applied sciences

MDPI

Review

Graphene-Based Functional Hybrid Membranes for Antimicrobial Applications: A Review

Xiaoting Zhang, Hao Kong *, Guozheng Yang, Danzhu Zhu, Xin Luan, Peng He and Gang Wei *

College of Chemistry and Chemical Engineering, Qingdao University, Qingdao 266071, China; zhangxiaotting@163.com (X.Z.); yangguozheng123@outlook.com (G.Y.); zhudanzhu11@outlook.com (D.Z.); luanxin0924@outlook.com (X.L.); hepeng_qdu@outlook.com (P.H.)

* Correspondence: konghao5@outlook.com (H.K.); weigroup@qdu.edu.cn (G.W.); Tel.: +86-15066242101 (G.W.)

Abstract: Graphene-based nanomaterials have shown wide applications in antimicrobial fields due to their accelerated rate of pathogen resistance and good antimicrobial properties. To apply graphene materials in the antimicrobial test, the graphene materials are usually fabricated as two-dimensional (2D) membranes. In addition, to improve the antimicrobial efficiency, graphene membranes are modified with various functional nanomaterials, such as nanoparticles, biomolecules, polymers, etc. In this review, we present recent advances in the fabrication, functional tailoring, and antimicrobial applications of graphene-based membranes. To implement this goal, we first introduce the synthesis of graphene materials and then the fabrication of 2D graphene-based membranes with potential techniques such as chemical vapor deposition, vacuum filtration, spin-coating, casting, and layer-by-layer self-assembly. Then, we present the functional tailoring of graphene membranes by adding metal and metal oxide nanoparticles, polymers, biopolymers, metal–organic frameworks, etc., with graphene. Finally, we focus on the antimicrobial mechanisms of graphene membranes, and demonstrate typical studies on the use of graphene membranes for antibacterial, antiviral, and antifungal applications. It is expected that this work will help readers to understand the antimicrobial mechanism of various graphene-based membranes and, further, to inspire the design and fabrication of functional graphene membranes/films for biomedical applications.

Keywords: graphene; hybrid materials; membranes; fabrication techniques; antibacterial; antiviral; antifungal

Citation: Zhang, X.; Kong, H.; Yang, G.; Zhu, D.; Luan, X.; He, P.; Wei, G. Graphene-Based Functional Hybrid Membranes for Antimicrobial Applications: A Review. *Appl. Sci.* **2022**, *12*, 4834. https://doi.org/10.3390/app12104834

Academic Editor: Antonio Di Bartolomeo

Received: 23 April 2022
Accepted: 9 May 2022
Published: 10 May 2022

1. Introduction

The rapid increase in population has accelerated water and air pollution, leading to the rapid development of infectious diseases and pathogens worldwide [1,2]. Due to the abuse of antibiotics and the rapid transfer of drug-resistance genes, pathogens are becoming resistant to traditional antibiotics at an alarming rate, which makes it extremely difficult to treat infections [3,4]. As microbial resistance to antibiotics kills millions of people worldwide [5], it is urgent to develop new types of antimicrobial materials with both extensive antimicrobial activity and suitable biocompatibility.

Graphene is a two-dimensional (2D), carbon-based, ultra-thin, biocompatible nanomaterial with excellent mechanical, thermal, and electrical properties [6,7]. Its derivatives graphene oxide (GO) and reduced graphene oxide (rGO) are very suitable for limiting microbial infection [8,9]. GO is the oxidation form of graphene, which is rich in oxygen-containing groups at its edges and defects, such as carboxyl (COOH), carbonyl (C=O) and hydroxyl (OH) [10,11]. These abundant groups promote interactions with biomolecules and induce bacterial death in the absence of intracellular processes [12,13]. The antibacterial effect of graphene generally involves physical and chemical effects [14]. Graphene-based materials can directly contact the cell membrane or wrap bacterial cells, thus causing physical damage to bacteria [15,16]. The chemical effect is mainly caused by oxidative stress

caused by reactive oxygen species (ROS) or charge transfer [17]. The functionalization of GO surface creates the possibility of obtaining highly effective antimicrobial agents [18]. The surface oxygen function of GO ensures that they are amphipathic, so the GO nanosheets can easily dissolve or disperse in water or other polar solvents [19]. Therefore, GO can successfully be used for further functionalization to form nanocomposites, or can uniformly deposited on different substrates as thin films from aqueous solutions. In addition, GO has been used as a carrier to disperse and stabilize a variety of nanomaterials, such as metals, metal oxides, polymers, carbon nanotube, biomaterials, and so on [20,21]. Due to their synergistic effect, graphene nanohybrids have high antimicrobial efficiency [22]. Due to its excellent antimicrobial properties and good biocompatibility, graphene nanocomposites have a wide range of potential applications in antimicrobial packaging, wound dressing and water disinfection [23]. In addition, it is an excellent drug-delivery vehicle to deliver various antibiotics and antibacterial enzymes [24,25].

The antimicrobial effect of graphene-based nanomaterials is of great significance for effective antimicrobial therapy, because its mechanism is independent of antibiotics [26]. To maintain the bulk properties of graphene materials and reduce the cost, graphene membranes are used, which have high efficiency, little or no side effects and a long service life [27]. Graphene-based membranes prepared from graphene nanomaterials can even be directly applied to antimicrobials [28,29].

Many reviews have been released of graphene-based materials' potential for antimicrobial applications [18,23,30]. However, to date, there is no comprehensive review on the antimicrobial application of graphene-based membrane. In this review, we summarize the research status of the antibacterial interaction of graphene-based membrane materials (Figure 1). Then, the preparation methods of graphene membranes are summarized, including the casting method, coating method, layer-by-layer (LBL) self-assembly method and vacuum/pressure-assisted method. A variety of graphene-based composites developed for antimicrobial applications are summarized, with an emphasis on the mechanism and application of graphene-based composites in antimicrobial applications. We hope that this review will provide valuable insights, stimulate wider attention, and promote the further development of this promising field.

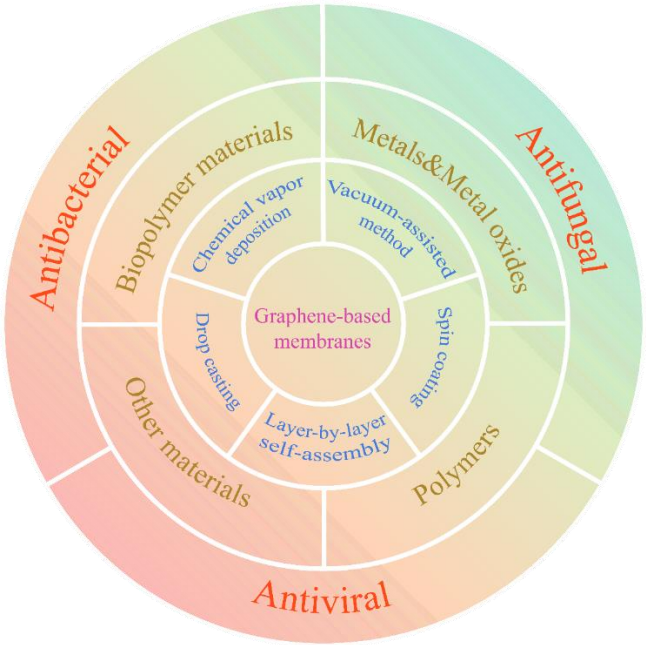

Figure 1. Schematic presentation of synthesis, functional tailoring, and antimicrobial applications of graphene-based membranes.

2. Properties and Synthesis of Graphene

2.1. Physical and Chemical Properties

Graphene is the simplest form of carbon and the thinnest material ever produced. Graphene materials have high electrical and thermal conductivity, as well as excellent mechanical properties [31]. It is well known that the electrical and optical properties of graphene thin films are strongly affected by graphene grain size and boundary [32].

Graphene is a material with high mechanical strength. Membrane-formed graphene can provide stronger support and adjustable spacing. Macroscopic membrane formation can increase the mechanical strength and controllability of graphene materials [33]. Graphene has a very high specific surface area (about 2675 m^2/g), and the inherent capacitance of the original graphene is 0.21 F/m^2, which exceeds that of all carbon-based materials [34]. Li et al. showed through experiments that graphene has a very high specific surface area and is a highly efficient additive, promoting the heterogeneous nucleation of water [35]. The addition of a surfactant can improve the hydrophilicity and stability of nano-materials. Leenaerts et al. pointed out that graphene is hydrophobic using density functional theory [18]. The binding energy between water droplets and graphene is weaker than that between water molecules, so water droplets tend to absorb more water molecules than graphene. This phenomenon shows that graphene is hydrophobic. The existence of a micro-nanostructure on the surface of graphene enhances its hydrophobicity. Water molecules can easily pass between graphene sheets without attaching, and the water flux increases. In addition, due to the existence of hydrophobic properties, particles are not easily attached to the membrane surface, which can reduce the membrane fouling rate [33]. In addition to the above properties, graphene has many other properties, such as a high surface area (2630 $m^2\ g^{-1}$), high Young's modulus (~1100 GPa), carrier mobility at room temperature (~15,000 $cm^2\ V^{-1}\ s^{-1}$), good optical transparency (~97.7%), excellent electrical conductivity (3000–5000 $W\ m^{-1}\ K^{-1}$) and the quantum Hall effect [33]. GO is not as hydrophobic as graphene due to the abundance of oxygen-containing functional groups in the planes and edges. Wang et al. prepared graphene oxide by oxidizing natural graphite and then reducing graphene by hydrazine hydrate [36]. The contact angle of water droplets on the surface of graphene oxide is 67.4°, which indicates that GO is hydrophilic. This is due to the existence of oxygen-containing functional groups on GO, which can form hydrogen bonds with water molecules, and the existence of oxidized functional groups increases the surface hydrophilicity [37]. Graphene-based nanomaterials are different in size, surface area, roughness, hydrophilicity, dispersion and functionalization, all of which can affect the interaction between graphene-based nanomaterials and bacterial cells, either alone or mutually.

2.2. Synthetic Methods

The development of high-yield and cheap preparation technology are the basis of the wide application of graphene. Many methods, such as mechanical stripping, epitaxial growth, redox, etc., have been proved to be effective methods of graphene synthesis [33].

