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News and opinions

Can graphene take part in the fight against COVID-19?

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ABSTRACT

The pneumonia outbreak of coronavirus disease 2019 (COVID-19) represents a global issue. The bidimensional material graphene has captured much attention due to promising antimicrobial applications and has also demonstrated antiviral efficacy. In response to this global outbreak, we summarized the current state of knowledge of graphene and virus interaction as well as possible successful applications to fight COVID-19. Antibody-conjugated graphene sheets can rapidly detect targeted virus proteins and can be useful for large population screening, but also for the development of environmental sensors and filters, given the low cost of graphene materials. Functionalized graphene has demonstrated a good viral capture capacity that, combined with heat or light-mediated inactivation, could be used as a disinfectant. Graphene sensors arrays can be implemented on standard utility textiles and drug efficacy screening. Thanks to its high versatility, we foresee that graphene may have a leading role in the fight against COVID-19.

Introduction

The unprecedented pneumonia outbreak of coronavirus disease 2019 (COVID-19) is tearing global health systems apart. While government bodies are struggling in preventing further spread of COVID-19, researchers immediately started tests on vaccines and a clinical trial is currently underway with potential treatments for severe acute respiratory syndrome coronavirus – 2 (SARS-CoV-2) [1]. The coronavirus corona is formed by surface proteic projections on the viral lipid envelope enclosing single-stranded positive-sense RNA (see Fig. 1a) [2].

In the last decade, the two-dimensional material graphene has captured much attention due to its superb electronic properties and promising applications, including approaches to fight or detect drug-resistant bacterial infections. Pristine graphene is a single-atom-thick sheet of hexagonally arranged carbon atoms, graphene oxide (GO) is its oxidized counterpart. The reduced GO (rGO) is produced from GO after the removal of oxygen groups by reducing agents to obtain a material analog to pristine graphene. Being a single layer of carbon atoms, graphene has an exceptionally high surface to mass ratio. With such a remarkable specific surface area, it can achieve single-molecule detection. Even when a single biomolecule comes in contact with the graphene surface it can modulate its electrical properties, making this nanomaterial a perfect sensor [3]. These bidimensional materials also have a strong interaction with light: a single layer of graphene can absorb 2.3% of incident visible light [4]. This property is extremely important for heat generation and sterilization of materials. GO oxygen groups make its surface more hydrophilic compared to rGO and graphene. Further, surface oxygen provides reaction sites for adsorption or functionalization with proteins, enzymes, and nucleic acids. With chemically selective functionalization, made easy by the variety of groups on the surface, GO can specifically target analytes [3]. We can ask ourselves how graphene research can take part in the fight against SARS-CoV-2. Unique behaviors have been observed from pathogens that come in contact with bidimensional carbon flakes [5].

It has been demonstrated that bacteria touching graphene surface lose integrity [6,7], while effects on viruses have been less well characterized. Indeed most of virus-related graphene research has been focused on the development of sensors for disease diagnostics [8]. In this direction, antibody-conjugated GO sheets can rapidly detect targeted virus proteins and can be coupled to nanomaterial electronic properties for signal amplification [9,10]. This can be useful not only in point-of-care or large population screening, given the low cost of graphene materials, but also for the development of environmental sensors.

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Graphene materials interact with viruses

It has been demonstrated that graphene has good viral inhibition capacity. The first evidence of graphene antiviral effects was reported in 2012, when thin films of rGO – tungsten oxide were exploited for photoinactivation of bacteriophages under visible light irradiation [11]. Broad-spectrum antivirals like heparin or heparin-like drugs and sulfate-rich polymers mimic the cell surface sugars responsible for viral attachment, such as heparan sulfate (HS) [12,13]. The large surface of graphene provides the highest ligand contact area for the adsorption of negatively charged sulfates. These can interact with virions' positively charged residues and block microorganisms [14]. Ziem and colleagues synthesized thermal rGO sulfate derivatives and demonstrated their antiviral activity against African swine fever virus, orthopoxvirus and herpesvirus strains [14,15]. In particular, rGO has been functionalized with dendritic polyglycerol (dPG) which was then modified by sulfation. dPGS binds to the heparin-binding domain of surface protein A27 on different orthopoxvirus strains [15].

The interactions between these rGO sulfated derivatives and viruses were sulfation degree and polymer density-dependent [14]. The higher the degree of sulfation and the smaller the size, the more potent was the effect on herpesvirus. This was explained as a combined result of the easiest bending and cooperative encapsulation by two or more small GO sheets [14]. Indeed GO flakes can wrap and confine microorganisms by enclosing them in an insulating carbon blanket [16].

