	Inflammation of the brain occurs	147
Simian immunodeficiency virus	SIV	Lentivirus	Retroviridae	Non-human primates	Simian AIDS	139
Moloney murine leukemia virus	M-MuLV	Gammaretrovirus	Retroviridae	Mouse	Cancer	139
Zika virus	ZIKV	Flavivirus	Flaviviridae	Humans through Aedes mosquitoes	Zika fever	24,147
Dengue virus	DENV	Flavivirus	Flaviviridae	Humans through *Aedes* mosquitoes	Dengue fever	24,147
Hepatitis C virus	HCV	Hepacivirus	Flaviviridae	Humans	Hepatitis C	143
Severe acute respiratory syndrome coronavirus 2	SARS-CoV-2	Betacoronavirus	Coronaviridae	Humans	COVID-19	156

Mechanism of Action of CBNs against Viral Infection

The antiviral mechanisms of fullerenes, carbon dots, graphene and related carbon-nanomaterials are still not completely understood. However, some progress in understanding the mechanism of action of these promising antiviral agents has been achieved. Details of these suggested mechanisms are discussed below.

Fullerene and Derivatives

Antiviral fullerene derivatives can inhibit viral entry, modify its morphology and functions, and block viral replication.[30] Studies about the antiviral mechanism of action of C_{60}-fullerene derivatives against viruses such as HIV-1 and HIV-2 suggest that the C_{60} carbon sphere fits well to the active site of some HIV enzymes such as the HIV-protease.[136,138,144] In the same research line, a diamido diacid diphenyl fulleroid derivative (2c) was found to be an inhibitor of HIV- 1 and HIV-2 protease and reverse transcriptase at low micromolar concentration.[144] More recently, the series of fullerene derivatives shown in Figure 3 exhibited potential inhibition of hepatitis C virus (HCV) NS5B polymerase and HCV NS3/4 protease.[143] On the other hand, the mechanism of aqueous fullerene preparations of C_{60}-fullerene combined with PVP (C_{60}/PVP) against influenza A virus has been reported.[168] Influenza A virus is a negative-sense single-stranded virus that belongs to the Baltimore group V.[167] However, it is an enveloped RNA virus like SARS-CoV-2. The antiviral mechanism of action of the C_{60}/PVP complexes are attributed mainly to the lipid component of virus membranes as a membranotropic agent.[168] Thus, the round and oval morphology of the vial particles before being in contact with the C_{60}/PVP preparation was observed by transmission electron microscopy. The well-defined surface glycoproteins in the form of "brush" lost their integrity of viral envelopes when they were in contact with the C_{60}/PVP complexes by breaking the lipoprotein envelope structure and these complexes fused with the virion-like aggregates.

Carbon Dots and Derivatives

In the context of antiviral mechanisms, CDs could inhibit viral replication by activating the interferon response for the porcine reproductive and respiratory syndrome virus (PRRSV).[63] Furthermore, CDs conjugated with carboxyl phenylboronic acid (CBBA) prevented the entry of HIV-1 viruses into cells by suppressing syncytium (Figure 5).[61]

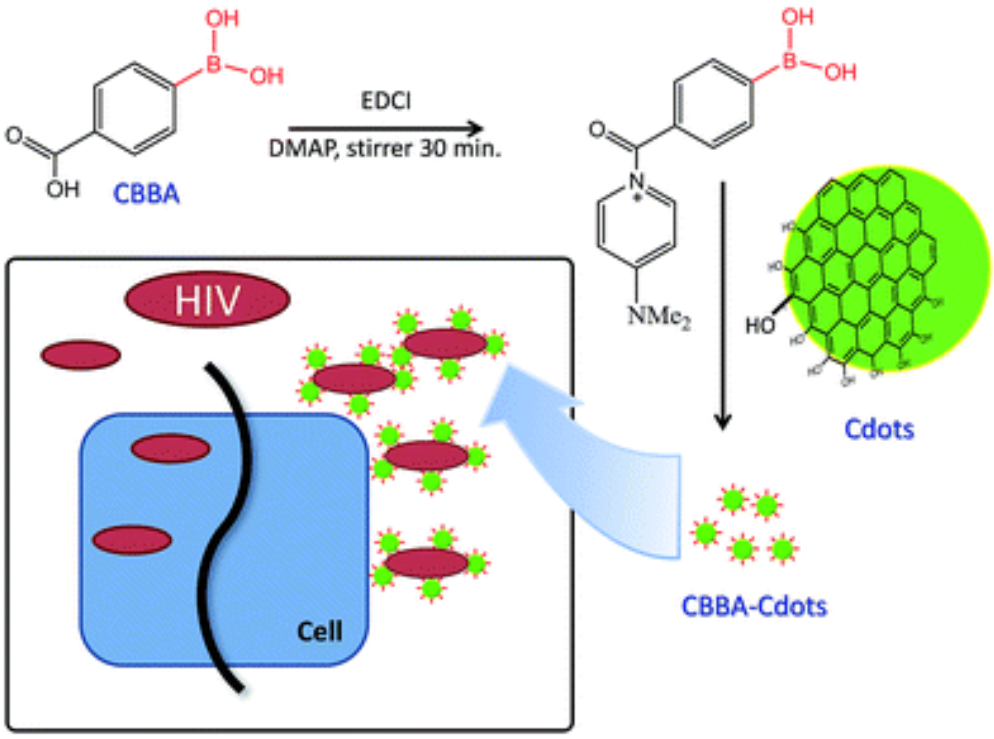

Figure 5. Schematic illustration of conjugating carboxyl phenylboronic acid (CBBA) on Cdots (CBBA-Cdots) and different mechanisms of inhibition entry - *Published by The (RSC)*. Reprinted with permission from ref 61. Copyright 2016 Royal Society of Chemistry.

The antiviral studies of functionalized CDs produced from 4-aminophenylboronic acid against human coronavirus (HCoV) infections in human Huh-7 liver cells showed inhibition of the viral entry and effects at the replication steps which was ascribed to the interaction of the CBNs' functional groups with the viral entry receptor DPP4[22] (see Figure 6).

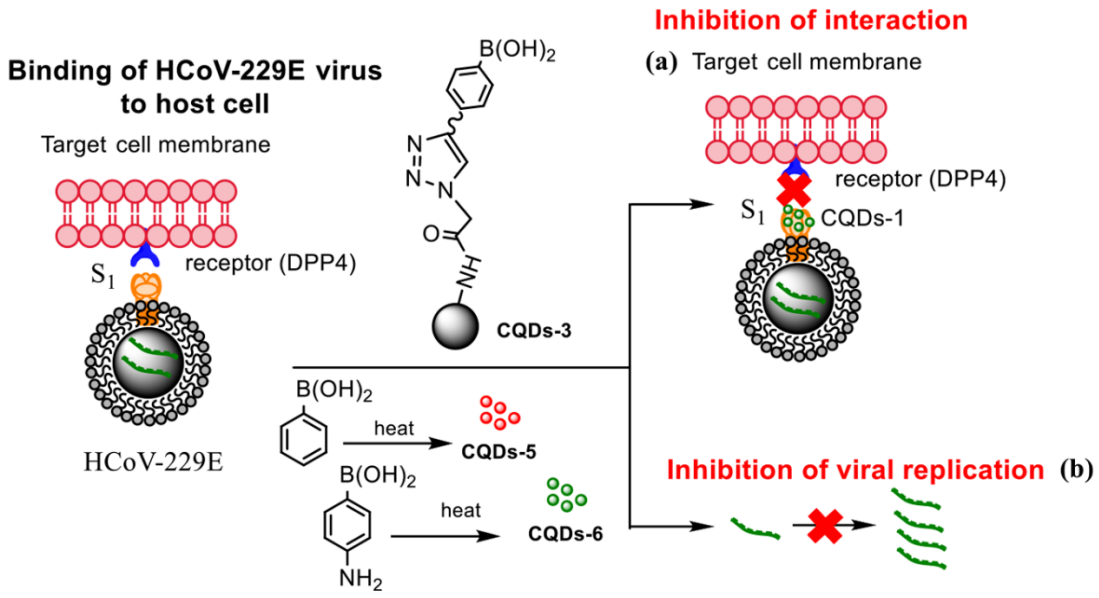

Figure 6. Influence of carbon quantum dots (CQDs) on the binding of HCoV229E virus to cells: (a) inhibition of protein S receptor interaction (b) inhibition of viral replication. Reprinted with permission from ref 22. Copyright 2019 American Chemical Society.

Graphene and Related Carbon-Based Nanomaterials

Graphene and GO can destroy the virus surface proteins, and extract their RNA by bioreduction.[165] The high binding affinity of graphene to the essential target proteins HIV Vpr, Nef, and Gag during HIV infections was reported in 2014.[148] Evaluation of the antiviral capacity of GO and rGO against PEDV showed significant viral inhibition by inactivating the virus before entering the cell (Figure 7).[28]

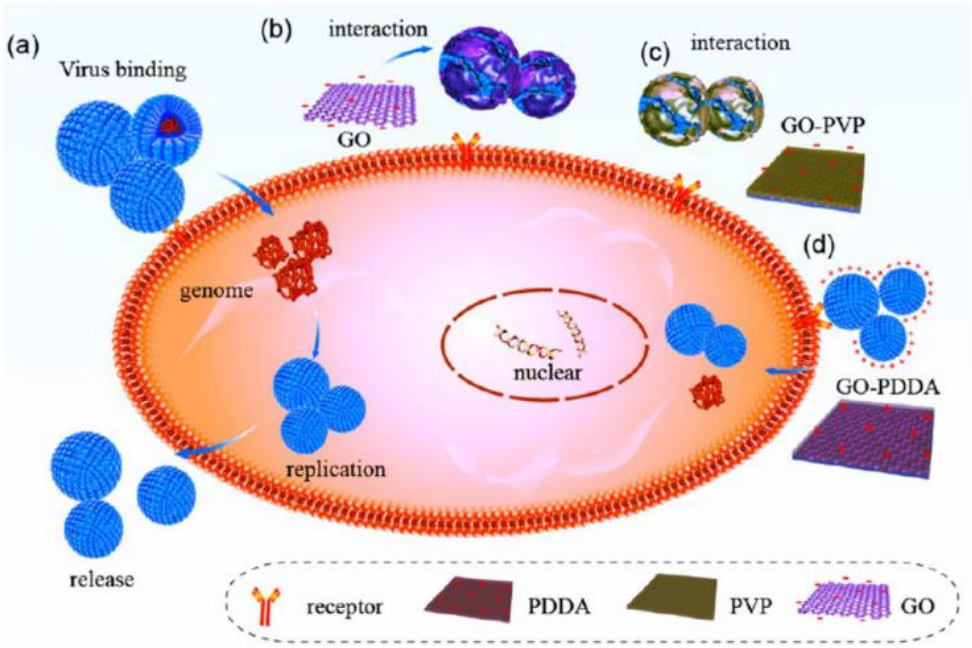

Figure 7. Possible antiviral mechanisms of graphene oxide (GO): (a) Infection initiation: virus binding by interaction with cell receptors; (b) Interaction of negatively charged GO nanosheet with the positively charged viruses, producing virus damage and infection inhibition; (c) GO conjugated with nonionic PVP blocked infection but GO with cationic PDDA did not (d). Reprinted with permission from ref 28. Copyright 2015 American Chemical Society.