Mechanical stripping is the simplest and most primitive preparation method, which makes independent graphene a reality. In this technique, a piece of graphite is stripped off several times and then transferred to the substrate. The number of layers in graphene can then be assessed using characterization methods such as optical microscope, Raman spectroscopy, atomic force microscope and scanning tunneling microscope. This method can still produce the highest quality crystals, but is limited by the inability to scale-up the process and is, therefore, only suitable for laboratory-scale experiments and prototyping [38]. Sinclair et al. studied the preparation of graphene by tape exfoliation from the perspective of molecular dynamics [39]. This was achieved by using molecular dynamics force field GraFF to represent the dispersion interaction of graphene. For nanoscale graphene sheets, two different exfoliation mechanisms (mixed-shear and positive-mode exfoliation) were observed, depending on the polymer binder used. The exfoliation method under these two mechanisms facilitates the synthesis of graphene rather than multilayer graphite.

Epitaxial growth is also a common method for the preparation of graphene. Xu et al. demonstrated the growth of graphene thin films with (5 × 50) cm^2 size in 20 min, and the ultra-high grain orientation of the graphene films is more than 99% [40]. The growth is the epitaxial growth in graphene on a meter-scale single crystal copper (111) surface as a template, and the seamless fusion of graphene facilitates the ultrafast growth of graphene films with high single crystallinity. This was achieved through the preparation of single crystal Cu (111) from industrial polycrystalline copper foils by temperature-gradient driven annealing and the wonderful effect of a continuous oxygen supply to adjacent oxides.

Graphite is chemically oxidized to GO and then reduced, which is a common method for the large-scale production of graphene or rGO. Among the high number of chemical-reducing agents used to prepare rGO, hydrazine ($N_2H_4 \cdot H_2O$) is the most famous, which can provide rGO with better electrical and structural properties [41]. DeSilva et al. reviewed the chemical reduction in GO by ascorbic acid, which is a non-toxic and cheap organic acid [42]. As hthe obtained rGO has high quality, high productivity, and can produce stable dispersions that are necessary for many applications, chemical reduction has a higher priority than non-chemical reduction. Guex et al. investigated the electrical conductivity of rGO preparation by the reduction in GO using sodium borohydride ($NaBH_4$) as function of time (2 min to 24 h) and temperature (20 °C to 80 °C) [43]. They used an inexpensive water reduction route that relies on sodium borohydride ($NaBH_4$), resulting in the highest conductivity of reduced graphene oxide to date.

3. Fabrication Techniques of Graphene-Based Membranes

2D graphene and its derivatives are the cornerstone of advanced thin membranes [44]. The molecular and ionic separation mechanism of graphene oxide films is basically selective transport through spacer channels between graphene oxide flakes, as well as defects and wrinkles in the graphene oxide flakes (Figure 2a) [45]. Compared with inorganic membranes or polymer membranes, graphene membranes have a better separation performance and application potential. However, the predominant graphene-based membranes show high permeation fluxes, which are fabricated by reducing their thickness to less than 50 nm. Graphene frameworks with polar functional groups are considered the key properties for obtaining ideal membranes in nanotechnology [44]. The main methods used to prepare graphene membranes include chemical vapor deposition (CVD), the vacuum-assisted method, spin-coating, dropping, and LBL self-assembly. Of the above methods, vacuum filtration and spin-coating are considered the most efficient for the preparation defect-free GO membranes [46].

3.1. Chemical Vapor Deposition

CVD is a chemical technology, which mainly uses one or more gas-phase compounds or elemental substances containing thin-film elements to perform chemical reactions on the surface of the substrate, to form thin films. CVD is an efficient method for the mass production of high-quality, graphene-based membranes. Different types of CVD methods are available, such as plasma-enhanced CVD, thermal CVD, thermal/cold wall CVD and many others. CVD is an advantageous method that enables the continuous preparation of graphene films by growing graphene on various substrates (nickel, copper, platinum, ruthenium or iridium) [32]. Due to the chemical inertness of graphene, the transfer of graphene from the growth substrate to other substrate can be difficult, and may cause defects and wrinkles in the graphene membrane during the transfer process. Thermal fluctuations can also affect the stability of the grown material [38]. Using this method, the complex transfer process of graphene growth on Cu or Ni substrates is unavoidable. On such substrates, high synthesis temperatures (>1200 °C) or high costs are required and often degrade the electrical and optical properties of graphene. Liu's group reported a new method for the direct synthesis of graphene on various insulating substrates (SiO_2, Al_2O_3, etc.) by the remote catalysis of copper nanoparticles by atmospheric pressure CVD, which

provides a low-defect preparation method. This is an efficient method for the production of large-area graphene membranes [47].

3.2. Vacuum Filtration-Assisted Method

Vacuum-assisted filtration methods are the most common and straightforward methods for the large-scale fabrication of free-standing, GO-based nanofiltration (NF), microfiltration, and ultrafiltration membranes [48]. Generally, GO membranes are prepared by filtering GO suspensions in water through porous supports, and the selection of porous supports can optimize the roughness and wettability of the membrane surface to obtain uniform GO membranes [46]. Baskoro et al. studied the interaction between GO laminates and salt solutions. The neatly arranged graphene oxide laminates are distributed on the top surface of the polyvinylidene fluoride-polyacrylic acid (PVDF-PAA) microporous layer by vacuum filtration. The PVDF-PAA layer was cast on the non-woven scaffold and formed by immersion in a water coagulation bath. The interaction of graphene oxide with salts plays an important role in tuning the properties of graphene oxide films (Figure 2b) [49]. Zhang et al. used the layer filtration construction technique to prepare GO-based membranes with improved separation efficiency and permeability [50]. The ultrathin GO framework layer was successfully deposited on the modified Torlon hollow fiber scaffold using the construction method, resulting in a composite membrane with excellent NF performance. Alayande et al. reported improved the antibacterial properties of rGO by preparing rGO-CuO nanocomposite films [51]. The rGO-CuO nanocomposite was synthesized using a simple hydrothermal method, and the nanocomposite membrane was prepared by filtration through a polytetrafluoroethylene (PTFE) filter, with the aid of a vacuum filtration device. The nanocomposite membrane exhibited good bacterial inactivation effect on *Pseudomonas aeruginosa* (*PAO1*). The antibacterial properties are due to the nanocomposite membrane's ability to transfer electrons from bacterial cells, resulting in bacterial cell inactivation. During the preparation of rGO-CuO nanocomposites, the hydrothermal treatment reduces graphene oxide and generates CuO on the reduced graphene oxide. Therefore, rGO-CuO aggregates to form a rod-like/sphere-like structure. Furthermore, the formed nanorods were able to penetrate bacterial cells, thereby enhancing the antibacterial properties of the nanocomposites.

3.3. Spin Coating

The spin-coating method has the advantages of a controllable thickness, high uniformity, short processing time and low cost. The size and shape of the substrate is a major limitation to implementing this technology on an industrial scale. Due to the unique two-dimensional structure of graphene oxide, the nanosheets may align during the coating process (Figure 2c) [52]. In this process flow, GO suspensions are first deposited on different substrates (silica, silicon nitride, glass, polymers, copper foils, etc.) to prepare thin and uniform graphene films. Then, the substrate is rotated at different speeds as needed, and the procedure is repeated until a uniform film is obtained. The addition of etchants corresponding to different substrates allows for the substrates to be removed, and independent GO films can be obtained after washing and drying. Since the face-to-face attractive capillary force generated by spin coating can overcome the repulsive force between the edges of GO nanosheets, thin films with dense GO layers can be obtained by this method. Kim et al. reported GO films with multilayered nanosheets, whose permeability was affected by the spin-coating operation, indicating that the microstructure of GO laminates relies on the precise control of the fabrication process [53]. Chi et al. also used spin coating, but they performed it at a higher temperature to accelerate the evaporation of the solvent [54]. A faster melting rate (in a boiling water bath) was used to generate an irreversible change between the shrinkage and expansion of graphene oxide for better exfoliation. The prepared GO membranes are ultra-thin and ordered, and have good separation application prospects.

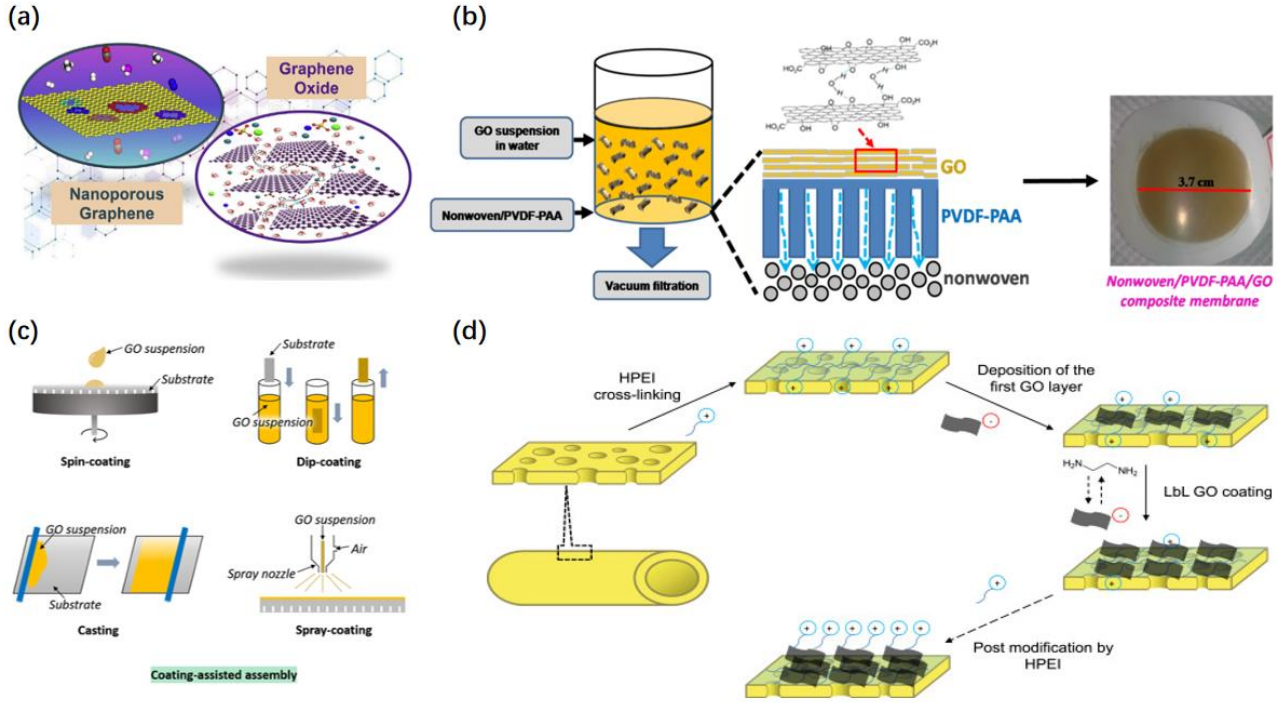

Figure 2. (**a**) Schematic diagram of two graphene-based films, including NPG film (left) and GO film (right). (Printed image with permission from Ref. [45], copyright 2021, American Chemistry Society). (**b**) Vacuum filtration method. (Printed image with permission from Ref. [49], copyright 2018, Elsevier Ltd.). (**c**) Coating-assisted assembly. (Printed image with permission from Ref. [52], copyright 2016, Nature Publication). (**d**) Schematic diagram of the procedure for constructing LbL GO framework films. (Printed image with permission from Ref. [50], copyright 2016, Elsevier Ltd.).