Interestingly, it has been recently demonstrated how the SARS-CoV-2 Spike S1 protein receptor-binding domain can interact with heparin and change conformation. This has implications for the development of a first-line therapeutic by repurposing heparin and glycosamminoglycans-based antivirals [17], including sulfated derivatives of GO. After capturing, the light absorbance of graphene can be used to destroy viral particles. Sulfonated magnetic nanoparticles functionalized with rGO have been successfully used to capture and photothermally destroy herpes simplex virus type 1 (HSV-1) using near-infrared (NIR) light [18]. These data point out how GO capture could be coupled with NIR treatments of lungs. Indeed absorbers like graphene and GO are in NIR tissue transparency window and allow deep penetration in the body of incident light, as demonstrated for lung metastases treatment.

Carbon dots combined with the natural antimicrobial polyphenol curcumin have been demonstrated to be effective against coronavirus models [19]. β-cyclodextrins-functionalized sulfonated graphene has been combined with curcumin (curcumin-loaded β-cyclodextrins functionalized sulfonated graphene, GSCC) to treat the respiratory syncytial virus (RSV) [20]. The sulfonate groups on the GSCC can mimic the cell surface and inhibit RSV infection through a competitive inhibition mechanism, simulating cell receptors used for virus attachment. GSCC nanoparticle effects are due to a double mechanism, by curcumin-mediated viral inactivation and also by inhibition of the virus attachment tothe host cell membranes [20]. The above examples highlight how sulfonate residues interact electrostatically with proteins of virus cellular recognition. However, the disruption of the coronavirus lipid bilayer is also of fundamental importance. Polyglycerol sulfated graphene functionalized with fatty amine chains can wrap and inactivate HSV-1 (see Fig. 1b). A strong correlation between the toxicity of graphene composites and the length of fatty amines (from 3 to 18 carbon atoms) reveals the crucial role of the hydrophobic interactions with these chain and virus lipid bilayer [21]. So what happens when a graphene surface comes in contact with viruses? GO interacts with viruses mainly by hydrogen bonding, electrostatic interactions and redox reaction [22]. An early study of 2014 highlighted how differences between GO and its sulfated rGO derivative were not visible in the antiviral activity against HSV-1. These materials had a similar negative charge density, which was probably the principal factor affecting virus inhibition [23,24]. It should be noted that GO and rGO have an intrinsic ability to adsorb charged lipids and destroy membranes after association with their aromatic plane [25,26]. This property has been used to explain GO activity against feline coronavirus compared to its inefficacy against non-enveloped infectious bursal disease virus [27]. Graphene derivatives have been exploited also as drug delivery systems for antiviral compounds like reverse transcriptase inhibitors conjugated with graphene quantum dots to treat HIV [28], and hypericin-GO against reovirus [29]. Remarkably, hypericin is on the list of computationally identified possible treatments against COVID-19 [30]. Furthermore, graphene is a potent immunomodulator and GO-silver nanocomposites enhance the production of natural antiviral defenses (interferon-α and IFN-stimulating genes) [31]. Finally, one of the early studies on GO demonstrated how this nanomaterial could be useful in testing coronavirus helicase inhibitors as drug candidates for antivirus therapy [32]. Helicases are enzymes that unwind double-stranded nucleic acids into single-stranded during virus replication and proliferation. For this reason, helicases have been targeted for antiviral therapy. However, the conventional assay for helicase activity is time-consuming and inefficient because of the lengthy preparation time and methodological pitfalls. GO has a preferential binding of single stranded DNA on its surface. This has been used to obtain a cost-effective monitoring of helicase activity. More importantly, GO testing is high throughput and allows parallel assays for screening of helicase inhibition by drugs [32].

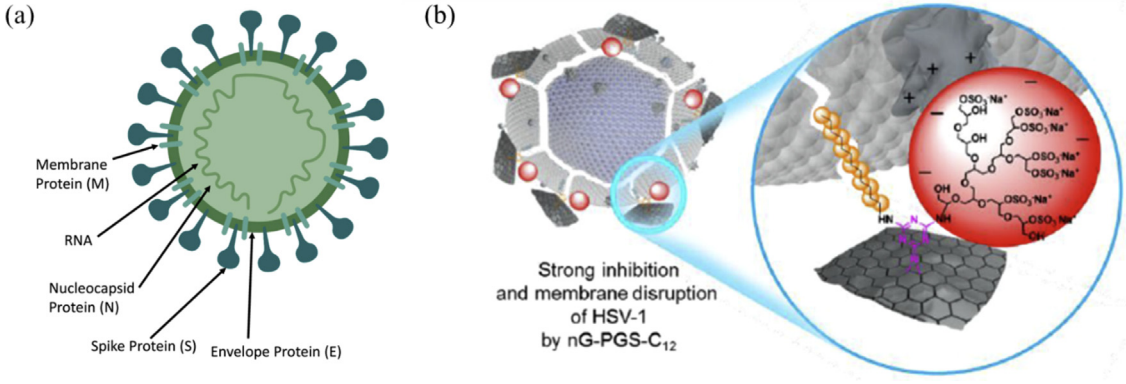

Fig. 1. (a) Main structure of coronavirus, reproduced with permission from [2], Creative Commons Printed with permission from Springer Nature. (b) Representation of HSV wrapping by sulfated graphene derivatives and illustration of long alkyl chain disrupting virus envelope. Reproduced with permission from [21]. Copyright 2019 Royal Society of Chemistry. Printed with permission from the Royal Society of Chemistry.