It was reported that, the sharp edges of the GO or rGO nanosheet inactivated the virus by physical disruption of its biological structure through direct interaction which resulted in the outflow of intracellular metabolites.[28] Since rGO and GO exhibited a similar viral inhibition capacity in that study, the surface functional groups present in these CBNs may play a minor role. The antiviral activity was concentration and time-dependent incubation. Therefore, the potent antiviral activity of both GO and rGO can be attributed to the negative charge, which favors the electrostatic interaction with the positive charge of the virus and the single nanosheet-layer structure.[30] On the other hand, graphite (Gt) showed no viral inhibition and graphite oxide (GtO) exhibited weaker viral inhibition than monolayer GO and rGO, suggesting that the nanosheet form plays a significant role in the antiviral activity.[28] In the same research line, GO-AgNPs nanocomposites prevented PRRSV from entering the host cells (~59.2% inhibition) and improved the production of interferon-α (IFN-α) and ISGs, which directly block the proliferation of PRRSV.[153] Furthermore, graphene sheets with defined dual sulfate/alkyl functionalities have shown potent antiviral activity against FCoV and SARS-CoV-2.[156] Here, graphene

acted as a 2D platform to allow the interaction of the negatively charged polyglycerol sulfate (PGS) branches with the positively charged patches of the virus particles, and then the long aliphatic chains (C11) ruptured the membrane of the lipid envelop. Thus, G-PGS-C11 displayed the strongest antiviral activity against SARS-CoV-2 without exhibiting significant toxicity against eukaryotic host cells (Figure 8).

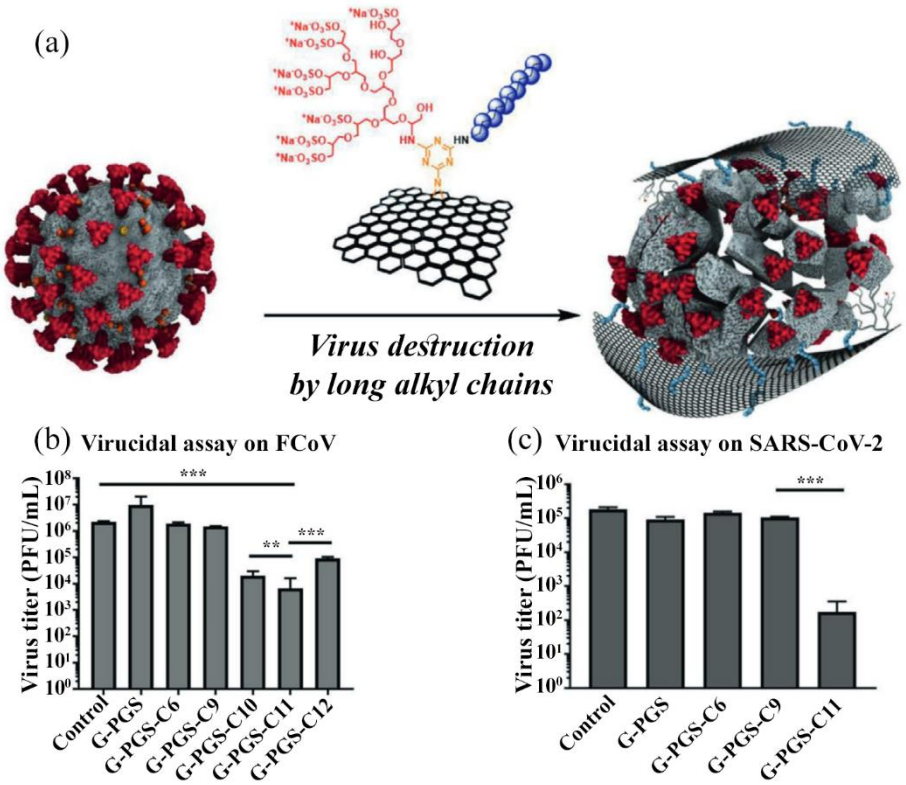

Figure 8. Schematic representation of virus rupturing by G-PGS-C11(a). Virucidal assays for the functionalized graphene platforms against FCoV (b) and SARS-CoV-2 (c). Values are expressed as mean ± standard deviation (n = 4). Adapted with permission under a Creative Commons CC BY 4.0 License from ref 156. Copyright 2021 Wiley-VCH GmbH.

Carbon-based nanomaterials may exercise multiple mechanisms against positive-sense single-stranded RNA viruses according to their physical configuration, chemical modifications, and metabolism. Hence, CBNs could work directly against the virus particle by distorting the envelope or the capsid organization; additionally, they may exert a steric hindrance effect by physically occupying a catalytic site of an essential viral enzyme or a receptor cavity.[137] When chemically modified with charged residues or metabolically activated, CBNs could perform both a direct disrupting effect on the virion structure and an indirect antiviral activity due to tuning of the redox signaling and the homeostatic innate/inflammatory response of the target cells.[30] All of the above functions, coupled with a negligible toxicity, may account for the anti-SARS-CoV-2 therapeutic potential of carbon compounds.[156] An even synergistically increased inhibitory effect may be played by CBNs when loaded with specific antiviral drugs.[30] Thus, the direct and intrinsic antiviral activities of the compounds plus the loaded antiviral drug will work synergistically against the viral particles which may increase the viral destruction potential.[169] Another characteristic feature of the CBNs is based on the immunostimulatory potentials,[170,171] which will derive and propagate the antiviral cell-mediated immunity, which also works synergistically with the CBNs-drug leading to

increase the virally infected cells and/or viral particle accessibility for CBN's or CBNs-drug achieving a more efficient viral infection clearance (Figure 9) through many suggested scenarios.[172]

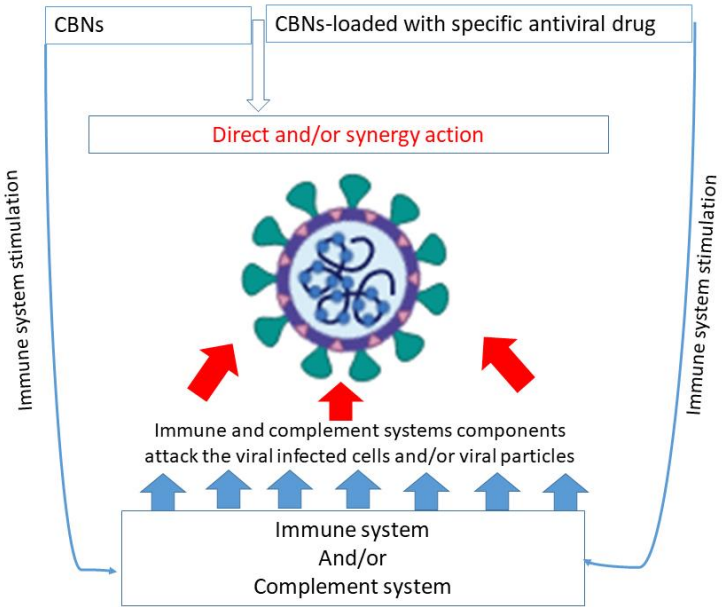

Figure 9. The suggested mechanism of action of CBNs against viral infection. The CBNs could work against viral and secondary infection in three scenarios, 1. CBNs alone or 2. in synergism with loaded antiviral drugs 3. and/or in synergism with the immune system components depending on the CBN's immunostimulatory potentials.

The most versatile interesting feature of CBNs is selecting one or more entities of them and functionalizing the selection base on the target function. The planning interactions between empty or loaded CBNs and the immune system, including the complement system, can be reciprocal in the context which immune system components would involve.[172,173]

Toxicological Aspects of Carbon-Based Nanomaterials

There are widespread concerns on the toxicological aspects of CBNs because some studies have reported that depending on the type of CBNs, dimensions, oxidation degree, functionalization, concentration, and exposure time, CBNs may exert cytotoxicity effects on host cells.[95–102] Nevertheless, we focused our attention on the toxicological studies performed with the CBNs tested against the 13 enveloped single-stranded positive-sense RNA viruses analyzed in this review (Table 1). Details of these studies are discussed below.

Fullerene and Derivatives

An anti- HIV-1 fullerene derivative (compound 1) showed no cytotoxicity on human PBM cells in 1993.[136] Another study reported non-cytotoxicity for concentrations up to 100 μM of an anti-HIV bis(monosuccinimide) derivative of p,p'-bis(2-aminoethyl)diphenyl-C_{60} in peripheral blood mononuclear (PBM) cells and H9 human embryonic stem cells, Vero, and CEM cells.[145] In 1996, nine anti-HIV functional derivatives of C60-fullerene showed no cytotoxicity for concentrations also up to 100 μM. However, a series of anti-HIV-1 bis-functionalized fullerene derivatives were

developed after that in 2003 and only one of them (derivative 1) exhibited moderate toxicity.[140] Nonetheless, low cytotoxic properties were reported for anti-HIV cationic fullerene derivatives and anti-HIV water-soluble fullerene derivatives in 2005 and 2007, respectively.[141,142] A later study in 2011 showed that anti-HIV derivatives of C_{70}-fullerene also exhibited low toxicity *in vitro* and *in vivo*.[26] More recently, an anti-HCV fullerene derivative (derivative 1a, Figure 3) showed no cytotoxicity in the low micromolar range.[25] Furthermore, very recently, anti-ZIKV and anti-DENV tridecafullerenes appended with up to 360 1,2- mannobiose showed no cytotoxicity in the picomolar range.[24]

Carbon Dots and Derivatives

CDs are highly promising nanomedicine tools for antiviral applications due to their low toxicity and potent antiviral activity.[158] Thus, CDs showed antiviral activity against PRRSV and low cytotoxicity in MARC-145 cells.[63] Furthermore, the modified CDs or their combination with other compounds significantly decreased the cytotoxicity of these carbon-based antiviral nanomaterials.[22,54,61,147] For example, in the cytotoxicity tests of both Cdots and CBBA-Cdots at high concentrations (up to 300 $mg{\cdot}mL^{-1}$), the proliferation of human cells was as fast as that of the untreated cells (control) after 24 hours incubation suggesting the absence of cytotoxic effects for both types of nanomaterials.[61] However, this study did not assess ROS or cytokine generation. Immunomodulatory effects are known to be induced by other carbon-based nanoparticles such as graphene (see the next paragraph). Anti-HCoV CDs produced from 4-aminophenylboronic acid, BZM-CDs with anti-JEV, anti-ZIKV and anti-DENV properties, and anti-PEDV and anti-PRRSV Gly-CDs manifested no cytotoxicity.[22,54,147]

Graphene and Related Carbon-Based Nanomaterials

The toxicological analysis of pristine MWCNT, ox-MWCNT and MWCNT-C-CHI36 with anti-HIV-1 properties showed opposite results.[149] Thus, ox-MWCNT and MWCNT-C-CHI36 showed low cytotoxicity contrary to pristine MWCNT which exhibited high cytotoxicity. However, GO showed potent anti-PEDV using a non-cytotoxic concentration (≤ 6 μg/mL).[28] Anti-FCoV GO-AgNPs nanocomposites and neat GO could also be prepared at very low or non-cytotoxic concentrations.[152] In the same way, anti-PRRSV GO-AgNPs nanocomposites could be prepared at a non-cytotoxic concentration (≤ 4 μg/mL).[153] Finally, a water-soluble GQD with a conjugated antiretroviral agent (GQD-CHI499) was selected as an excellent potential candidate due to its high antiviral activity against HIV and low cytotoxicity.[155] Anti-FCoV and anti-SARS-CoV-2 graphene platforms have shown a large concentration window without significant toxicity against human cells.[156] Therefore, the lack of toxicity could provide great potential in the development of safe therapeutics based on carbon-based nanomaterials to combat COVID-19 in the microbial resistant era.