3.4. Drop Casting

Drop casting is also a simple fabrication process for GO-based membranes. In this method, GO suspensions are drop-cast on smooth-surfaced substrates (such as silica or paper), and then dried at room temperature. A freestanding and uniform GO film can be separated from the substrate by direct lift-off. The drop casting technique has received extensive attention due to its simplicity, ease of operation, rapidity, and small substrate size. However, even under near-ideal conditions, differences in the evaporation rate or concentration gradient of the liquid phase across the substrate can lead to changes in the thickness or internal structure of the entire film [46]. Cobos et al. prepared polyvinyl alcohol (PVA) composite films filled with GO-Ag NPs with antibacterial activity using the solution casting method [55]. Characterization of its microstructural and morphological indicated that the fillers had good dispersion in the polymer matrix. In addition, the exfoliated structure of the nanocomposite results in enhancements to the thermal stability, mechanical properties, and water resistance of the film. The PVA membranes filled with 0.5–5 wt% GO-Ag NPs showed antibacterial effects against *Escherichia coli* and *Staphylococcus aureus*, and the antibacterial effects were stronger for composites with a higher GO-Ag NPs content. Compared with *E. coli*, the PVA/GO-Ag NPs membrane showed a stronger inactivation of *Staphylococcus aureus*. The direct contact between bacteria and composite membranes offered a higher bactericidal effect than contact with the leachate of these membranes. Akbari et al. reported that high-concentration graphene oxide suspensions containing discotic nematic phases can be shear-aligned by blade casting to form ordered graphene oxide laminates [56].

3.5. Layer-by-Layer Self-Assembly

LBL allows for more precise control of the concentration, the geometry of the film to be deposited, the elasticity of the monolayer, etc., and the surface state. The LBL self-assembly

technique, which can construct multilayer films at the nanoscale, has received increasing attention in the fields of biomimicry, clean energy, and microelectronics [57]. Jia et al. used the LBL self-assembly technique to construct ordered dispersed GO and polymer multilayers, demonstrating an efficient and facile method to improve the quality of proton-exchange membranes. $(PU/GO/PDDA/GO)_{200}$ films were prepared by the alternate deposition of polyurethane (PU), positively charged poly(diallyldimethylammonium chloride) PDDA, and negatively charged GO. In addition to electrostatic attraction, intermolecular hydrogen bonding can also drive the completion of the LBL self-assembly process. Furthermore, due to the presence of intermolecular hydrogen bonds, phosphoric acid (PA) molecules are trapped and form PA-doped films. As expected, the $(PU/GO/PDDA/GO)_{200}/PA$ membrane showed high and stable proton conductivity, benefiting from the formation of free PA molecular chains and the reduced resistance to proton conduction. The excellent performance of LBL self-assembly technology in terms of proton conductivity, assembly, and mechanical stability provides a new strategy for the application of GO in the field of anhydrous proton-exchange membranes.

Shi et al. [58] prepared porous carbon nitride nanosheets (MCU-C_3N_4) with excellent photocatalytic properties using the supramolecular polymerization of melamine (C), melamine (M), and urea (U) in dimethyl sulfoxide (DMSO). They used the LBL assembly method for the first time to intercalate MCU-C_3N_4 and carbon nanotubes (CNTs) into GO on PVDF films, and the CNT/MCU-C_3N_4/GO materials were immobilized on the polymer. Three-dimensional heterostructured photocatalytic membranes (PMs) were fabricated on polyelectrolyte (PE)-modified PVDF [58]. Compared with pristine GO film and an MCU-C_3N_4/GO film prepared using the same method, the PMs with abundant nanochannels have excellent mechanical strength, good water permeability (14.35 L m^{-2} h^{-1} bar^{-1}) and synergetic removal efficiency of rhodamine B (RhB, 98.31%). In long-term operations, Rhodamine B is extremely harmful to the human body. It can cause acute and chronic poisoning injuries when it is ingested and when it comes into contact with the skin. It can be used as a model pollutant. Wang et al. assembled GO nanosheets with polycations on a polyacrylonitrile substrate using a LBL self-assembly strategy to form a multilayer film [59]. The migration and rearrangement of PE chains are restricted due to the confinement effect of adjacent GO nanosheets. As a result, the anti-swelling properties of the polycation/GO multilayer films are enhanced, making their properties more stable. Due to the anti-fouling ability of GO nanosheets, the fouling resistance of the multilayer films was also improved. Compared with pure PE multilayers, the separation performance, stability, and fouling resistance of polycation/GO membranes were improved. Zhang et al. successfully deposited ultrathin GO framework layers on a modified Torlon® hollow fiber scaffolds using the LBL approach, enabling composite membranes with excellent nanofiltration performance (Figure 2d) [50].

4. Functional Tailoring of Graphene-Based Membranes

Graphene has a stable structure and is not easy to react with other substances. Oxygen-containing functional groups make graphene oxide more functional and endow many excellent properties. GO can be very stably dispersed in water without dispersants and stabilizers, which is beneficial to its subsequent functional regulation. Since oxygen-containing groups are prone to chemical reactions, the surface of GO can undergo various chemical modifications to form graphene nanocomposites [33]. Graphene membranes have a nanoporous structure and functional feasibility, which make them ideal for achieving a higher permeation flux, higher selectivity, and better stability through control of the pore size and shape of membranes [60].

Graphene nanocomposites are promising antibacterial materials. Polymer matrix composites can give full play to the advantages of graphene and polymers on the basis of controllable graphene content [61]. Nanocomposites can overcome the limitations of individual components, and their structures have good properties, such as excellent electrical conductivity, easy modification, good dispersion and thermal stability, high

specific surface area, high mechanical strength, and good flexibility [62]. The following sections describe graphene-based bicomponent and multicomponent nanocomposites for antibacterial activity. These nanocomposites include metals, metal oxides, polymers, biopolymers, and other composite materials.

4.1. Metals and Metal Oxides

The doping of graphene films with alkali, transition or noble metals results in increased binding energy [60]. Compared with other nanoparticles, Ag NPs have shown good performance in water purification because they limit the growth of microorganisms such as algae, bacteria and fungi [44]. Silver ions can penetrate cells, destroy cell membranes and inactivate bacteria. In order to improve the anti-biofouling performance of polyvinylidene fluoride (PVDF) membranes, Li et al. synthesized GO-Ag composites as membrane antibacterial agents using a simple and environmentally friendly method [63]. GO-Ag composite films were prepared by phase inversion method using GO-silver composites with different concentrations (0.00–0.15%) as raw materials. Compared with the unmodified PVDF membrane, the hydrophilicity, mechanical properties and permeability of the GO-Ag composite modified membrane were improved. Silver phosphate (AP), as a silver ion (Ag^+) source, has a higher bactericidal effect than silver nanoparticles in membrane applications. Li et al. synthesized AP-supported GO quantum dots (GOQDs) multifunctional nanocomposites via a facile electrostatic drive method. Subsequently, GOQDs/APs were intercalated into dense polyamide (PA) layers via interfacial polymerization [64]. The results show that the thin film nanocomposite (TFN)-GOQD/AP membrane has a strong bactericidal performance against *Escherichia coli*, with a sterilization rate of 99.9%, good stability and excellent antifouling performance during reverse osmosis. Zhu et al. synthesized rGO-copper (rGOC) nanocomposites by in situ reduction as a novel and efficient biocide [65]. A loose NF membrane was designed using a fast (2 h) biomimetic strategy, in which the rGOC nanocomposite was tightly co-deposited with polydopamine (PDA) on the ultrafiltration support. After the PDA-rGOC-modified membrane was contacted with *Escherichia coli* for 3 h, the rGOC-functionalized membrane exhibited strong antibacterial properties, reducing the number of viable *Escherichia coli* bacteria by 97.9%. Combined with the release of Cu ions, the antibacterial activity of GO-functionalized films was enhanced by subsequent ROS-induced oxidative stress in the rGOC composite.

The photocatalytic properties of metal oxide–graphene composites have also been exploited for antibacterial applications, and increasing the heterojunction density can improve antibacterial activity due to enhanced absorption in the visible region [5]. Metal oxides can kill bacteria by releasing metal ions and generating ROS upon contact with bacteria. The adhesion of graphene oxide to bacterial membrane surfaces is inherently repulsive, and these studies demonstrate that graphene nanostructures can disrupt bacterial membranes to a degree that depends on their size and composition. Abadikhah et al. prepared a multifunctional thin-film nanocomposite (MTFN) by incorporating a novel nanocomposite-structured rGO@TiO_2@Ag into the PA active layer [66]. This film has a high desalination performance, dye retention and antimicrobial properties. The special properties of graphene-based nanocomposites synthesized by a microwave-assisted irradiation process facilitate water channelization and provide superhydrophilic and antibacterial properties for MTFN membranes. Zhao et al. fabricated GO-based composite films with high antifouling properties using a phase inversion process [67]. Strongly hydrophilic GO and antibacterial copper oxide (Cu_xO) were used as nanofillers and blended with PVDF to obtain hybrid membranes with excellent antibacterial properties. Due to the hydrophilic and bactericidal properties of the membrane surface, the composite membrane exhibits excellent antifouling properties, including higher flux recovery, better resistance to accumulated pollutants, and lower filtration resistance, especially lower irreversible resistance. The antifouling properties, especially the anti-irreversible scaling properties, are significantly improved, showing great engineering potential.