Fig. 2. Schematic of envisioned application space for the GO screen printed flexible impedance biosensors. Sensors arrays implemented on textiles and interfaced with flexible electronics can report the location of positives and identify sources of the outbreak. Reproduced with permission from [35], Creative Commons. Printed with permission from IOP Publishing.

Graphene textiles for pandemic spread control

Besides the treatment approaches based on graphene capturing, this nanomaterial has also applicability in the control of the epidemiological spreading of the disease. Graphene filters have been produced for capturing particulates and bacteria to decrease the transmission of nosocomial infections [33]. The bacteria blocked on the filter are not able to proliferate and by increasing filter temperature over 300 °C microorganisms along with molecules that can cause diseases are destroyed. Filters can be used also for sampling bioaerosols for characterization of transmission in hospital settings and exposure risk of workers in general [34]. Graphene has relevant applications in electronic, optical, and thermoelectric devices including transparent and flexible conductors, electrical and optical sensors. In 2018, Kinnamon and colleagues developed a textile screen-printed biosensor based on a GO transduction film for the detection of environmental exposure to influenza A virus H1N1 [35].

Nucleoprotein specific Influenza A antibody was conjugated on a GO surface to create a sensor integrated with textile. This had a detection limit of 10 ng/mL and is capable of uncovering exposure to the virus before symptoms manifest (see Fig. 2 from [35]). Similarly, future research could envisage the development of SARS-CoV-2 sensors for epidemiological control of virus spreading through protective clothing.

GO films are also useful for creating breathable barrier layers in fabrics. The water permeation rate of graphene is superior to many other technologies and this nanomaterial also can exclude bacteria, viruses and other molecular agents sterically [36]. The protective effect of facemasks is maintained when the surface layer is hydrophobic and dry, otherwise microorganisms may be able to penetrate the protective layers. Coatings of graphene could be exploited as previously described with nanoparticles of silver nitrate and titanium dioxide [37], to trap pathogens. Protective graphene facemasks could be recycled by photocatalysis [11] or heat [22]. Indeed graphene heat and electronic transduction can than be used to sterilize textile and filters that came in contact with viruses. Indeed, it has been demonstrated how viruses can be denatured after adsorption on graphene and mild heat treatment at 56 °C for 30 minutes [22,38].

Limitations and Challenges

Even if the antiviral efficacy of graphene has been demonstrated, the immediate use of graphene for treatment of COVID-19 is unrealistic. The only available in vivo test for virus treatment demonstrated the efficacy of GO-hypericin in ducklings infected with the Novel duck reovirus [29]. We should however point out that in vivo toxicity of graphene is still a matter of debate. Graphene materials can cross biological barriers but are less cytotoxic toward macrophages, important cells for immunological response, compared with other carbon nanomaterials. Conclusions on graphene toxicity are however made difficult due to the infinite combinations of dose, surface chemistry, exposure route used for the evaluation [39]. Its instability and aggregation in solution arefurther problems, given the mandatory stability required for drugs and vaccine storage prior use [40].

Summary and Outlook

Graphene is a nanomaterial with a thousand advantages. Although the urgency of treatment and vaccines necessary for SARS-CoV-2 combined with the lack of a complete toxicity characterization make an in vivo use farther off than we expected, there are other chances to exploit graphene and its composites against COVID-19. World Health Organization continues to highlight the need for the prioritization of personal protective equipment supplies for frontline healthcare workers and graphene could be used as coatings of facemasks to minimize the risk of transmission. Graphene textile applications for epidemiological exposure detection and for filtering are possible allies of health systems against pandemic spreading. Furthermore, high-throughput diagnostics and drug screening based on graphene sensors have been successfully demonstrated and some of these sensors have begun to make their way to the market place [3]. In response to this global outbreak, we have summarized the current state of knowledge of graphene-virus interaction as well as possible graphene uses transferrable to COVID-19 diagnosis and treatment. Further research is undoubtedly required but we foresee that graphene may have a leading role in the world fight against COVID-19.

Author Statement

Massimiliano Papi and Valentina Palmieri conceptualized and wrote the manuscript.

Declaration of Competing Interest

The authors declare that there are no conflicts of interest.

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