Conclusions

Carbon-based nanomaterials have been evaluated for their antiviral activity against 13 enveloped viruses (HCoV, PRRSV, PEDV, HIV-1, HIV-2, FCoV, JEV, SIV, M-MuLV, ZIKV, DENV, HCV and SARS-CoV-2), all single-stranded positive-sense RNA viruses belonging to the Baltimore group IV. Most of the studies have shown a potent antiviral activity and from low to no toxicity supporting the potential for the use of CBNs to

combat SARS-CoV-2. As a revolutionary technology approach to treat COVID-19, these carbon-based therapeutics can provide a significant breakthrough as these nanomaterials allow the targeting of microbial resistance issues and can potentially induce tissue regeneration at the same time. Furthermore, these antimicrobial nanoweapons could be employed to deal with SARS-CoV-2 alone or with other types of viruses, bacteria, or fungi, causing pneumonia, including multidrug-resistant strains. The chance of success applying these wide-spectrum antimicrobial nanomaterials is very high because of the preliminary antiviral results reported for 13 viruses and the fact that the proposed approach could be extended to other types of pneumonia caused by other important pathogens.

Author Contributions

ÁSA conceived the idea of this work, wrote the draft manuscript and major editing. KT, ATM, MS, VNU, KL, PA, AAAA, GC, RK, AL, BDU, DB, EMR, NGB, and YKM edited the manuscript, and SSH, PPC, GP, MMT, TMAEA, SS, and AMB proof-read the manuscript.

Competing interests

The authors do not have any conflicts of interest to declare.

Vocabulary

Coronavirus disease 2019 (COVID-19), viral infectious disease that has spread throughout the world causing the current COVID-19 pandemic; **Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2)** is the causative pathogen responsible for the COVID-19 disease; **Carbon-based nanomaterials (CBNs)** are materials mainly constituted by carbon atoms that possess a broad range of antimicrobial properties; **Enveloped positive-sense single-stranded RNA viruses** are viruses that belong to the IV Baltimore group similar to SARS-CoV-2; **Pneumonia** is an infection of the lungs caused by bacteria, viruses or fungi.

References

(1) WHO. Coronavirus Disease (COVID-19) Pandemic https://www.who.int/emergencies/diseases/novel-coronavirus-2019.

(2) Verity, R.; Okell, L. C.; Dorigatti, I.; Winskill, P.; Whittaker, C.; Imai, N.; Cuomo-Dannenburg, G.; Thompson, H.; Walker, P. G. T.; Fu, H.; Dighe, A.; Griffin, J. T.; Baguelin, M.; Bhatia, S.; Boonyasiri, A.; Cori, A.; Cucunubá, Z.; FitzJohn, R.; Gaythorpe, K.; Green, W. *et al.* Estimates of the Severity of Coronavirus Disease 2019: A Model-Based Analysis. *Lancet Infect. Dis.* **2020**, *20* (6), 669–677. https://doi.org/10.1016/S1473-3099(20)30243-7.

(3) Chan, J. F. W.; Kok, K. H.; Zhu, Z.; Chu, H.; To, K. K. W.; Yuan, S.; Yuen, K. Y. Genomic Characterization of the 2019 Novel Human-Pathogenic Coronavirus Isolated from a Patient with Atypical Pneumonia after Visiting Wuhan. *Emerg. Microbes Infect.* **2020**, *9* (1), 221–236. https://doi.org/10.1080/22221751.2020.1719902.

(4) Lu, R.; Zhao, X.; Li, J.; Niu, P.; Yang, B.; Wu, H.; Wang, W.; Song, H.; Huang, B.; Zhu, N.; Bi, Y.; Ma, X.; Zhan, F.; Wang, L.; Hu, T.; Zhou, H.; Hu, Z.; Zhou, W.; Zhao, L.; Chen, J. *et al.* Genomic Characterisation and Epidemiology of 2019 Novel Coronavirus: Implications for Virus Origins and Receptor Binding. *Lancet* **2020**, *395* (10224), 565–574. https://doi.org/10.1016/S0140-6736(20)30251-8.

(5) Zhu, N.; Zhang, D.; Wang, W.; Li, X.; Yang, B.; Song, J.; Zhao, X.; Huang, B.; Shi, W.; Lu, R.; Niu, P.; Zhan, F.; Ma, X.; Wang, D.; Xu, W.; Wu, G.; Gao, G. F.; Tan, W. A Novel Coronavirus from Patients with Pneumonia in China, 2019. *N. Engl. J. Med.* **2020**, *382* (8), 727–733. https://doi.org/10.1056/NEJMoa2001017.

(6) Andersen, K. G.; Rambaut, A.; Lipkin, W. I.; Holmes, E. C.; Garry, R. F. The Proximal Origin of SARS-CoV-2. *Nat. Med.* **2020**, *26* (3), 450–452.

https://doi.org/10.1038/s41591-020-0820-9.

(7) Seyran, M.; Pizzol, D.; Adadi, P.; El-Aziz, T. M. A.; Hassan, S. S.; Soares, A.; Kandimalla, R.; Lundstrom, K.; Tambuwala, M.; Aljabali, A. A. A.; Lal, A.; Azad, G. K.; Choudhury, P. P.; Uversky, V. N.; Sherchan, S. P.; Uhal, B. D.; Rezaei, N.; Brufsky, A. M. Questions Concerning the Proximal Origin of SARS-CoV-2. *J. Med. Virol.* **2020**, *93* (3):1204-1206. jmv.26478. https://doi.org/10.1002/jmv.26478.

(8) Seyran, M.; Takayama, K.; Uversky, V. N.; Lundstrom, K.; Palù, G.; Sherchan, S. P.; Attrish, D.; Rezaei, N.; Aljabali, A. A. A.; Ghosh, S.; Pizzol, D.; Chauhan, G.; Adadi, P.; Mohamed Abd El-Aziz, T.; Soares, A. G.; Kandimalla, R.; Tambuwala, M.; Hassan, S. S.; Azad, G. K.; Pal Choudhury, P. *et al.* The Structural Basis of Accelerated Host Cell Entry by SARS-CoV-2. *FEBS J.* **2020**, Dec 2;10.1111, febs.15651. https://doi.org/10.1111/febs.15651.

(9) Hassan, S. S.; Ghosh, S.; Attrish, D.; Choudhury, P. P.; Aljabali, A. A. A.; Uhal, B. D.; Lundstrom, K.; Rezaei, N.; Uversky, V. N.; Seyran, M.; Pizzol, D.; Adadi, P.; Soares, A.; El-Aziz, T. M. A.; Kandimalla, R.; Tambuwala, M. M.; Azad, G. K.; Sherchan, S. P.; Baetas-da-Cruz, W.; Takayama, K. *et al.* Possible Transmission Flow of SARS-CoV-2 Based on ACE2 Features. *Molecules* **2020**, *25* (24), 5906. https://doi.org/10.3390/molecules25245906.

(10) Yang, X.; Yu, Y.; Xu, J.; Shu, H.; Xia, J.; Liu, H.; Wu, Y.; Zhang, L.; Yu, Z.; Fang, M.; Yu, T.; Wang, Y.; Pan, S.; Zou, X.; Yuan, S.; Shang, Y. Clinical Course and Outcomes of Critically Ill Patients with SARS-CoV-2 Pneumonia in Wuhan, China: A Single-Centered, Retrospective, Observational Study. *Lancet Respir. Med.* **2020**, *8* (5), 475–481. https://doi.org/10.1016/S2213-2600(20)30079-5.

(11) Rothberg, M. B.; Haessler, S. D.; Brown, R. B. Complications of Viral Influenza. *Am. J. Med.* **2008**, *121* (4), 258-64. https://doi.org/10.1016/j.amjmed.2007.10.040.

(12) Su, I. C.; Lee, K. L.; Liu, H. Y.; Chuang, H. C.; Chen, L. Y.; Lee, Y. J. Severe Community-Acquired Pneumonia Due to *Pseudomonas Aeruginosa* Coinfection in an Influenza A(H1N1)Pdm09 Patient. *J. Microbiol. Immunol. Infect.* **2019**, *52* (2), 365–366. https://doi.org/10.1016/j.jmii.2018.05.007.

(13) Chou, C. C.; Shen, C. F.; Chen, S. J.; Chen, H. M.; Wang, Y. C.; Chang, W. S.; Chang, Y. T.; Chen, W. Y.; Huang, C. Y.; Kuo, C. C.; Li, M. C.; Lin, J. F.; Lin, S. P.; Ting, S. W.; Weng, T. C.; Wu, P. S.; Wu, U. I.; Lin, P. C.; Lee, S. S. J.; Chen, Y. S. *et al.* Recommendations and Guidelines for the Treatment of Pneumonia in Taiwan. *J. Microbiol. Immunol. Infect.* **2019**, *52* (1), 172-199. https://doi.org/10.1016/j.jmii.2018.11.004.

(14) Lee, J. Y.; Yang, P. C.; Chang, C.; Lin, I. T.; Ko, W. C.; Cia, C. T. Community-Acquired Adenoviral and Pneumococcal Pneumonia Complicated by Pulmonary Aspergillosis in an Immunocompetent Adult. *J. Microbiol. Immunol. Infect.* **2019**, *52* (5), 838-839. https://doi.org/10.1016/j.jmii.2019.05.014.

(15) Albrich, W. C.; Rassouli, F.; Waldeck, F.; Berger, C.; Baty, F. Influence of Older Age and Other Risk Factors on Pneumonia Hospitalization in Switzerland in the Pneumococcal Vaccine Era. *Front. Med.* **2019**, *6*, *286*. https://doi.org/10.3389/fmed.2019.00286.

(16) Lundstrom, K. Coronavirus Pandemic-Therapy and Vaccines. *Biomedicines*. **2020**, *8* (5), 109. https://doi.org/10.3390/BIOMEDICINES8050109.

(17) Chauhan, G.; Madou, M. J.; Kalra, S.; Chopra, V.; Ghosh, D. Nanotechnology for COVID-19: Therapeutics and Vaccine Research. *ACS Nano*. **2020**, *14* (7),

7760-7782. https://doi.org/10.1021/acsnano.0c04006.

(18) Martí, M.; Tuñón-Molina, A.; Aachmann, F. L.; Muramoto, Y.; Noda, T.; Takayama, K.; Serrano-Aroca, Á. Protective Face Mask Filter Capable of Inactivating SARS-CoV-2, and Methicillin-Resistant *Staphylococcus Aureus* and *Staphylococcus Epidermidis*. *Polymers (Basel).* **2021**, *13* (2), 207. https://doi.org/10.3390/polym13020207.

(19) Feikin, D. R.; Schuchat, A.; Kolczak, M.; Barrett, N. L.; Harrison, L. H.; Lefkowitz, L.; McGeer, A.; Farley, M. M.; Vugia, D. J.; Lexau, C.; Stefonek, K. R.; Patterson, J. E.; Jorgensen, J. H. Mortality from Invasive Pneumococcal Pneumonia in the Era of Antibiotic Resistance, 1995-1997. *Am. J. Public Health* **2000**, *90* (2), 223–229. https://doi.org/10.2105/AJPH.90.2.223.

(20) Huttner, B.; Cappello, B.; Cooke, G.; Gandra, S.; Harbarth, S.; Imi, M.; Loeb, M.; Mendelson, M.; Moja, L.; Pulcini, C.; Sharland, M.; Tacconnelli, E.; Zeng, M.; Magrini, N. 2019 Community-Acquired Pneumonia Treatment Guidelines: There Is a Need for a Change toward More Parsimonious Antibiotic Use. *Am. J. Respir. Crit. Care Med.* **2020**, *201* (10), 1315–1316. https://doi.org/10.1164/rccm.201911-2226LE.

(21) Lal, A.; Akhtar, J.; Ullah, A.; Abraham, G. M. First Case of Pleural Empyema Caused by *Staphylococcus Simulans* : Review of the Literature. *Case Rep. Infect. Dis.* **2018**, *2018*, 1–5. https://doi.org/10.1155/2018/7831284.