4.2. Polymers

Graphene can be coupled with a variety of polymers to form membrane materials [68–71]. By optimizing the properties of graphene and polymers and the way of conjugation, the physical and chemical properties of graphene composite membranes can be optimized. Lim et al. prepared covalently cross-linked GO membranes using a facile vacuum filtration method, and cross-linked tannic acid-functionalized GO (TA-GO) and hyperbranched polyethyleneimine (PEI) [72]. The cross-linked GO membrane exhibits good dimensional stability and ion separation performance. TA-containing cross-linked GO films exhibited excellent antibacterial activity against *Escherichia coli* due to the inherent bactericidal properties of the TA moiety. Zhang et al. prepared covalently cross-linked GO membranes by vacuum-assisted filtration self-assembly after the functionalization of dopamine (GO-PDA) and branched polyethyleneimine (GO-PDA-PEI) [73]. Compared with GO and GO-PDA, the GO-PDA-PEI film exhibited extraordinary stability. Notably, Graphene oxide exhibits excellent photothermal properties under 795 nm near-infrared laser irradiation. Under 795 nm near-infrared (NIR) laser irradiation, the GO-PDA-PEI membrane exhibited excellent antibacterial properties against both Gram-positive *Staphylococcus aureus* and Gram-negative *Escherichia coli*, with an antibacterial efficiency of over 99%.

Kaneda et al. developed a facile method to graft biocidal GO onto chemically inactive membrane materials using benzophenone as an anchor [74]. The GO nanosheets undergo an amide coupling reaction and are functionalized with benzophenone. Then, the functionalized GO nanosheets were grafted onto the inert membrane surface via benzophenone-induced cross-linking, which was irreversible under UV irradiation (Figure 3a). When exposed to model bacteria (*Escherichia coli*), the GO-functionalized PVDF and polysulfone membranes exhibited robust antibacterial activity, reducing the number of viable cells by 90% and 75%, respectively, compared to the pristine membrane. He et al. prepared a bilayer polymer membrane with blood compatibility and antibacterial properties by coating a GO and sulfonated polyanion co-doped hydrogel film (GO-SPHF) on the underlying membrane substrate [75]. The double-layer membrane showed strong antibacterial ability, and had an excellent bactericidal ability for both *E. coli* and *S. aureus*. Zhang et al. developed an ultrafiltration PES membrane with dual antifouling and antibacterial properties (Figure 3b) [76]. First, the ampholyte hydrogel was UV-grafted onto the PES membrane surface (p-PES membrane). Then, the hydrogel was loaded into GO nanosheets using a vacuum filtration strategy (GO-p-PES membrane). Static adsorption and dynamic filtration experiments showed that p-PES membranes had similar organic fouling propensities to GO-p-PES membranes. Furthermore, the GO loading induced the antibacterial properties of the membrane, as demonstrated by contact kill and antibiotic fouling filtration experiments.

4.3. Biopolymers

Biopolymers have been proven to be excellent dopants for antimicrobial materials due to their rich functions, diverse nanostructures, and excellent biocompatibility [77]. Chitosan is a polysaccharide extracted from the exoskeleton of crustaceans and the mycelium of some fungi. Due to its unique properties, chitosan is considered a promising reagent for molecular separation, food packaging, artificial skin, bone substitutes, water treatment, electrochemical sensors, biosensors, and several other applications. Chitosan is an interesting graphene immobilization matrix, which can be modified by chemical methods such as cross-linking, complexation and grafting, and different functional groups can also be introduced. Grande et al. studied the synthesis of chitosan-GO (CS-GO) nanocomposite films and their application prospects in food packaging [78]. Nanocomposite films were prepared by crosslinking GO and chitosan at a high temperature (120 °C). The results showed that the CS-GO membrane had better antibacterial properties against Gram-negative *Escherichia coli* and *Gram-positive Bacillus subtilis*. The GO/CS/ZnO composite exhibited impressive antibacterial activity against *Escherichia coli* and *Staphylococcus aureus*, and excellent adsorption performance for methylene blue [79].

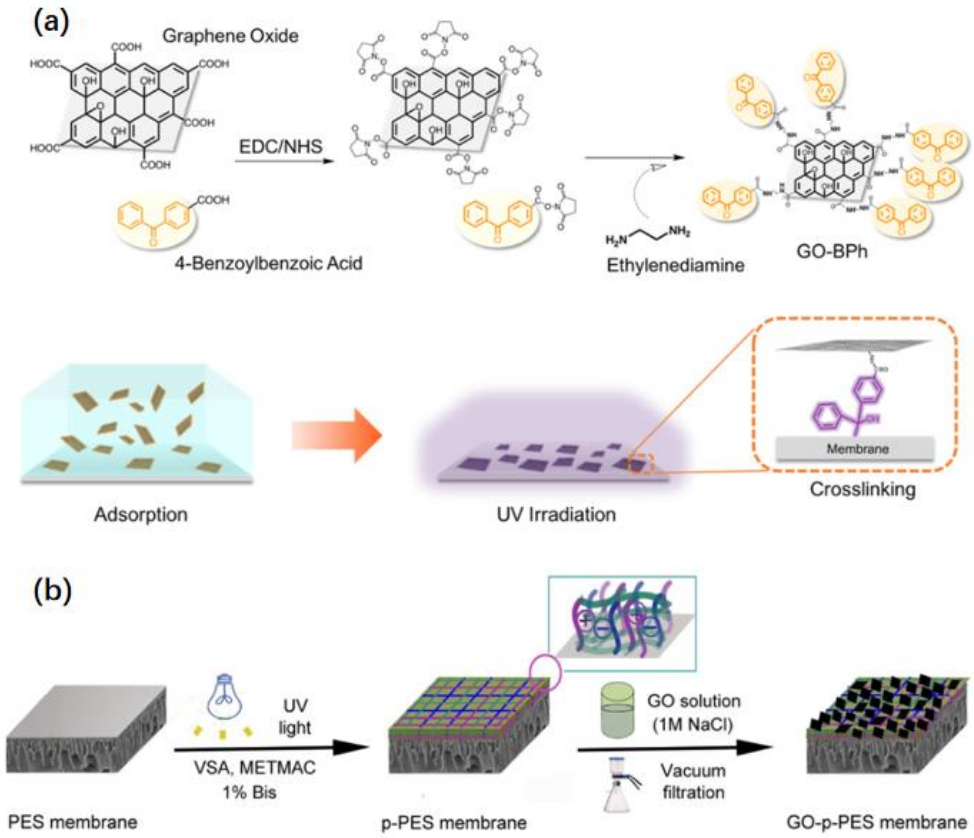

Figure 3. (**a**) Schematic diagram of the chemical surface modification process of benzophenone. (Printed image with permission from Ref. [74], copyright 2019, American Chemistry Society). (**b**) Schematic illustration of the two-step synthesis process of GO functionalized zwitterionic polyampholyte hydrogel polyethersulfone (PES) membrane. (Printed image with permission from Ref. [76], copyright 2018, Elsevier Ltd.).

Sreeprasad et al. studied a composite material containing GO, lactoferrin (LF) and chitosan [80]. In this composite, LF and chitosan enhanced the toxicity of GO to bacteria and reduced bacterial viability. Xie et al. investigated the synthesis, structure, and antibacterial properties of a novel nanocomposite film composed of bacterial cellulose (BC), GO, and copper oxide (CuO) nanosheets [81]. Nanocomposite films were synthesized by combining GO-CuO nanohybrid with BC matrix using a uniform blending method. The antibacterial activity of the nanocomposites against Gram-positive bacteria was superior to that of Gram-negative bacteria. The antibacterial activity of the BC/GO-CuO nanocomposite was higher than that of BC/CuO, and the antibacterial mechanism of the nanocomposite was elucidated. Lin et al. selected hydroxypropyl cellulose (H), CS and polyoxyethylene (P) as membrane materials to improve the hydrophilicity, antibacterial properties and yield of nanofibers, respectively [82]. In addition, graphene (G) was added to enhance the antibacterial properties of the membrane. The antibacterial activity of the membrane was demonstrated by experiments against *Escherichia coli* and *Staphylococcus aureus*. The highly hydrophilic HCP and HCPG membranes prevented bacterial adhesion. The presence of such membranes, especially graphene-embedded HCPG membranes, also greatly reduced bacterial growth. The small pore size of the HCP and HCPG nanofibrous membranes prevented bacterial penetration and infection.

4.4. Others

Yan et al. developed a spray-mediated, bio-inspired Ag assembly on reduced graphene-sodium alginate nanocomposite films for efficient wound healing [83]. The composite membrane can effectively inactivate *PAO1*, *Escherichia coli* and *C. albicans*, and has the

ability to protect wounds from infection by pathogenic microorganisms. Geng et al. studied the direct deposition of graphene films on germanium surfaces with different coverage areas by controlling the growth time [84]. Compared with bare graphene, the presence of graphene films enables graphene to have a good antibacterial ability against *S. aureus* and acceptable antibacterial ability against *E. coli*, and the underlying mechanism is thought to be phospholipids. The combined effect of interference and electron extraction occurs at the interface between graphene and biofilm (Figure 4). Electrostatic interactions between positively charged pristine graphene and negatively charged bacterial membranes bind the phospholipids to the graphene membranes. At the same time, this contact may cause translocation and the flipping of phospholipids, allowing for phospholipids to interact with hydrophobic tails due to hydrophobic interactions, further disrupting the function of the membrane. Dhanasekar et al. found that nanocomposites containing PVA, rGO, and Cu_2O or TiO_2 exhibited good inhibition against *Staphylococcus aureus*, *oral Streptococcus*, *Escherichia coli*, and *POA1* [85].

Zhu et al. synthesized zeolite imidazole framework-8 (ZIF-8) on the surface of GO by an in-situ synthesis, and used it as a novel efficient fungicide to prepare antibacterial membranes using the interfacial polymerization (IP) process [86]. The results showed that the ZIF-8/GO nanocomposites have good antioxidant properties, and the improvement in the antibacterial efficiency of ZIF/GO functionalized films was mainly due to the synergy of ZIF-8 (Zn^{2+} release) and GO (oxidative stress) towards the antibacterial effect. In another study, Kanchanapally et al. recently reported a 3D porous membrane composed of GO and the antimicrobial peptide nisin, which was able to efficiently identify, separate, and disinfect water from methicillin-resistant *Staphylococcus aureus* (MRSA) [29]. In the study by Peng et al. [87], hydrophilic metal–organic framework (UiO-66) nanoparticles were embedded into GO layers as microporous fillers to form ultrathin "sandwich" membranes to improve forward osmosis (FO) performance, the results showed that the nano-thick film matrix formed by the GO layer has antibacterial activity (90% antibacterial activity).

Zahid et al. prepared a novel, functionalized, GO-based cellulose acetate (CA) membrane using a phase inversion method [88]. The addition of a hydrophilic aminated graphene oxide (AGO) additive to the CA film enhanced the antibacterial activity of the AGO-CA film and improved the thermal stability of the film. Zhang et al. fabricated a series of TFN reverse osmosis membranes by incorporating nanofiller p-aminophenol-modified graphene oxide (mGO) into the PA skin layer by interfacial polymerization [89]. The bactericidal phenolic functional group's effect on mGO, the oxidative stress response of bacteria and the hydroxyl group's effect on aminophenol can cause an antibacterial effect. Statistics show that the sterilization rate of TFN reverse osmosis membrane with GO on *Escherichia coli* and *Staphylococcus aureus* with the addition of 0.005 wt% is 96.78% and 95.26%, respectively, which is much higher than the reverse osmosis membrane with GO added and original reverse osmosis membrane. This study provides a facile and feasible method for preparing reverse osmosis membranes with good separation performance and antibacterial properties.

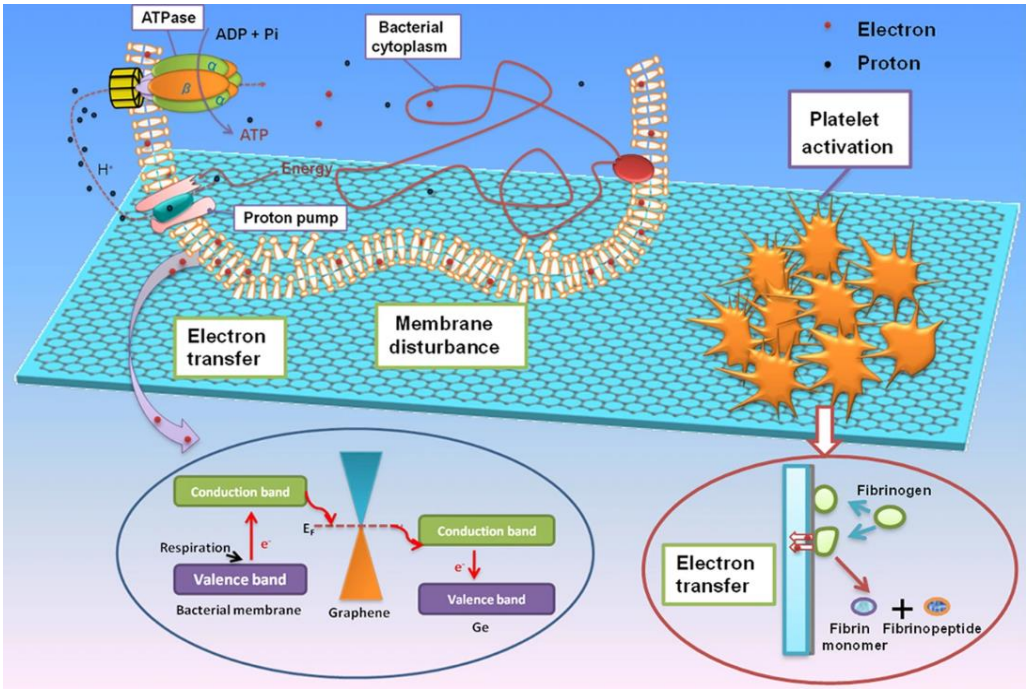

Figure 4. Schematic diagram of the interaction between bacteria (platelets) and graphene films covered with graphene substrates. (Printed image with permission from Ref. [84], copyright 2016, Nature Publication).

5. Antimicrobial Applications

5.1. Antimicrobial Mechanism

Graphene inactivates bacteria that contact it. Li et al. changed the conductive properties of the substrate on which the graphene films were deposited, compared the bactericidal effect, and demonstrated the charge transfer mechanism [90]. Interestingly, the antibacterial activity was clearly dependent on the electrical conductivity of the graphene metal substrate, and the inhibitory effects on the growth of *Staphylococcus aureus* and *Escherichia coli* were increased in the order of graphene/SiO_2 < graphene/Ge < graphene/Cu. When bacterial cells come into contact with graphene materials over time, they may eventually be destroyed due to several mechanisms: sharp-edged cutting effects, oxidative stress, cell trapping, and encapsulation sequestration. These mechanisms can act individually or together to inhibit bacterial growth in a bactericidal or bacteriostatic manner. A comprehensive understanding of its antibacterial mechanism is crucial to guide the design of graphene-based membrane nanomaterials for practical applications. Figure 5 outlines the antibacterial mechanisms of three common graphene-based materials and illustrates them in the following sections [91].

5.1.1. Sharp Edge Cutting

In brief, graphene-based membrane nanocomposites have sharp edges that physically damage the cell membrane when in direct contact with bacterial cells [91]. Pham et al. found that factors affecting the antibacterial activity induced by the sharp edges of graphene-based nanomaterials include edge density and the angle at which flakes are in contact with the cell membrane. The edge length is the lateral dimension of the two sides of the exfoliated graphene film [92]. The density of graphene edge length is called edge density, which is positively correlated with the antibacterial activity of graphene-based nanomaterials. They concluded that graphene with smooth edges has a greater edge density and better antibacterial effect than rough graphene. They concluded that when the contact angle was 37°, the graphene-based nanomaterials began to show antibacterial effects, and reached the biggest consequent at 90°. Besides the direct cleavage effect, which affects the

antibacterial activity, the charge transfer between the microbial cell membrane and the edge of graphene-based nanomaterials may also be beneficial to increase the antibacterial activity. Akhavan et al. showed that the reduced graphene-based nanomaterials are more damaging to bacterial cell membranes than the unreduced graphene-based nanomaterials, because of the stronger charge transfer effect between the edges of the reduced graphene-based nanomaterials and the cell membrane [93].

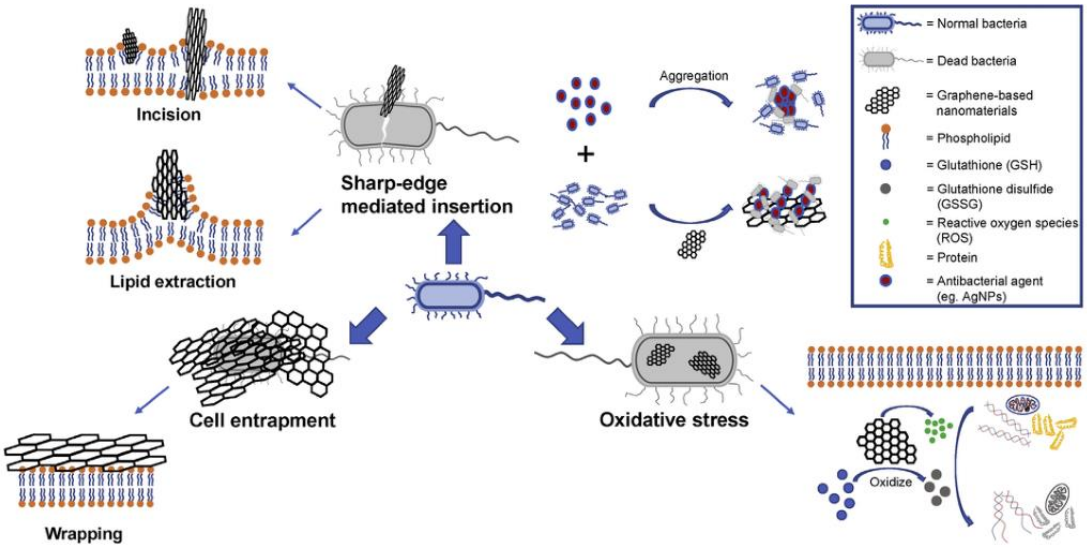

Figure 5. Schematic diagram of the antibacterial mechanism of graphene-based nanomaterials, including sharp edge cutting effect, oxidative stress and cell trapping. (Printed image with permission from Ref. [91], copyright 2019, Elsevier Ltd.).

5.1.2. Oxidative Stress

The main cause of microbial death is oxidative stress [94]. Due to the large surface area of graphene sheets, they can act as conductive bridges on which redox charge transfer occurs, further enhancing the oxidative stress potential [5]. Oxidative stress usually causes an imbalance between oxidation and antioxidants, which slows down the bacterial metabolism, disrupting cellular structure, and ultimately causing the bacteria to lose their vitality [91]. Gurunahan et al. showed that, after exposure to GO and rGO, glutathione (GSH) content was significantly reduced and ROS content increased by 3.8-fold and 2.7-fold in *POA1*, respectively [95]. Graphene-based membrane nanomaterials generate large amounts of ROS and are thought to be responsible for bacterial oxidative stress. It is generally believed that GO produces more ROS and possesses a higher GSH oxidation capacity than reduced GO. However, Liu et al. found that the GSH concentration in *E. coli* decreased while the ROS concentration did not increase after exposure to four graphene-based membranes [96]. Interestingly, conductive rGO and graphite have higher oxidative capacity for GSH than insulating GO and graphite oxide.