(22) Łoczechin, A.; Séron, K.; Barras, A.; Giovanelli, E.; Belouzard, S.; Chen, Y. T.; Metzler-Nolte, N.; Boukherroub, R.; Dubuisson, J.; Szunerits, S. Functional Carbon Quantum Dots as Medical Countermeasures to Human Coronavirus. *ACS Appl. Mater. Interfaces* **2019**, *11* (46), 42964–42974. https://doi.org/10.1021/acsami.9b15032.

(23) Liu, X.; Pang, J.; Xu, F.; Zhang, X. Simple Approach to Synthesize Amino-Functionalized Carbon Dots by Carbonization of Chitosan. *Sci. Rep.* **2016**, *6* (1), 1–8. https://doi.org/10.1038/srep31100.

(24) Ramos-Soriano, J.; Reina, J. J.; Illescas, B. M.; De La Cruz, N.; Rodríguez-Pérez, L.; Lasala, F.; Rojo, J.; Delgado, R.; Martín, N. Synthesis of Highly Efficient Multivalent Disaccharide/[60]Fullerene Nanoballs for Emergent Viruses. *J. Am. Chem. Soc.* **2019**, *141* (38), 15403–15412. https://doi.org/10.1021/jacs.9b08003.

(25) Nakamura, S.; Mashino, T. Water-Soluble Fullerene Derivatives for Drug Discovery. *J. Nippon Med. Sch.* **2012**, *79* (4), 248–254. https://doi.org/10.1272/jnms.79.248.

(26) Kornev, A. B.; Peregudov, A. S.; Martynenko, V. M.; Balzarini, J.; Hoorelbeke, B.; Troshin, P. A. Synthesis and Antiviral Activity of Highly Water-Soluble Polycarboxylic Derivatives of [70]Fullerene. *Chem. Commun.* **2011**, *47* (29), 8298–8300. https://doi.org/10.1039/c1cc12209f.

(27) Tao, H.; Yang, K.; Ma, Z.; Wan, J.; Zhang, Y.; Kang, Z.; Liu, Z. *In Vivo* NIR Fluorescence Imaging, Biodistribution, and Toxicology of Photoluminescent Carbon Dots Produced from Carbon Nanotubes and Graphite. *Small* **2012**, *8* (2), 281–290. https://doi.org/10.1002/smll.201101706.

(28) Ye, S.; Shao, K.; Li, Z.; Guo, N.; Zuo, Y.; Li, Q.; Lu, Z.; Chen, L.; He, Q.; Han, H. Antiviral Activity of Graphene Oxide: How Sharp Edged Structure and Charge Matter. *ACS Appl. Mater. Interfaces* **2015**, *7* (38), 21578–21579. https://doi.org/10.1021/acsami.5b06876.

(29) Salesa, B.; Martí, M.; Frígols, B.; Serrano-Aroca, Á. Carbon Nanofibers in Pure Form and in Calcium Alginate Composites Films: New Cost-Effective

Antibacterial Biomaterials against the Life-Threatening Multidrug-Resistant *Staphylococcus Epidermidis*. *Polymers (Basel).* **2019**, *11* (3), 453. https://doi.org/10.3390/polym11030453.

(30) Innocenzi, P.; Stagi, L. Carbon-Based Antiviral Nanomaterials: Graphene, C-Dots, and Fullerenes. A Perspective. *Chem. Sci.* **2020**, *11* (26), 6606–6622. https://doi.org/10.1039/d0sc02658a.

(31) Wang, X.; Liu, X.; Chen, J.; Han, H.; Yuan, Z. Evaluation and Mechanism of Antifungal Effects of Carbon Nanomaterials in Controlling Plant Fungal Pathogen. *Carbon N. Y.* **2014**, *68*, 798–806. https://doi.org/10.1016/j.carbon.2013.11.072.

(32) Titirici, M. M.; White, R. J.; Brun, N.; Budarin, V. L.; Su, D. S.; Del Monte, F.; Clark, J. H.; MacLachlan, M. J. Sustainable Carbon Materials. *Chem. Soc. Rev.* **2015**, *44* (1), 250–290. https://doi.org/10.1039/c4cs00232f.

(33) Du, Y.; Ge, J.; Li, Y.; Ma, P. X.; Lei, B. Biomimetic Elastomeric, Conductive and Biodegradable Polycitrate-Based Nanocomposites for Guiding Myogenic Differentiation and Skeletal Muscle Regeneration. *Biomaterials* **2018**, *157*, 40–50. https://doi.org/10.1016/j.biomaterials.2017.12.005.

(34) Tandon, B.; Magaz, A.; Balint, R.; Blaker, J. J.; Cartmell, S. H. Electroactive Biomaterials: Vehicles for Controlled Delivery of Therapeutic Agents for Drug Delivery and Tissue Regeneration. *Adv. Drug Deliv. Rev.* **2018**, *129*, 148–168. https://doi.org/10.1016/j.addr.2017.12.012.

(35) Rivera-Briso, A. L.; Aachmann, F. L.; Moreno-Manzano, V.; Serrano-Aroca, Á. Graphene Oxide Nanosheets *versus* Carbon Nanofibers: Enhancement of Physical and Biological Properties of Poly(3-Hydroxybutyrate-Co-3-Hydroxyvalerate) Films for Biomedical Applications. *Int. J. Biol. Macromol.* **2020**, *143*, 1000–1008. https://doi.org/10.1016/j.ijbiomac.2019.10.034.

(36) Wu, X.; Ding, S. J.; Lin, K.; Su, J. A Review on the Biocompatibility and Potential Applications of Graphene in Inducing Cell Differentiation and Tissue Regeneration. *J. Mater. Chem. B* **2017**, *5* (17), 3084–3102. https://doi.org/10.1039/c6tb03067j.

(37) Lu, Y.; Li, L.; Li, M.; Lin, Z.; Wang, L.; Zhang, Y.; Yin, Q.; Xia, H.; Han, G. Zero-Dimensional Carbon Dots Enhance Bone Regeneration, Osteosarcoma Ablation, and Clinical Bacterial Eradication. *Bioconjug. Chem.* **2018**, *29* (9), 2982–2993. https://doi.org/10.1021/acs.bioconjchem.8b00400.

(38) Zhao, Z.; Lu, K.; Mao, B.; Liu, S.; Trilling, M.; Huang, A.; Lu, M.; Lin, Y. The Interplay between Emerging Human Coronavirus Infections and Autophagy. *Emerg. Microbes Infect*. **2021**, *10* (1), 196–205. https://doi.org/10.1080/22221751.2021.1872353.

(39) Samavati, L.; Uhal, B. D. ACE2, Much More Than Just a Receptor for SARS-COV-2. *Front. Cell. Infect. Microbiol.* **2020**, *10*, 317. https://doi.org/10.3389/fcimb.2020.00317.

(40) Patra, T.; Meyer, K.; Geerling, L.; Isbell, T. S.; Hoft, D. F.; Brien, J.; Pinto, A. K.; Ray, R. B.; Ray, R. SARS-CoV-2 Spike Protein Promotes IL-6 Transsignaling by Activation of Angiotensin II Receptor Signaling in Epithelial Cells. *PLoS Pathog.* **2020**, *16* (12), e1009128. https://doi.org/10.1371/journal.ppat.1009128.

(41) Uhal, B. D. Cell Cycle Kinetics in the Alveolar Epithelium. *Am. J. Physiol.* **1997**, *272* (6 Pt 1), L1031-1045. https://doi.org/10.1152/ajplung.1997.272.6.l1031.

(42) WHO | High Levels of Antibiotic Resistance Found Worldwide, New Data Shows. **2018**, https://www.who.int/news/item/29-01-2018-high-levels-of-

antibiotic-resistance-found-worldwide-new-data-shows (accessed April 1, 2021)

(43) Smith, R. A.; Sidwell, R. W.; Robins, R. K. Antiviral Mechanisms of Action. *Annu. Rev. Pharmacol. Toxicol.* **1980**, *20* (1), 259–284. https://doi.org/10.1146/annurev.pa.20.040180.001355.

(44) John Bennett, R. D. M. J. B. *Mandell, Douglas, and Bennett's Principles and Practice of Infectious Diseases - 9th Edition*; Elsevier: Amsterdam, 2019.

(45) Wood, A. J. J.; Whitley, R. J.; Gnann, J. W. Acyclovir: A Decade Later. *N. Engl. J. Med.* **1992**, *327* (11), 782–789. https://doi.org/10.1056/nejm199209103271108.

(46) Chono, K.; Katsumata, K.; Kontani, T.; Kobayashi, M.; Sudo, K.; Yokota, T.; Konno, K.; Shimizu, Y.; Suzuki, H. ASP2151, a Novel Helicase-Primase Inhibitor, Possesses Antiviral Activity against Varicella-Zoster Virus and Herpes Simplex Virus Types 1 and 2. *J. Antimicrob. Chemother.* **2010**, *65* (8), 1733–1741. https://doi.org/10.1093/jac/dkq198.

(47) Liu, Y.; Wang, X.; Yang, F.; Yang, X. Excellent Antimicrobial Properties of Mesoporous Anatase TiO2 and Ag/TiO2 Composite Films. *Microporous Mesoporous Mater.* **2008**, *114* (1–3), 431–439. https://doi.org/10.1016/j.micromeso.2008.01.032.

(48) Jia, Z.; Shen, D.; Xu, W. Synthesis and Antibacterial Activities of Quaternary Ammonium Salt of Chitosan. *Carbohydr. Res.* **2001**, *333* (1), 1–6. https://doi.org/10.1016/S0008-6215(01)00112-4.

(49) Chongsiriwatana, N. P.; Patch, J. A.; Czyzewski, A. M.; Dohm, M. T.; Ivankin, A.; Gidalevitz, D.; Zuckermann, R. N.; Barron, A. E. Peptoids That Mimic the Structure, Function, and Mechanism of Helical Antimicrobial Peptides. *Proc. Natl. Acad. Sci.* **2008**, *105* (8), 2794–2799. https://doi.org/10.1073/pnas.0708254105.

(50) Chen, Y.; Mant, C. T.; Farmer, S. W.; Hancock, R. E. W.; Vasil, M. L.; Hodges, R. S. Rational Design of Alpha-Helical Antimicrobial Peptides with Enhanced Activities and Specificity/Therapeutic Index. *J. Biol. Chem.* **2005**, *280* (13), 12316–12329. https://doi.org/10.1074/jbc.M413406200.

(51) Porter, E. A.; Wang, X.; Lee, H. S.; Weisblum, B.; Gellman, S. H. Non-Haemolytic Beta-Amino-Acid Oligomers. *Nature* **2000**, *404* (6778), 565. https://doi.org/10.1038/35007145.

(52) Henriques, P. C.; Borges, I.; Pinto, A. M.; Magalhães, F. D.; Gonçalves, I. C. Fabrication and Antimicrobial Performance of Surfaces Integrating Graphene-Based Materials. *Carbon*. **2018**, *132*, 709–732. https://doi.org/10.1016/j.carbon.2018.02.027.

(53) Gomes, R. N.; Borges, I.; Pereira, A. T.; Maia, A. F.; Pestana, M.; Magalhães, F. D.; Pinto, A. M.; Gonçalves, I. C. Antimicrobial Graphene Nanoplatelets Coatings for Silicone Catheters. *Carbon N. Y.* **2018**, *139*, 635–647. https://doi.org/10.1016/j.carbon.2018.06.044.

(54) Tong, T.; Hu, H.; Zhou, J.; Deng, S.; Zhang, X.; Tang, W.; Fang, L.; Xiao, S.; Liang, J. Glycyrrhizic-Acid-Based Carbon Dots with High Antiviral Activity by Multisite Inhibition Mechanisms. *Small* **2020**, *16* (13), e1906206. https://doi.org/10.1002/smll.201906206.