5.1.3. Cell Trapping

Another major mechanism for the antibacterial activity of graphene-based membranes is cell trapping, which describes bacterial cells being captured by graphene-based membrane sheets when they come into contact with them. These bacteria are isolated from the external environment, and the way they get nutrients is limited. This entrapment effect is related to the size of the graphene-based membranes. Larger graphene oxide flakes have a stronger antibacterial effect, which is attributed to the ability of larger graphene oxide flakes to completely cover bacterial cells and prevent their proliferation. The study found that the antibacterial activity of GO increased with the increase in its lateral size [97]. It is possible that the trapping effect temporarily limits the growth of bacteria rather than killing them. Interestingly, it was found that the size of graphene-based membranes has a

significant and complex effect on its antibacterial activity by affecting both cleavage efficacy and cell-trapping efficiency. Simply put, a large size favors the entrapment effect, but a small size is equivalent to high edge density, which is good for the cutting effect. The size of GO and rGOs also affect their UV absorption, probably because ultrasound reduces the size during oxidation. The size of GO and rGO also affects their UV absorption, which may be due to the possible size reduction caused by ultrasound during the oxidation process [91].

5.1.4. Parcel Isolation

Chen et al. studied the antimicrobial activity of GO to explore its mechanism of activity and reveal its strong inhibitory effect on the studied microorganisms [98]. GO killed about 90% of bacteria and inhibited about 80% of macroconidia germination. At a concentration of 500 μg/mL, some cells swelled and lysed. They propose a mechanism by which the interweaving of fungal and bacterial spores leads to local perturbations of the cell membrane. Intertwining leads to the leakage of fungal spore electrolytes and a decrease in bacterial membrane potential. They believe that one of the main toxic effects of GO on plant pathogens could be that GO interacts with pathogens through mechanical encapsulation, locally disrupting cell membranes, and causing cell lysis.

5.2. Antibacterial Applications

As the human microbiome develops, associations between certain bacteria and specific human diseases are gradually being established. *Escherichia coli* and *Staphylococcus aureus* are associated with infections in different parts of the body [99]. *Streptococcus mutans*, *Porphyromonas gingivalis*, and *Candida albicans* have been associated with oral disease [100–102]. *POA1* and *Klebsiella pneumoniae* are typical nosocomial pathogens [103,104]. *Salmonella typhimurium* is associated with gastrointestinal disease [105]. Due to its special physicochemical properties and unique antibacterial mechanism, graphene membrane material has great potential to become a novel antibiotic-independent antibacterial drug. The antibacterial mechanisms of graphene nanomaterials are diverse, and the main antibacterial mechanisms against different bacteria may differ. GO and rGO are toxic to both Gram-positive (*Staphylococcus aureus*) and Gram-negative (*Escherichia coli*) bacteria. Previous studies have shown that the inhibition of cell division may be the main antibacterial mechanism against Gram-positive bacteria, and mechanical damage may be the main antibacterial mechanism against Gram-negative bacteria [106]. The study by Karahan et al. showed that the antibacterial activity of GO was clearly dependent on the cellular physiology of Gram-negative and positive bacteria [107]. The sensitivity of bacteria to GO is highest in the exponential growth phase (i.e., the physiological growth phase), while cells in the stationary growth phase (i.e., the non-growing phase) have considerable resistance to GO. Importantly, the order of *E. coli* susceptibility to GO during growth stages is closely related to changes in the ultrastructure of the cell envelope.

Graphene-based membranes can be used to effectively inhibit the growth of planktonic *Streptococcus mutans* and their biofilm formation. For instance, Bregnocchi et al. synthesized graphene nanosheets (GR) and found that an oral adhesive filled with a graphene-based film had a significant inhibitory effect on the adhesion, growth and even biofilm formation of Streptococcus mutans [108], and that oral adhesives filled with graphene-based films exhibited a significant inhibition of adhesion. In the study of Huang et al. [109], azide-functionalized AGO was covalently immobilized on the surface of a reverse osmosis membrane (RO) by azide photochemistry. Surface modification was performed by coating the reverse osmosis membrane with an aqueous dispersion of AGO followed by UV exposure under ambient conditions (Figure 6a). This simple process produces a hydrophilic, smooth, antimicrobial membrane with limited reductions in water permeability or salt selectivity. The biofouling of GO-RO membranes was reduced 17-fold after exposure to *E. coli* for 24 h. Zhang et al. developed a simple, one-step probe sonication method to synthesize ZnO nanoparticle-functionalized rGO-ZnO nanocomposites by exfoliating graphite oxide in the presence of Zn^{2+} [110]. The antibacterial activity of nanocomposites against Gram-negative

bacteria (*Escherichia coli* and *Serratia marcescens*) and Gram-positive bacteria (*Bacillus subtilis*) is higher than that of GO and rGO. Furthermore, the incorporation of rGO-ZnO nanocomposites into PES membranes can inhibit the growth of biofilms compared to pristine PES membranes. To improve the anti-biofouling performance of ultrafiltration membranes, Liu et al. synthesized quaternized graphene oxide (QGO) as a membrane modified by the in-situ growth of quaternary ammonium salts on the surface of ultrafiltration membranes. For the first time, QGO with hydrophilic and antibacterial properties was prepared by chemically grafting quaternary ammonium salt groups with carboxyl and epoxy groups on graphene oxide as active sites [111]. Using QGD as a modifier, a hydrophilic antibacterial bifunctional membrane was prepared by the dipping phase inversion method. Compared with the unmodified PVDF membrane, the hydrophilicity, antibacterial property and mechanical properties of the modified membrane were significantly enhanced. Najjar et al. proposed a novel PES membrane prepared by the phase inversion method with different loadings of GO and 1 wt% gum arabic (AG) as nanofillers and pore formers [112]. The prepared membranes were tested for contamination with bovine serum albumin (BSA) solution, model bacterial suspensions of Gram-positive bacteria (*Bacillus subtilis*) and Gram-negative bacteria (*Escherichia coli*), and actual treated sewage (TSE) conduct a biofouling test. The results show that the novel PES/GO membrane has strong hydrophilicity and a negative surface charge, and increases the porosity, pore size, and water flux. The PES/GO membrane showed a superior antibacterial effect against both Gram-positive and Gram-negative bacteria, which means that the PES membrane with GO and AG as the novel membrane has high-throughput and high antibacterial properties. Zhang et al. prepared a novel synergistic antibacterial guanidine-functionalized graphene/polysulfone (GFG/PSF) mixed-matrix ultrafiltration membrane by a non-solvent-induced phase separation method (Figure 6b) [113]. The guanidized graphene nanosheets were prepared by a two-step grafting method of amination and guanidine. The graphene nanosheets showed high dispersibility in the casting solution and good compatibility with the polymer matrix. The bis-diester binding between guanidine and phosphate groups on the cell wall that it induces allows for high bactericidal rates, even at low concentrations. Compared with pure PSF membrane, GFG/PSF mixed matrix membrane not only exhibits excellent permeability and remarkable antifouling performance against BSA, but also exhibits good antibacterial activity against *Escherichia coli* and *Staphylococcus aureus* and long-term duration.

GOQDs with hydrophilic groups and single-atom-thick structures have attracted extensive interest in the field of membrane preparation. By providing additional water channels in the membrane, GOQDs can increase the permeability of the membrane without compromising the selectivity. As an effective bactericidal material, silver nanoparticles were uniformly deposited on GOQDs by a facile method. Yu et al. used interfacial polymerization to embed GOQD/Ag into a PA selective layer to obtain a nanocomposite thin film (TFN-GOQD/Ag) film [114]. Due to the synergistic effect of GOQD and Ag, the TFN-GOQD/Ag200 membrane exhibited significant bactericidal ability against both Gram-negative *Escherichia coli* (98.6%) and Gram-positive *Staphylococcus aureus* (96.5%).

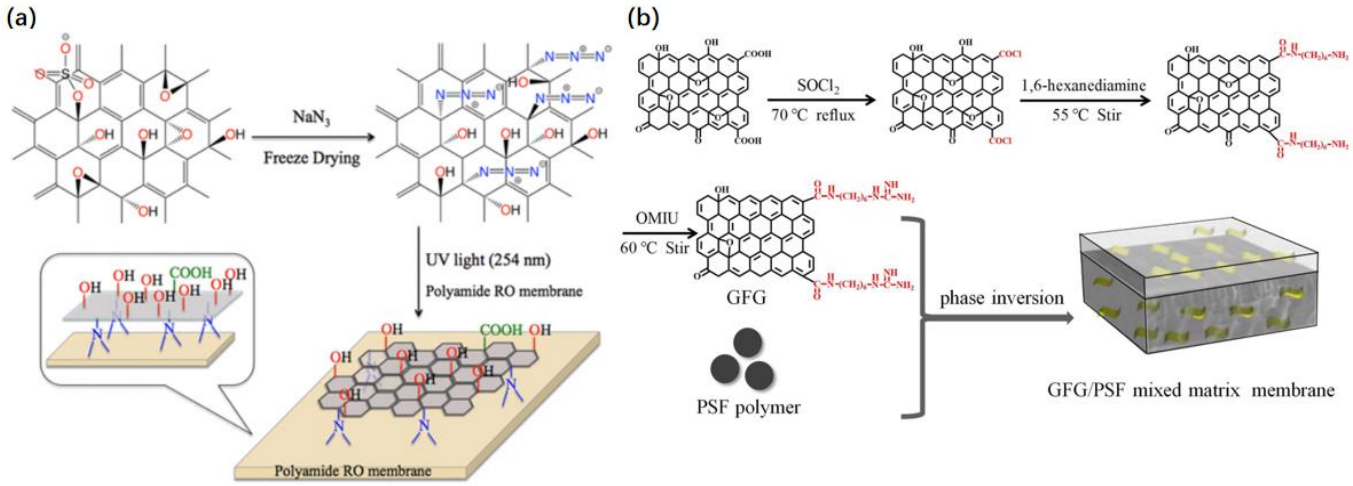

Figure 6. (**a**) Synthesis of AGO and its UV activation on the surface of PA reverse osmosis membrane. (Printed image with permission from Ref. [109], copyright 2016, American Chemistry Society). (**b**) Synthetic route of GFG/PSF mixed matrix membrane. (Printed image with permission from Ref. [113], copyright 2019, Elsevier Ltd.).