(55) Li, T. T.; Zhong, Y.; Yan, M.; Zhou, W.; Xu, W.; Huang, S. Y.; Sun, F.; Lou, C. W.; Lin, J. H. Synergistic Effect and Characterization of Graphene/Carbon Nanotubes/Polyvinyl Alcohol/Sodium Alginate Nanofibrous Membranes Formed Using Continuous Needleless Dynamic Linear Electrospinning. *Nanomaterials* **2019**, *9* (5), 714. https://doi.org/10.3390/nano9050714.

(56) Shams, E.; Yeganeh, H.; Naderi-Manesh, H.; Gharibi, R.; Mohammad Hassan, Z. Polyurethane/Siloxane Membranes Containing Graphene Oxide Nanoplatelets as Antimicrobial Wound Dressings: *In Vitro* and *in Vivo* Evaluations. *J. Mater. Sci. Mater. Med.* **2017**, *28* (5), 75. https://doi.org/10.1007/s10856-017-5881-z.

(57) Perreault, F.; De Faria, A. F.; Nejati, S.; Elimelech, M. Antimicrobial Properties of Graphene Oxide Nanosheets: Why Size Matters. *ACS Nano* **2015**, *9* (7), 7226–7236. https://doi.org/10.1021/acsnano.5b02067.

(58) Wu, X.; Tan, S.; Xing, Y.; Pu, Q.; Wu, M.; Zhao, J. X. Graphene Oxide as an Efficient Antimicrobial Nanomaterial for Eradicating Multi-Drug Resistant Bacteria *in Vitro* and *in Vivo*. *Colloids Surf. B Biointerfaces* **2017**, *157*, 1–9. https://doi.org/10.1016/j.colsurfb.2017.05.024.

(59) Melo, S. F.; Neves, S. C.; Pereira, A. T.; Borges, I.; Granja, P. L.; Magalhães, F. D.; Gonçalves, I. C. Incorporation of Graphene Oxide into Poly(ε-Caprolactone) 3D Printed Fibrous Scaffolds Improves Their Antimicrobial Properties. *Mater. Sci. Eng. C* **2020**, *109*, 110537. https://doi.org/10.1016/j.msec.2019.110537.

(60) Shi, L.; Chen, J.; Teng, L.; Wang, L.; Zhu, G.; Liu, S.; Luo, Z.; Shi, X.; Wang, Y.; Ren, L. The Antibacterial Applications of Graphene and Its Derivatives. *Small* **2016**, *12* (31), 4165–4184. https://doi.org/10.1002/smll.201601841.

(61) Fahmi, M. Z.; Sukmayani, W.; Khairunisa, S. Q.; Witaningrum, A. M.; Indriati, D. W.; Matondang, M. Q. Y.; Chang, J. Y.; Kotaki, T.; Kameoka, M. Design of Boronic Acid-Attributed Carbon Dots on Inhibits HIV-1 Entry. *RSC Adv.* **2016**, *6* (95), 92996–93002. https://doi.org/10.1039/c6ra21062g.

(62) Dong, X.; Moyer, M. M.; Yang, F.; Sun, Y. P.; Yang, L. Carbon Dots' Antiviral Functions against Noroviruses. *Sci. Rep.* **2017**, *7* (1), 1–10. https://doi.org/10.1038/s41598-017-00675-x.

(63) Du, T.; Liang, J.; Dong, N.; Liu, L.; Fang, L.; Xiao, S.; Han, H. Carbon Dots as Inhibitors of Virus by Activation of Type I Interferon Response. *Carbon N. Y.* **2016**, *110*, 278–285. https://doi.org/10.1016/j.carbon.2016.09.032.

(64) Zou, X.; Zhang, L.; Wang, Z.; Luo, Y. Mechanisms of the Antimicrobial Activities of Graphene Materials. *J. Am. Chem. Soc.* **2016**, *138* (7), 2064–2077. https://doi.org/10.1021/jacs.5b11411.

(65) Tegou, E.; Magana, M.; Katsogridaki, A. E.; Ioannidis, A.; Raptis, V.; Jordan, S.; Chatzipanagiotou, S.; Chatzandroulis, S.; Ornelas, C.; Tegos, G. P. Terms of Endearment: Bacteria Meet Graphene Nanosurfaces. *Biomaterials*. **2016**, *89*, 38–55. https://doi.org/10.1016/j.biomaterials.2016.02.030.

(66) Lara, H. H.; Ayala-Nuñez, N. V.; Ixtepan-Turrent, L.; Rodriguez-Padilla, C. Mode of Antiviral Action of Silver Nanoparticles against HIV-1. *J. Nanobiotechnology* **2010**, *8* (1), 1–10. https://doi.org/10.1186/1477-3155-8-1.

(67) Fujimori, Y.; Sato, T.; Hayata, T.; Nagao, T.; Nakayam, M.; Nakayam, T.; Sugamat, R.; Suzuki, K. Novel Antiviral Characteristics of Nanosized Copper(i) Iodide Particles Showing Inactivation Activity against 2009 Pandemic H1N1 Influenza Virus. *Appl. Environ. Microbiol.* **2012**, *78* (4), 951–955. https://doi.org/10.1128/AEM.06284-11.

(68) Akhtar, S.; Shahzad, K.; Mushtaq, S.; Ali, I.; Rafe, M. H.; Fazal-Ul-Karim, S. M. Antibacterial and Antiviral Potential of Colloidal Titanium Dioxide (TiO2) Nanoparticles Suitable for Biological Applications. *Mater. Res. Express* **2019**, *6* (10), 105409. https://doi.org/10.1088/2053-1591/ab3b27.

(69) Ghaffari, H.; Tavakoli, A.; Moradi, A.; Tabarraei, A.; Bokharaei-Salim, F.; Zahmatkeshan, M.; Farahmand, M.; Javanmard, D.; Kiani, S. J.; Esghaei, M.; Pirhajati-Mahabadi, V.; Ataei-Pirkooh, A.; Monavari, S. H. Inhibition of H1N1

Influenza Virus Infection by Zinc Oxide Nanoparticles: Another Emerging Application of Nanomedicine. *J. Biomed. Sci.* **2019**, *26* (1), 70. https://doi.org/10.1186/s12929-019-0563-4.

(70) Cooksey, D. A. Molecular Mechanisms of Copper Resistance and Accumulation in Bacteria. *FEMS Microbiol. Rev.* **1994**, *14* (4), 381–386. https://doi.org/10.1111/j.1574-6976.1994.tb00112.x.

(71) Li, X. Z.; Nikaido, H.; Williams, K. E. Silver-Resistant Mutants of *Escherichia Coli* Display Active Efflux of Ag+ and Are Deficient in Porins. *J. Bacteriol.* **1997**, *179* (19), 6127–6132. https://doi.org/10.1128/CMR.00043-12.

(72) Salas Orozco, M. F.; Niño-Martínez, N.; Martínez-Castañón, G. A.; Méndez, F. T.; Ruiz, F. Molecular Mechanisms of Bacterial Resistance to Metal and Metal Oxide Nanoparticles. *Int. J. Mol. Sci.* **2019**, *20* (11), 2808. https://doi.org/10.3390/ijms20112808.

(73) Rosen, B. P. Bacterial Resistance to Heavy Metals and Metalloids. *J.Biol Inorg. Chem.***1996**, *1*, 273–277. https://doi.org/10.1007/s007750050053.

(74) Gunawan, C.; Marquis, C. P.; Amal, R.; Sotiriou, G. A.; Rice, S. A.; Harry, E. J. Widespread and Indiscriminate Nanosilver Use: Genuine Potential for Microbial Resistance. *ACS Nano*. **2017**, *11* (4), 3438–3445. https://doi.org/10.1021/acsnano.7b01166.

(75) Choudhury, R.; Srivastava, S. Zinc Resistance Mechanisms in Bacteria. *Curr. Microbiol*. **2001**, *43* (5), 316-321. https://doi.org/10.2307/24106396.

(76) Yazdankhah, S.; Rudi, K.; Bernhoft, A. Zinc and Copper in Animal Feed – Development of Resistance and Co-Resistance to Antimicrobial Agents in Bacteria of Animal Origin. *Microb. Ecol. Heal. Dis.* **2014**, *25* (0). https://doi.org/10.3402/mehd.v25.25862.

(77) Bondarenko, O.; Juganson, K.; Ivask, A.; Kasemets, K.; Mortimer, M.; Kahru, A. Toxicity of Ag, CuO and ZnO Nanoparticles to Selected Environmentally Relevant Test Organisms and Mammalian Cells *in Vitro*: A Critical Review. *Arch. Toxicol.* **2013**, *87* (7), 1181–1200. https://doi.org/10.1007/s00204-013-1079-4.

(78) Ahamed, M.; AlSalhi, M. S.; Siddiqui, M. K. J. Silver Nanoparticle Applications and Human Health. *Clin. Chim. Acta*. **2010**, 411 (23-24), 1841–1848. https://doi.org/10.1016/j.cca.2010.08.016.

(79) Wätjen, W.; Haase, H.; Biagioli, M.; Beyersmann, D. Induction of Apoptosis in Mammalian Cells by Cadmium and Zinc. *Environ. Health Perspect.* **2002**, *110* (SUPPL. 5), 865–867. https://doi.org/10.1289/ehp.02110s5865.

(80) Frígols, B.; Martí, M.; Hernández-Oliver, C.; Aarstad, O.; Teialeret Ulset, A.-S.; Inger Sætrom, G.; Lillelund Aachmann, F.; Serrano-Aroca, Á. Graphene Oxide in Zinc Alginate Films: Antibacterial Activity, Water Sorption, Wettability and Opacity. PLoS One. **2019**, *14* (3), e0212819. 10.1371/journal.pone.0212819

(81) Fahmy, B.; Cormier, S. A. Copper Oxide Nanoparticles Induce Oxidative Stress and Cytotoxicity in Airway Epithelial Cells. *Toxicol. Vitr.* **2009**, *23* (7), 1365–1371. https://doi.org/10.1016/j.tiv.2009.08.005.

(82) Yousefi, M.; Dadashpour, M.; Hejazi, M.; Hasanzadeh, M.; Behnam, B.; de la Guardia, M.; Shadjou, N.; Mokhtarzadeh, A. Anti-Bacterial Activity of Graphene Oxide as a New Weapon Nanomaterial to Combat Multidrug-Resistance Bacteria. *Mater Sci Eng C Mater Biol Appl.* **2017**, *75*, 568–581. https://doi.org/10.1016/j.msec.2016.12.125.

(83) Mejías Carpio, I. E.; Santos, C. M.; Wei, X.; Rodrigues, D. F. Toxicity of a Polymer–Graphene Oxide Composite against Bacterial Planktonic Cells,

Biofilms, and Mammalian Cells. *Nanoscale* **2012**, *4* (15), 4746-4756. https://doi.org/10.1039/c2nr30774j.

(84) Ye, S.; Shao, K.; Li, Z.; Guo, N.; Zuo, Y.; Li, Q.; Lu, Z.; Chen, L.; He, Q.; Han, H. Antiviral Activity of Graphene Oxide: How Sharp Edged Structure and Charge Matter. *ACS Appl. Mater. Interfaces* **2015**, *7* (38), 21578–21579. https://doi.org/10.1021/acsami.5b06876.

(85) Martí, M.; Frígols, B.; Salesa, B.; Serrano-Aroca, Á. Calcium Alginate/Graphene Oxide Films: Reinforced Composites Able to Prevent *Staphylococcus Aureus* and Methicillin-Resistant *Staphylococcus Epidermidis* Infections with No Cytotoxicity for Human Keratinocyte HaCaT Cells. *Eur. Polym. J.* **2019**, *110*, 14-21. https://doi.org/10.1016/j.eurpolymj.2018.11.012.

(86) Zhang, X.; Yin, J.; Peng, C.; Hu, W.; Zhu, Z.; Li, W.; Fan, C.; Huang, Q. Distribution and Biocompatibility Studies of Graphene Oxide in Mice after Intravenous Administration. *Carbon N. Y.* **2011**, *49* (3), 986–995. https://doi.org/10.1016/j.carbon.2010.11.005.

(87) Drasler, B.; Kucki, M.; Delhaes, F.; Buerki-Thurnherr, T.; Vanhecke, D.; Korejwo, D.; Chortarea, S.; Barosova, H.; Hirsch, C.; Petri-Fink, A.; Rothen-Rutishauser, B.; Wick, P. Single Exposure to Aerosolized Graphene Oxide and Graphene Nanoplatelets Did Not Initiate an Acute Biological Response in a 3D Human Lung Model. *Carbon N. Y.* **2018**, *137*, 125–135. https://doi.org/10.1016/j.carbon.2018.05.012.

(88) Kurapati, R.; Mukherjee, S. P.; Martín, C.; Bepete, G.; Vázquez, E.; Pénicaud, A.; Fadeel, B.; Bianco, A. Degradation of Single-Layer and Few-Layer Graphene by Neutrophil Myeloperoxidase. *Angew. Chemie - Int. Ed.* **2018**, *57* (36), 11722–11727. https://doi.org/10.1002/anie.201806906.

(89) Kurapati, R.; Russier, J.; Squillaci, M. A.; Treossi, E.; Ménard-Moyon, C.; Del Rio-Castillo, A. E.; Vazquez, E.; Samorì, P.; Palermo, V.; Bianco, A. Dispersibility-Dependent Biodegradation of Graphene Oxide by Myeloperoxidase. *Small* **2015**, *11* (32), 3985–3994. https://doi.org/10.1002/smll.201500038.

(90) Li, D.; Hu, X.; Zhang, S. Biodegradation of Graphene-Based Nanomaterials in Blood Plasma Affects Their Biocompatibility, Drug Delivery, Targeted Organs and Antitumor Ability. *Biomaterials* **2019**, *202*, 12–25. https://doi.org/10.1016/j.biomaterials.2019.02.020.

(91) Girish, C. M.; Sasidharan, A.; Gowd, G. S.; Nair, S.; Koyakutty, M. Confocal Raman Imaging Study Showing Macrophage Mediated Biodegradation of Graphene *in Vivo*. *Adv. Healthc. Mater.* **2013**, *2* (11), 1489–1500. https://doi.org/10.1002/adhm.201200489.

(92) Kotchey, G. P.; Allen, B. L.; Vedala, H.; Yanamala, N.; Kapralov, A. A.; Tyurina, Y. Y.; Klein-Seetharaman, J.; Kagan, V. E.; Star, A. The Enzymatic Oxidation of Graphene Oxide. *ACS Nano* **2011**, *5* (3), 2098–2108. https://doi.org/10.1021/nn103265h.

(93) Mukherjee, S. P.; Gliga, A. R.; Lazzaretto, B.; Brandner, B.; Fielden, M.; Vogt, C.; Newman, L.; Rodrigues, A. F.; Shao, W.; Fournier, P. M.; Toprak, M. S.; Star, A.; Kostarelos, K.; Bhattacharya, K.; Fadeel, B. Graphene Oxide Is Degraded by Neutrophils and the Degradation Products Are Non-Genotoxic. *Nanoscale* **2018**, *10* (3), 1180–1188. https://doi.org/10.1039/c7nr03552g.

(94) Liu, J. H.; Yang, S. T.; Wang, H.; Chang, Y.; Cao, A.; Liu, Y. Effect of Size and Dose on the Biodistribution of Graphene Oxide in Mice. *Nanomedicine* **2012**, *7* (12), 1801–1812. https://doi.org/10.2217/nnm.12.60.

(95) Pinto, A. M.; Gonçalves, I. C.; Magalhães, F. D. Graphene-Based Materials Biocompatibility: A Review. *Colloids Surf.B: Biointerfaces*. **2013**, *111*, 188–202. https://doi.org/10.1016/j.colsurfb.2013.05.022.

(96) Piperno, A.; Scala, A.; Mazzaglia, A.; Neri, G.; Pennisi, R.; Sciortino, M. T.; Grassi, G. Cellular Signaling Pathways Activated by Functional Graphene Nanomaterials. *Int. J. Mol. Sci.* **2018**, *19* (11), 3365. https://doi.org/10.3390/ijms19113365.

(97) Zhang, X.; Zhou, Q.; Zou, W.; Hu, X. Molecular Mechanisms of Developmental Toxicity Induced by Graphene Oxide at Predicted Environmental Concentrations. *Environ. Sci. Technol.* **2017**, *51* (14), 7861–7871. https://doi.org/10.1021/acs.est.7b01922.

(98) Martinez Paino, I. M.; Santos, F.; Zucolotto, V. Biocompatibility and Toxicology Effects of Graphene Oxide in Cancer, Normal, and Primary Immune Cells. *J. Biomed. Mater. Res. - Part A* **2017**, *105* (3), 728–736. https://doi.org/10.1002/jbm.a.35946.

(99) Fadeel, B.; Bussy, C.; Merino, S.; Vázquez, E.; Flahaut, E.; Mouchet, F.; Evariste, L.; Gauthier, L.; Koivisto, A. J.; Vogel, U.; Martín, C.; Delogu, L. G.; Buerki-Thurnherr, T.; Wick, P.; Beloin-Saint-Pierre, D.; Hischier, R.; Pelin, M.; Candotto Carniel, F.; Tretiach, M.; Cesca, F.; Benfenati, F.; Scaini, D.; Ballerini, L.; Kostarelos, K.; Prato, M.; Bianco, A. Safety Assessment of Graphene-Based Materials: Focus on Human Health and the Environment. *ACS Nano*. **2018**, *12* (11), 10582–10620. https://doi.org/10.1021/acsnano.8b04758.

(100) Pinto, A. M.; Goncąlves, C.; Sousa, D. M.; Ferreira, A. R.; Moreira, J. A.; Goncąlves, I. C.; Magalhães, F. D. Smaller Particle Size and Higher Oxidation Improves Biocompatibility of Graphene-Based Materials. *Carbon N. Y.* **2016**, *99*, 318–329. https://doi.org/10.1016/j.carbon.2015.11.076.

(101) Cheng, C.; Nie, S.; Li, S.; Peng, H.; Yang, H.; Ma, L.; Sun, S.; Zhao, C. Biopolymer Functionalized Reduced Graphene Oxide with Enhanced Biocompatibility *via* Mussel Inspired Coatings/Anchors. *J. Mater. Chem. B* **2013**, *1* (3), 265–275. https://doi.org/10.1039/c2tb00025c.

(102) Freixa, A.; Acuña, V.; Sanchís, J.; Farré, M.; Barceló, D.; Sabater, S. Ecotoxicological Effects of Carbon Based Nanomaterials in Aquatic Organisms. *Sci.Total Environ.* **2018**, *619-620*, 328–337. https://doi.org/10.1016/j.scitotenv.2017.11.095.

(103) Hu, W.; Peng, C.; Lv, M.; Li, X.; Zhang, Y.; Chen, N.; Fan, C.; Huang, Q. Protein Corona-Mediated Mitigation of Cytotoxicity of Graphene Oxide. *ACS Nano* **2011**, *5* (5), 3693–3700. https://doi.org/10.1021/nn200021j.

(104) Chong, Y.; Ge, C.; Yang, Z.; Garate, J. A.; Gu, Z.; Weber, J. K.; Liu, J.; Zhou, R. Reduced Cytotoxicity of Graphene Nanosheets Mediated by Blood-Protein Coating. *ACS Nano* **2015**, *9* (6), 5713–5724. https://doi.org/10.1021/nn5066606.

(105) Duan, G.; Kang, S. G.; Tian, X.; Garate, J. A.; Zhao, L.; Ge, C.; Zhou, R. Protein Corona Mitigates the Cytotoxicity of Graphene Oxide by Reducing Its Physical Interaction with Cell Membrane. *Nanoscale* **2015**, *7* (37), 15214–15224. https://doi.org/10.1039/c5nr01839k.

(106) Li, Y.; Feng, L.; Shi, X.; Wang, X.; Yang, Y.; Yang, K.; Liu, T.; Yang, G.; Liu, Z. Surface Coating-Dependent Cytotoxicity and Degradation of Graphene Derivatives: Towards the Design of Non-Toxic, Degradable Nano-Graphene. *Small* **2014**, *10* (8), 1544–1554. https://doi.org/10.1002/smll.201303234.

(107) Wibroe, P. P.; Petersen, S. V.; Bovet, N.; Laursen, B. W.; Moghimi, S. M. Soluble and Immobilized Graphene Oxide Activates Complement System

Differently Dependent on Surface Oxidation State. *Biomaterials* **2016**, *78*, 20–26. https://doi.org/10.1016/j.biomaterials.2015.11.028.

(108) Orecchioni, M.; Ménard-Moyon, C.; Delogu, L. G.; Bianco, A. Graphene and the Immune System: Challenges and Potentiality. *Adv. Drug Deliv. Rev.* **2016**, *105* (Pt B), 63–175. https://doi.org/10.1016/j.addr.2016.05.014.

(109) Orecchioni, M.; Jasim, D. A.; Pescatori, M.; Manetti, R.; Fozza, C.; Sgarrella, F.; Bedognetti, D.; Bianco, A.; Kostarelos, K.; Delogu, L. G. Molecular and Genomic Impact of Large and Small Lateral Dimension Graphene Oxide Sheets on Human Immune Cells from Healthy Donors. *Adv. Healthc. Mater.* **2016**, *5* (2), 276–287. https://doi.org/10.1002/adhm.201500606.

(110) Ma, J.; Liu, R.; Wang, X.; Liu, Q.; Chen, Y.; Valle, R. P.; Zuo, Y. Y.; Xia, T.; Liu, S. Crucial Role of Lateral Size for Graphene Oxide in Activating Macrophages and Stimulating Pro-Inflammatory Responses in Cells and Animals. *ACS Nano* **2015**, *9* (10), 10498–10515. https://doi.org/10.1021/acsnano.5b04751.

(111) Russier, J.; Treossi, E.; Scarsi, A.; Perrozzi, F.; Dumortier, H.; Ottaviano, L.; Meneghetti, M.; Palermo, V.; Bianco, A. Evidencing the Mask Effect of Graphene Oxide: A Comparative Study on Primary Human and Murine Phagocytic Cells. *Nanoscale* **2013**, *5* (22), 11234–11247. https://doi.org/10.1039/c3nr03543c.

(112) Li, Y.; Liu, Y.; Fu, Y.; Wei, T.; Le Guyader, L.; Gao, G.; Liu, R. S.; Chang, Y. Z.; Chen, C. The Triggering of Apoptosis in Macrophages by Pristine Graphene through the MAPK and TGF-Beta Signaling Pathways. *Biomaterials* **2012**, *33* (2), 402–411. https://doi.org/10.1016/j.biomaterials.2011.09.091.

(113) Zhou, H.; Zhao, K.; Li, W.; Yang, N.; Liu, Y.; Chen, C.; Wei, T. The Interactions between Pristine Graphene and Macrophages and the Production of Cytokines/Chemokines *via* TLR- and NF-KB-Related Signaling Pathways. *Biomaterials* **2012**, *33* (29), 6933–6942. https://doi.org/10.1016/j.biomaterials.2012.06.064.