5.3. Antiviral Applications

In general, GO and its derivatives have broad-spectrum antiviral properties, such as against positive-sense and negative-sense viruses, RNA and DNA viruses, enveloped and non-enveloped viruses, etc. Therefore, these materials have great potential for developing antiviral surfaces and coatings to prevent contamination by virulent and infectious viruses, including *SARS-CoV-2*, and to control disease transmission. Graphene has shown potential to fight viral diseases by developing excellent diagnostic devices, and to control the spread of infection by developing various components and coatings. Goswami et al. revealed the development of masks with air filters fitted with functionalized graphene (fG) on 3D-printed mask replicas [115]. Mask replicas were fabricated using Fused Deposition Modeling (FDM). The fG combined with nanosheets has additional adsorption capacity, with a higher surface area to volume ratio. The fG coating was applied on polypropylene (PP) cloth by the dip-coating method to enhance antiviral and antibacterial properties.

Valentini et al. evaluated the filtration effect of GO-coated textiles on a model of *human herpesvirus 6A* (*HHV-6A*) infection of human glioblastoma cells (U373) [116]. Textiles coated with different amounts of GO were able to reduce the *HHV-6A* infection of U373 cells in a dose-dependent manner. The antifouling, antiviral and antibacterial properties of LIG have been proven in air and water filtration applications. Gupta et al. developed a polyimide (PI) nonwoven supported laser-induced graphene (LIG) air filter with a negligible change in pressure drop compared to nonwoven supported materials, while a low current density can inhibit aerosolized bacteria [117]. Its low-pressure antibacterial mechanism was elucidated through antibacterial experiments on titanium surface and LIG surface prepared on high-density PI film. The results show that, at extremely low voltages, low current density levels are sufficient to inactivate a high number of bacteria and viruses. Das Jana et al. found that copper-graphene (Cu-Gr) nanocomposites possess strong antiviral activity [118]. Using PVA as a confinement agent, we were able to generate a highly transparent coating based on Cu-Gr nanocomposites that retains the original antiviral activity in solid form, thus potentially realizing a wide variety of surfaces to reduce the spread of respiratory viral infections (Figure 7).

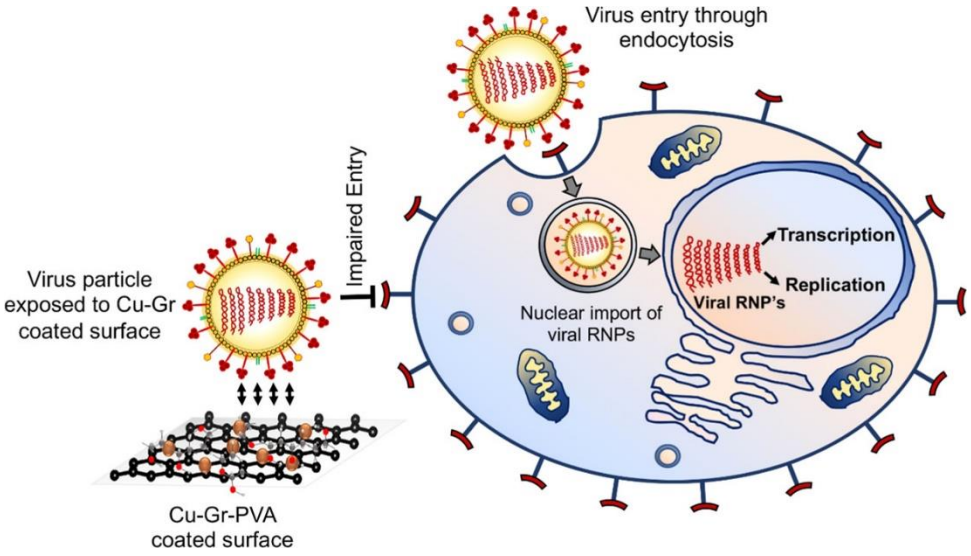

Figure 7. Mechanism of action of Cu-Gr coating on antiviral surfaces. (Printed image with permission from Ref. [118], copyright 2020, American Chemistry Society).

5.4. Antifungal Applications

In the study of Montes et al. [119], cellulose nanocrystals (CNC) and GR were combined in two different ratios and added to polylactic acid (PLA) using the melt-blending technique. The obtained PLA-CNC/GR nanocomposites were hot-pressed to prepare films, their antifungal properties against aspergillus niger were evaluated, and it was found that the CNC/GR hybrid films had better antifungal activity. Huang et al. aimed to introduce GO to oral PE membranes for the delivery of antifungal drugs and investigated the effect of GO on membrane properties, drug release properties, and antifungal activity [120]. Using chitosan and alginate as binders, a PE membrane was formed with the antifungal drug clotrimazole. The excipients were mixed with clotrimazole, and the oral formulation was prepared by probe sonication, followed by film casting and drying. The results showed that the electrostatic hydrophobic interaction between GO and clotrimazole also affected the antifungal effect of clotrimazole. Gontarek-Castro et al. prepared several graphene/polyvinylidene (PVDF/G) films with different graphene loadings (0–10 wt%) by a phase-inversion method [121]. The antifungal activity test of PVDF/G film showed that the inhibition rate of PVDF/G film against *curvularia* strain was increased. However, the antifungal surface properties were found to be the result of a synergistic effect of graphene toxicity and surface topography. The GO-AgNPs nanocomposites synthesized by Chen et al. were first utilized to investigate *graminearum*. Additionally, for the first time, the antibacterial activity of GO-AgNPs against wheat head blight was studied in vitro and in vivo. Damage to spores and hyphae, possibly due to the remarkable synergy of GO-AgNPs, created an antibacterial mechanism, resulting in physical damage and the production of chemical reactive oxygen species. More importantly, the fungal spore-mediated chemical reduction in GO may be responsible for its high antibacterial activity. The in vitro leaf experiments confirmed that the nanocomposite can significantly control *graminearum* and protect plants from pathogen infection (Figure 8) [122].

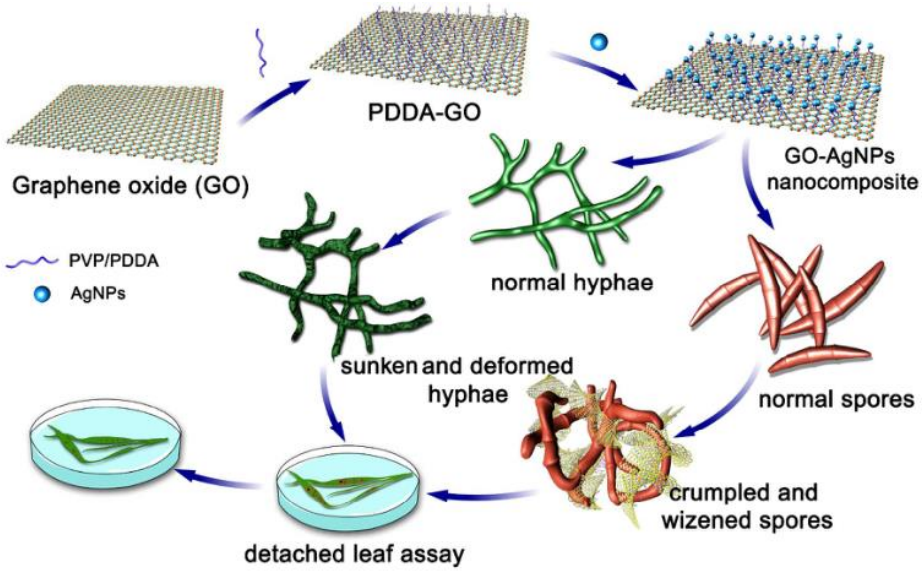

Figure 8. Schematic diagram of the preparation of graphene-AgNPs nanocomposites and their antifungal properties in vitro and in vivo. (Printed image with permission from Ref. [122], copyright 2016, American Chemistry Society).

The above introduction on antimicrobial applications of graphene-based membranes is summarized in Table 1.

Table 1. Antimicrobial materials, activity, and application of graphene-based membranes.

GMBs	Pathogens	Comments	Ref.
GNPs	*Streptococcus mutans*	Inhibit adhesion and growth	[108]
GO-RO	*Escherichia coli*	Reduced water permeability, reduced salt selectivity, reduced pollution	[109]
rGO-Zno-PES	*Escherichia coli* *Serratia marcescens* *Bacillus subtilis*	Inhibit biofilm growth	[110]
QGO	*Escherichia coli*	Hydrophilic, antibacterial and mechanical properties are significantly enhanced	[111]
GO-AG-PES	*Escherichia coli* *Bacillus subtilis*	Superior antibacterial effect	[112]
GFG/PSF	*Escherichia coli* *Staphylococcus aureus*	Good antibacterial activity, long duration	[113]
TFN-GOQD/Ag200	*Escherichia coli* *Staphylococcus aureus*	Significant bactericidal power	[114]
fG-PP	*SARS-CoV-2*	Good inhibition effect	[115]
GO-textile	*HHV-6A*	Prevent cell infection, has antiviral properties	[116]
LIG-PI	*Pseudomonas aeruginosa* *T4 virus*	Inactivation of a large number of bacteria and viruses	[117]
Cu-Gr	*Influenza virus*	Virus particles are inactivated, slowing infection	[118]
PLA-CNC/GR	*Aspergillus niger*	High antifungal activity	[119]
CS/alginate/GO/clotrimazole	*Oral candidiasis*	GO can be used as a functional excipient for delivery of antifungal drugs.	[120]
PVDF/G	*Curvuria*	High inhibition rate	[121]
GO/AgNP	*graminearum*	High antibacterial activity and inhibit pathogen infection	[122]

6. Conclusions and Outlook

In conclusion, in this review, we comprehensively and systemically summarize the synthesis and functional tailing of graphene-based membranes, as well as their applications in antimicrobial fields. From the above introduction and discussion, it can be concluded that macroscopic graphene-based membranes can be successfully synthesized by traditional techniques such as CVD, vacuum-assisted filtration, spin-coating, drop-casting, and LBL self-assembly. The addition of other functional nanomaterials such as metals, metal oxide, polymers, biomolecules, and biopolymers, enables the functional regulation of graphene-based membrane materials with an improved antimicrobial performance, and the nanocomposite-based hybrid membranes can overcome the limitations of individual component-based graphene membranes. The graphene-based functional membranes exhibit excellent antibacterial, antiviral and antifungal effects. In addition, we introduce the mechanisms of graphene-based membranes for antimicrobial resistance, which are crucial to guide the design and fabrication of graphene-based membranes for biomedical applications.