(114) Mukherjee, S. P.; Bottini, M.; Fadeel, B. Graphene and the Immune System: A Romance of Many Dimensions. *Front. Immunol.* **2017**, *8*, 673. https://doi.org/10.3389/fimmu.2017.00673.

(115) Sasidharan, A.; Panchakarla, L. S.; Sadanandan, A. R.; Ashokan, A.; Chandran, P.; Girish, C. M.; Menon, D.; Nair, S. V.; Rao, C. N. R.; Koyakutty, M. Hemocompatibility and Macrophage Response of Pristine and Functionalized Graphene. *Small* **2012**, *8* (8), 1251–1263. https://doi.org/10.1002/smll.201102393.

(116) Kenry; Lee, W. C.; Loh, K. P.; Lim, C. T. When Stem Cells Meet Graphene: Opportunities and Challenges in Regenerative Medicine. *Biomaterials*. **2018**, *155*, 236–250. https://doi.org/10.1016/j.biomaterials.2017.10.004.

(117) Uversky, V. N.; Elrashdy, F.; Aljadawi, A.; Ali, S. M.; Khan, R. H.; Redwan, E. M. Severe Acute Respiratory Syndrome Coronavirus 2 Infection Reaches the Human Nervous System: How? *J. Neurosci. Res.* **2021**, *99* (3), 750–777. https://doi.org/10.1002/jnr.24752.

(118) Xue, D.; Chen, E.; Zhong, H.; Zhang, W.; Wang, S.; Joomun, M. U.; Yao, T.; Tan, Y.; Lin, S.; Zheng, Q.; Pan, Z. Immunomodulatory Properties of Graphene Oxide for Osteogenesis and Angiogenesis. *Int. J. Nanomedicine* **2018**, *13*, 5799–5810. https://doi.org/10.2147/IJN.S170305.

(119) Li, T.; Zhang, T. The Application of Nanomaterials in Angiogenesis. *Curr. Stem Cell Res. Ther.* **2020**, *16* (1), 74–82.

https://doi.org/10.2174/1574888x15666200211102203.

(120) Shao, D.; Lu, M.; Xu, D.; Zheng, X.; Pan, Y.; Song, Y.; Xu, J.; Li, M.; Zhang, M.; Li, J.; Chi, G.; Chen, L.; Yang, B. Carbon Dots for Tracking and Promoting the Osteogenic Differentiation of Mesenchymal Stem Cells. *Biomater. Sci.* **2017**, *5* (9), 1820–1827. https://doi.org/10.1039/c7bm00358g.

(121) Leng, Z.; Zhu, R.; Hou, W.; Feng, Y.; Yang, Y.; Han, Q.; Shan, G.; Meng, F.; Du, D.; Wang, S.; Fan, J.; Wang, W.; Deng, L.; Shi, H.; Li, H.; Hu, Z.; Zhang, F.; Gao, J.; Liu, H.; Li, X. *et al.* Transplantation of ACE2- Mesenchymal Stem Cells Improves the Outcome of Patients with Covid-19 Pneumonia. *Aging Dis.* **2020**, *11* (2), 216–228. https://doi.org/10.14336/AD.2020.0228.

(122) Shi, L.; Huang, H.; Lu, X.; Yan, X.; Jiang, X.; Xu, R.; Wang, S.; Zhang, C.; Yuan, X.; Xu, Z.; Huang, L.; Fu, J.-L.; Li, Y.; Zhang, Y.; Yao, W.-Q.; Liu, T.; Song, J.; Sun, L.; Yang, F.; Zhang, X. *et al.* Effect of Human Umbilical Cord-Derived Mesenchymal Stem Cells on Lung Damage in Severe COVID-19 Patients: A Randomized, Double-Blind, Placebo-Controlled Phase 2 Trial. *Signal Transduct. Target. Ther.* **2021**, *6* (1), 1–9. https://doi.org/10.1038/s41392-021-00488-5.

(123) Pinky; Gupta, S.; Krishnakumar, V.; Sharma, Y.; Dinda, A. K.; Mohanty, S. Mesenchymal Stem Cell Derived Exosomes: A Nano Platform for Therapeutics and Drug Delivery in Combating COVID-19. *Stem Cell Rev Reports*. 2020, 17 (1), 33–43. https://doi.org/10.1007/s12015-020-10002-z.

(124) Khavinson, V. K.; Kuznik, B. I.; Trofimova, S. V.; Volchkov, V. A.; Rukavishnikova, S. A.; Titova, O. N.; Akhmedov, T. A.; Trofimov, A. V.; Potemkin, V. V.; Magen, E. Results and Prospects of Using Activator of Hematopoietic Stem Cell Differentiation in Complex Therapy for Patients with COVID-19. *Stem Cell Rev. Rep.* **2021**, *17* (1), 285–290. https://doi.org/10.1007/s12015-020-10087-6.

(125) NIH. ClinicalTrials.gov https://clinicaltrials.gov/ (accessed Feb 24, 2021).

(126) Sahu, K. K.; Siddiqui, A. D.; Cerny, J. Mesenchymal Stem Cells in COVID-19: A Journey from Bench to Bedside. *Lab. Med.* **2021**, *52* (1), 24–35. https://doi.org/10.1093/labmed/lmaa049.

(127) Jeyaraman, M.; John, A.; Koshy, S.; Ranjan, R.; Anudeep, T. C.; Jain, R.; Swati, K.; Jha, N. K.; Sharma, A.; Kesari, K. K.; Prakash, A.; Nand, P.; Jha, S. K.; Reddy, P. H. Fostering Mesenchymal Stem Cell Therapy to Halt Cytokine Storm in COVID-19. *Biochim Biophys Acta - Mol Basis Dis.* **2021**, *1876* (2), 166014. https://doi.org/10.1016/j.bbadis.2020.166014.

(128) Chen, Y.; Chan, V. S. F.; Zheng, B.; Chan, K. Y. K.; Xu, X.; To, L. Y. F.; Huang, F. P.; Khoo, U. S.; Lin, C. L. S. A Novel Subset of Putative Stem/Progenitor CD34+ Oct-4 + Cells Is the Major Target for SARS Coronavirus in Human Lung. *J. Exp. Med.* **2007**, *204* (11), 2529–2536. https://doi.org/10.1084/jem.20070462.

(129) Hashemian, S. M. R.; Aliannejad, R.; Zarrabi, M.; Soleimani, M.; Vosough, M.; Hosseini, S. E.; Hossieni, H.; Keshel, S. H.; Naderpour, Z.; Hajizadeh-Saffar, E.; Shajareh, E.; Jamaati, H.; Soufi-Zomorrod, M.; Khavandgar, N.; Alemi, H.; Karimi, A.; Pak, N.; Rouzbahani, N. H.; Nouri, M.; Sorouri, M. *et al.* Mesenchymal Stem Cells Derived from Perinatal Tissues for Treatment of Critically Ill COVID-19-Induced ARDS Patients: A Case Series. *Stem Cell Res. Ther.* **2021**, *12* (1), 91. https://doi.org/10.1186/s13287-021-02165-4.

(130) Weiss, C.; Carriere, M.; Fusco, L.; Fusco, L.; Capua, I.; Regla-Nava, J. A.; Pasquali, M.; Pasquali, M.; Pasquali, M.; Scott, J. A.; Vitale, F.; Vitale, F.; Unal,

M. A.; Mattevi, C.; Bedognetti, D.; Merkoçi, A.; Merkoçi, A.; Tasciotti, E.; Tasciotti, E.; Yilmazer, A. *et al.* Toward Nanotechnology-Enabled Approaches against the COVID-19 Pandemic. *ACS Nano* **2020**, *14* (6), 6383–6406. https://doi.org/10.1021/acsnano.0c03697.

(131) Saburi, E.; Abazari, M. F.; Hassannia, H.; Mansour, R. N.; Eshaghi-Gorji, R.; Gheibi, M.; Rahmati, M.; Enderami, S. E. The Use of Mesenchymal Stem Cells in the Process of Treatment and Tissue Regeneration after Recovery in Patients with Covid-19. *Gene* **2021**, *777*, 145471. https://doi.org/10.1016/j.gene.2021.145471.

(132) Rauti, R.; Musto, M.; Bosi, S.; Prato, M.; Ballerini, L. Properties and Behavior of Carbon Nanomaterials When Interfacing Neuronal Cells: How Far Have We Come? *Carbon N. Y.* **2019**, *143*, 430–446. https://doi.org/10.1016/j.carbon.2018.11.026.

(133) Nguyen, T. P.; Nguyen, D. L. T.; Nguyen, V. H.; Le, T. H.; Vo, D. V. N.; Ly, Q. V.; Kim, S. Y.; Van Le, Q. Recent Progress in Carbon-Based Buffer Layers for Polymer Solar Cells. *Polymers*. **2019**, *11* (11), 1858. https://doi.org/10.3390/polym11111858.

(134) Gacem, M. A.; Gacem, H.; Ould-El-Hadj-Khelil, A. Nanocarbons: Antibacterial, Antifungal, and Antiviral Activity and the Underlying Mechanism. *Carbon Nanomater. Agri-Food Environ. Appl.* **2020**, 505–533. https://doi.org/10.1016/b978-0-12-819786-8.00022-0.

(135) Castro, E.; Garcia, A. H.; Zavala, G.; Echegoyen, L. Fullerenes in Biology and Medicine. *J. Mater. Chem. B*. **2017**, *5* (32), 6523–6535. https://doi.org/10.1039/c7tb00855d.

(136) Friedman, S. H.; DeCamp, D. L.; Kenyon, G. L.; Sijbesma, R. P.; Srdanov, G.; Wudl, F. Inhibition of the HIV-1 Protease by Fullerene Derivatives: Model Building Studies and Experimental Verification. *J. Am. Chem. Soc.* **1993**, *115* (15), 6506–6509. https://doi.org/10.1021/ja00068a005.

(137) Toniolo, C.; Bianco, A.; Maggini, M.; Scorrano, G.; Prato, M.; Marastoni, M.; Tomatis, R.; Spisani, S.; Palú, G.; Blair, E. D. A Bioactive Fullerene Peptide. *J. Med. Chem.* **1994**, *37* (26), 4558–4562. https://doi.org/10.1021/jm00052a015.

(138) Schuster, D. I.; Wilson, S. R.; Schinazi, R. F. Anti-Human Immunodeficiency Virus Activity and Cytotoxicity of Derivatized Buckminsterfullerenes. *Bioorganic Med. Chem. Lett.* **1996**, *6* (11), 1253–1256. https://doi.org/10.1016/0960-894X(96)00210-7.

(139) Nacsa, J.; Segesdi, J.; Gyuris, Á.; Braun, T.; Rausch, H.; Buvári-Barcza, Á.; Barcza, L.; Minarovits, J.; Molnár, J. Antiretroviral Effects of Nonderivatized C60 *in Vitro*. *Fuller. Sci. Technol.* **1997**, *5* (5), 969–976. https://doi.org/10.1080/15363839708013310.

(140) Bosi, S.; Da Ros, T.; Spalluto, G.; Balzarini, J.; Prato, M. Synthesis and Anti-HIV Properties of New Water-Soluble Bis- Functionalized[60]Fullerene Derivatives. *Bioorganic Med. Chem. Lett.* **2003**, *13* (24), 4437–4440. https://doi.org/10.1016/j.bmcl.2003.09.016.

(141) Marchesan, S.; Da Ros, T.; Spalluto, G.; Balzarini, J.; Prato, M. Anti-HIV Properties of Cationic Fullerene Derivatives. *Bioorganic Med. Chem. Lett.* **2005**, *15* (15), 3615–3618. https://doi.org/10.1016/j.bmcl.2005.05.069.

(142) Troshina, O. A.; Troshin, P. A.; Peregudov, A. S.; Kozlovskiy, V. I.; Balzarini, J.; Lyubovskaya, R. N. Chlorofullerene C60Cl6: A Precursor for Straightforward Preparation of Highly Water-Soluble Polycarboxylic Fullerene Derivatives Active against HIV. *Org. Biomol. Chem.* **2007**, *5* (17), 2783–2791.

https://doi.org/10.1039/b705331b.
(143) Kataoka, H.; Ohe, T.; Takahashi, K.; Nakamura, S.; Mashino, T. Novel Fullerene Derivatives as Dual Inhibitors of Hepatitis C Virus NS5B Polymerase and NS3/4A Protease. *Bioorganic Med. Chem. Lett.* **2016**, *26* (19), 4565–4567. https://doi.org/10.1016/j.bmcl.2016.08.086.
(144) Sijbesma, R.; Srdanov, G.; Wudl, F.; Castoro, J. A.; Wilkins, C.; Friedman, S. H.; DeCamp, D. L.; Kenyon, G. L. Synthesis of a Fullerene Derivative for the Inhibition of HIV Enzymes. *J. Am. Chem. Soc.* **1993**, *115* (15), 6510–6512. https://doi.org/10.1021/ja00068a006.
(145) Schinazi, R. F.; Sijbesma, R.; Srdanov, G.; Hill, C. L.; Wudl, F. Synthesis and Virucidal Activity of a Water-Soluble, Configurationally Stable, Derivatized C60 Fullerene. *Antimicrob. Agents Chemother*. **1993**, *37* (8), 1707–1710. https://doi.org/10.1128/AAC.37.8.1707.
(146) Barras, A.; Pagneux, Q.; Sane, F.; Wang, Q.; Boukherroub, R.; Hober, D.; Szunerits, S. High Efficiency of Functional Carbon Nanodots as Entry Inhibitors of Herpes Simplex Virus Type 1. *ACS Appl. Mater. Interfaces* **2016**, *8* (14), 9004–9013. https://doi.org/10.1021/acsami.6b01681.
(147) Huang, S.; Gu, J.; Ye, J.; Fang, B.; Wan, S.; Wang, C.; Ashraf, U.; Li, Q.; Wang, X.; Shao, L.; Song, Y.; Zheng, X.; Cao, F.; Cao, S. Benzoxazine Monomer Derived Carbon Dots as a Broad-Spectrum Agent to Block Viral Infectivity. *J. Colloid Interface Sci.* **2019**, *542*, 198–206. https://doi.org/10.1016/j.jcis.2019.02.010.
(148) Rathinam, N. K.; Saravanan, C.; Parimal, P.; Perumal, V.; Perumal, M. Molecular Interactions of Graphene with HIV-Vpr, Nef and Gag Proteins: A New Approach for Treating HIV Infections. *Korean J. Chem. Eng.* **2014**, *31* (5), 744–747. https://doi.org/10.1007/s11814-014-0049-8.
(149) Iannazzo, D.; Pistone, A.; Galvagno, S.; Ferro, S.; De Luca, L.; Monforte, A. M.; Da Ros, T.; Hadad, C.; Prato, M.; Pannecouque, C. Synthesis and Anti-HIV Activity of Carboxylated and Drug-Conjugated Multi-Walled Carbon Nanotubes. *Carbon N. Y.* **2015**, *82* (C), 548–561. https://doi.org/10.1016/j.carbon.2014.11.007.
(150) Liu, W.; Zhang, J.; Li, C.; Tang, L.; Zhang, Z.; Yang, M. A Novel Composite Film Derived from Cysteic Acid and PDDA-Functionalized Graphene: Enhanced Sensing Material for Electrochemical Determination of Metronidazole. *Talanta* **2013**, *104*, 204–211. https://doi.org/10.1016/j.talanta.2012.11.013.
(151) Li, D.; Müller, M. B.; Gilje, S.; Kaner, R. B.; Wallace, G. G. Processable Aqueous Dispersions of Graphene Nanosheets. *Nat. Nanotechnol.* **2008**, *3* (2), 101–105. https://doi.org/10.1038/nnano.2007.451.
(152) Chen, Y. N.; Hsueh, Y. H.; Hsieh, C. Te; Tzou, D. Y.; Chang, P. L. Antiviral Activity of Graphene–Silver Nanocomposites against Non-Enveloped and Enveloped Viruses. *Int. J. Environ. Res. Public Health* **2016**, *13* (4), 430. https://doi.org/10.3390/ijerph13040430.
(153) Du, T.; Lu, J.; Liu, L.; Dong, N.; Fang, L.; Xiao, S.; Han, H. Antiviral Activity of Graphene Oxide-Silver Nanocomposites by Preventing Viral Entry and Activation of the Antiviral Innate Immune Response. *ACS Appl. Bio Mater.* **2018**, *1* (5), 1286–1293. https://doi.org/10.1021/acsabm.8b00154.
(154) Iannazzo, D.; Pistone, A.; Salamò, M.; Galvagno, S.; Romeo, R.; Giofré, S. V.; Branca, C.; Visalli, G.; Di Pietro, A. Graphene Quantum Dots for Cancer Targeted Drug Delivery. *Int. J. Pharm.* **2017**, *518* (1–2), 185–192. https://doi.org/10.1016/j.ijpharm.2016.12.060.

(155) Iannazzo, D.; Pistone, A.; Ferro, S.; De Luca, L.; Monforte, A. M.; Romeo, R.; Buemi, M. R.; Pannecouque, C. Graphene Quantum Dots Based Systems As HIV Inhibitors. *Bioconjug. Chem.* **2018**, *29* (9), 3084–3093. https://doi.org/10.1021/acs.bioconjchem.8b00448.

(156) Donskyi, I. S.; Nie, C.; Ludwig, K.; Trimpert, J.; Ahmed, R.; Quaas, E.; Achazi, K.; Radnik, J.; Adeli, M.; Haag, R.; Osterrieder, K. Graphene Sheets with Defined Dual Functionalities for the Strong SARS-CoV-2 Interactions. *Small* **2021**, *17* (11), e2007091. https://doi.org/10.1002/smll.202007091.

(157) Ruiz-Hitzky, E.; Darder, M.; Wicklein, B.; Ruiz-Garcia, C.; Martín-Sampedro, R.; del Real, G.; Aranda, P. Nanotechnology Responses to COVID-19. *Adv. Healthc. Mater.* **2020**, *9* (19), e2000979. https://doi.org/10.1002/adhm.202000979.

(158) Lim, S. Y.; Shen, W.; Gao, Z. Carbon Quantum Dots and Their Applications. *Chem. Soc. Rev.* **2015**, *44* (1), 362–381. https://doi.org/10.1039/c4cs00269e.

(159) Lee, C. Porcine Epidemic Diarrhea Virus: An Emerging and Re-Emerging Epizootic Swine Virus. *Virol. J.* **2015**, *12*, 193. https://doi.org/10.1186/s12985-015-0421-2.

(160) Serrano-Aroca, Á.; Ruiz-Pividal, J. F.; Llorens-Gámez, M. Enhancement of Water Diffusion and Compression Performance of Crosslinked Alginate Films with a Minuscule Amount of Graphene Oxide. *Sci. Rep.* **2017**, *7* (1), 1–8. https://doi.org/10.1038/s41598-017-10260-x.

(161) Llorens-Gámez, M.; Serrano-Aroca, Á. Low-Cost Advanced Hydrogels of Calcium Alginate/Carbon Nanofibers with Enhanced Water Diffusion and Compression Properties. *Polymers (Basel).* **2018**, *10* (4), 405. https://doi.org/10.3390/polym10040405.

(162) Llorens-Gámez, M.; Salesa, B.; Serrano-Aroca, Á. Physical and Biological Properties of Alginate/Carbon Nanofibers Hydrogel Films. *Int. J. Biol. Macromol.* **2020**, *151*, 499–507. https://doi.org/10.1016/j.ijbiomac.2020.02.213.

(163) Serrano-Aroca, Á.; Iskandar, L.; Deb, S. Green Synthetic Routes to Alginate-Graphene Oxide Composite Hydrogels with Enhanced Physical Properties for Bioengineering Applications. *Eur. Polym. J.* **2018**, *103*, 198–206. https://doi.org/10.1016/j.eurpolymj.2018.04.015.

(164) Elias, L.; Taengua, R.; Frígols, B.; Serrano-Aroca, Á.; Salesa, B. Carbon Nanomaterials and LED Irradiation as Antibacterial Strategies against Gram-Positive Multidrug-Resistant Pathogens. *Int. J. Mol. Sci.* **2019**, *20* (14), 3603. https://doi.org/10.3390/ijms20143603.

(165) Song, Z.; Wang, X.; Zhu, G.; Nian, Q.; Zhou, H.; Yang, D.; Qin, C.; Tang, R. Virus Capture and Destruction by Label-Free Graphene Oxide for Detection and Disinfection Applications. *Small* **2015**, *11* (9–10), 1771–1776. https://doi.org/10.1002/smll.201401706.

(166) Sanmartín-Santos, I.; Gandía-Llop, S.; Salesa, B.; Martí, M.; Lillelund Aachmann, F.; Serrano-Aroca, Á. Enhancement of Antimicrobial Activity of Alginate Films with a Low Amount of Carbon Nanofibers (0.1% w/W). *Appl. Sci.* **2021**, *11* (5), 2311. https://doi.org/10.3390/app11052311.

(167) Baltimore, D. Expression of Animal Virus Genomes. *Bacteriol. Rev.* **1971**, *35* (3), 235–241. https://doi.org/10.1128/mmbr.35.3.235-241.1971.

(168) Sirotkin, A. K.; Zarubaev, V. V.; Poznyiakova, L. N.; Dumpis, M. A.; Muravieva, T. D.; Krisko, T. K.; Belousova, I. M.; Kiselev, O. I.; Piotrovsky, L. B. Pristine Fullerene C60: Different Water Soluble Forms - Different Mechanisms of Biological Action. In *Fullerenes Nanotubes and Carbon*

Nanostructures; Taylor & Francis Group, 2006; Vol. 14, pp 327–333. https://doi.org/10.1080/15363830600665375.

(169) Riley, P. R.; Narayan, R. J. Recent Advances in Carbon Nanomaterials for Biomedical Applications: A Review. *Curr. Opin. Biomed. Eng.* **2021**, *17*, 100262. https://doi.org/10.1016/j.cobme.2021.100262.

(170) Gupta, T. K.; Budarapu, P. R.; Chappidi, S. R.; Y.B., S. S.; Paggi, M.; Bordas, S. P. Advances in Carbon Based Nanomaterials for Bio-Medical Applications. *Curr. Med. Chem.* **2018**, *26* (38), 6851–6877. https://doi.org/10.2174/0929867326666181126113605.

(171) Maiti, D.; Tong, X.; Mou, X.; Yang, K. Carbon-Based Nanomaterials for Biomedical Applications: A Recent Study. *Front. Pharmacol.* **2019**, *9*, 1401. https://doi.org/10.3389/fphar.2018.01401.

(172) Bhattacharya, K.; Mukherjee, S. P.; Gallud, A.; Burkert, S. C.; Bistarelli, S.; Bellucci, S.; Bottini, M.; Star, A.; Fadeel, B. Biological Interactions of Carbon-Based Nanomaterials: From Coronation to Degradation. *Nanomedicine.* **2016**, *12* (2), 333–351. https://doi.org/10.1016/j.nano.2015.11.011.

(173) Moghimi, S. M.; Wibroe, P. P.; Wu, L.; Farhangrazi, Z. S. Insidious Pathogen-Mimicking Properties of Nanoparticles in Triggering the Lectin Pathway of the Complement System. *Eur. J. Nanomed.* **2015**, *7* (3), 263–268. https://doi.org/10.1515/ejnm-2015-0014.

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