Although great achievements have been made regarding the antimicrobial applications of graphene-based membranes in recent years, it is necessary for researchers to develop novel techniques and materials to promote the development of this promising research topic. First, it is necessary to modify graphene materials with highly active biomolecules such as antimicrobial biopolymers and antimicrobial peptides to enhance the biological properties of graphene-based membranes. The addition of these biomolecules could improve the biocompatibility and antimicrobial performances of graphene-based membranes. Second, the fabrication techniques of 2D graphene membranes should be developed for large-scale production, with a low cost and high sustainability, as these are highly required for practical applications in biomedical and other fields. Third, graphene-based new materials should be studied to improve and optimize the performance of graphene-based hybrid membranes, which are expected to only show toxicity to the microorganisms, but reveal high biocompatibility with human cells, to realize the next generation of the most advantageous antimicrobial materials. Fourth, other 2D materials, such as transition metal oxides, MXenes, metal–organic frameworks (MOFs), and others could be utilized to fabricate functional 2D membranes for antimicrobial applications, which could lead to surprising findings and effects in the future.

Author Contributions: Conceptualization, G.W. and H.K.; software, X.Z. and H.K.; resources, X.Z. and H.K.; data curation, X.Z., G.Y., D.Z., X.L. and P.H.; writing—original draft preparation, X.Z. and H.K.; writing—review and editing, X.Z., H.K. and G.Y.; supervision, H.K. and G.W.; project administration, G.W.; funding acquisition, G.W. All authors have read and agreed to the published version of the manuscript.

Funding: The authors thank the financial support from the National Natural Science Foundation of China (No. 51873225), the Taishan Scholars Program of Shandong Province (No. tsqn201909104), and the High-Grade Talents Plan of Qingdao University. This work is partly supported by the Innovation and Entrepreneurship Program for College Students of Qingdao University.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

Acknowledgments: The authors acknowledge the Endnote editing support by Bin Liu and Yun Chen (Qingdao University).

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Fatima, N.; Qazi, U.Y.; Mansha, A.; Bhatti, I.A.; Javaid, R.; Abbas, Q.; Nadeem, N.; Rehan, Z.A.; Noreen, S.; Zahid, M. Recent developments for antimicrobial applications of graphene-based polymeric composites: A review. *J. Ind. Eng. Chem.* **2021**, *100*, 40–58. [CrossRef]
2. Dong, A.; Wang, Y.J.; Gao, Y.Y.; Gao, T.Y.; Gao, G. Chemical Insights into Antibacterial N-Halamines. *Chem. Rev.* **2017**, *117*, 4806–4862. [CrossRef] [PubMed]
3. Hegab, H.M.; ElMekawy, A.; Zou, L.; Mulcahy, D.; Saint, C.P.; Ginic-Markovic, M. The controversial antibacterial activity of graphene-based materials. *Carbon* **2016**, *105*, 362–376. [CrossRef]
4. Sweileh, W.M. Global research publications on irrational use of antimicrobials: Call for more research to contain antimicrobial resistance. *Glob. Health* **2021**, *17*, 94. [CrossRef] [PubMed]
5. Rojas-Andrade, M.D.; Chata, G.; Rouholiman, D.; Liu, J.; Saltikov, C.; Chen, S. Antibacterial mechanisms of graphene-based composite nanomaterials. *Nanoscale* **2017**, *9*, 994–1006. [CrossRef]
6. Kong, H.; Chen, Y.; Yang, G.Z.; Liu, B.; Guo, L.; Wang, Y.; Zhou, X.; Wei, G. Two-dimensional material-based functional aerogels for treating hazards in the environment: Synthesis, functional tailoring, applications, and sustainability analysis. *Nanoscale Horiz.* **2022**, *7*, 112–140. [CrossRef]
7. Li, T.; Yin, W.; Gao, S.W.; Sun, Y.N.; Xu, P.L.; Wu, S.H.; Kong, H.; Yang, G.Z.; Wei, G. The Combination of Two-Dimensional Nanomaterials with Metal Oxide Nanoparticles for Gas Sensors: A Review. *Nanomaterials* **2022**, *12*, 982. [CrossRef]
8. Diez-Pascual, A.M. Antibacterial Action of Nanoparticle Loaded Nanocomposites Based on Graphene and Its Derivatives: A Mini-Review. *Int. J. Mol. Sci.* **2020**, *21*, 3563. [CrossRef]
9. Agarwal, V.; Zetterlund, P.B. Strategies for reduction of graphene oxide—A comprehensive review. *Chem. Eng. J.* **2021**, *405*, 127018. [CrossRef]
10. Su, W.T.; Kumar, N.; Krayev, A.; Chaigneau, M. In situ topographical chemical and electrical imaging of carboxyl graphene oxide at the nanoscale. *Nat. Commun.* **2018**, *9*, 2891. [CrossRef]
11. Fatima, Q.; Haidry, A.A.; Yao, Z.J.; He, Y.; Li, Z.; Sun, L.C.; Xie, L.J. The critical role of hydroxyl groups in water vapor sensing of graphene oxide. *Nanoscale Adv.* **2019**, *1*, 1319–1330. [CrossRef]
12. Yang, G.Z.; Kong, H.; Chen, Y.; Liu, B.; Zhu, D.Z.; Guo, L.; Wei, G. Recent advances in the hybridization of cellulose and carbon nanomaterials: Interactions, structural design, functional tailoring, and applications. *Carbohydr. Polym.* **2022**, *279*, 118947. [CrossRef] [PubMed]
13. Ege, D.; Kamali, A.R.; Boccaccini, A.R. Graphene Oxide/Polymer-Based Biomaterials. *Adv. Eng. Mater.* **2017**, *19*, 1700627. [CrossRef]
14. Nanda, S.S.; Yi, D.K.; Kim, K. Study of antibacterial mechanism of graphene oxide using Raman spectroscopy. *Sci. Rep.* **2016**, *6*, 28443. [CrossRef]
15. Yaragalla, S.; Bhavitha, K.B.; Athanassiou, A. A Review on Graphene Based Materials and Their Antimicrobial Properties. *Coatings* **2021**, *11*, 1197. [CrossRef]
16. Shi, L.; Chen, J.R.; Teng, L.J.; Wang, L.; Zhu, G.L.; Liu, S.; Luo, Z.T.; Shi, X.T.; Wang, Y.J.; Ren, L. The Antibacterial Applications of Graphene and Its Derivatives. *Small* **2016**, *12*, 4165–4184. [CrossRef]
17. 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* **2017**, *74*, 568–581. [CrossRef]
18. Kumar, A.; Sharma, K.; Dixit, A.R. A review of the mechanical and thermal properties of graphene and its hybrid polymer nanocomposites for structural applications. *J. Mater. Sci.* **2019**, *54*, 5992–6026. [CrossRef]
19. Kuziel, A.W.; Milowska, K.Z.; Chau, P.L.; Boncel, S.; Koziol, K.K.; Yahya, N.; Payne, M.C. The True Amphipathic Nature of Graphene Flakes: A Versatile 2D Stabilizer. *Adv. Mater.* **2020**, *32*, 2000608. [CrossRef]
20. Van Thanh, D.; Duc, D.N.; Thi, T.C.; Phuoc, H.L.; Dai, L.T.; Ngoc, M.P.; Van, C.N. Recent trends in preparation and application of carbon nanotube-graphene hybrid thin films. *Adv. Nat. Sci.-Nanosci. Nanotechnol.* **2016**, *7*, 033002. [CrossRef]
21. Zhao, B.; Yang, S.H.; Deng, J.P.; Pan, K. Chiral Graphene Hybrid Materials: Structures, Properties, and Chiral Applications. *Adv. Sci.* **2021**, *8*, 2003681. [CrossRef] [PubMed]
22. Lukowiak, A.; Kedziora, A.; Strek, W. Antimicrobial graphene family materials: Progress, advances, hopes and fears. *Adv. Colloid Interface Sci.* **2016**, *236*, 101–112. [CrossRef] [PubMed]
23. Ji, H.; Sun, H.; Qu, X. Antibacterial applications of graphene-based nanomaterials: Recent achievements and challenges. *Adv. Drug Deliv. Rev.* **2016**, *105*, 176–189. [CrossRef] [PubMed]
24. Priyadarsini, S.; Mohanty, S.; Mukherjee, S.; Basu, S.; Mishra, M. Graphene and graphene oxide as nanomaterials for medicine and biology application. *J. Nanostruct. Chem.* **2018**, *8*, 123–137. [CrossRef]
25. Tashan, H.; Khosravi-Darani, K.; Yazdian, F.; Omidi, M.; Sheikhpour, M.; Farahani, M.; Omri, A. Antibacterial Properties of Graphene Based Nanomaterials: An Emphasis on Molecular Mechanisms, Surface Engineering and Size of Sheets. *Mini-Rev. Org. Chem.* **2019**, *16*, 159–172. [CrossRef]
26. Barbolina, I.; Woods, C.R.; Lozano, N.; Kostarelos, K.; Novoselov, K.S.; Roberts, I.S. Purity of graphene oxide determines its antibacterial activity. *2D Mater.* **2016**, *3*, 025025. [CrossRef]

27. Ma, J.X.; Ping, D.; Dong, X.F. Recent Developments of Graphene Oxide-Based Membranes: A Review. *Membranes* **2017**, *7*, 52. [CrossRef]
28. Musico, Y.L.F.; Santos, C.M.; Dalida, M.L.P.; Rodrigues, D.F. Surface Modification of Membrane Filters Using Graphene and Graphene Oxide-Based Nanomaterials for Bacterial Inactivation and Removal. *ACS Sustain. Chem. Eng.* **2014**, *2*, 1559–1565. [CrossRef]
29. Kanchanapally, R.; Viraka Nellore, B.P.; Sinha, S.S.; Pedraza, F.; Jones, S.J.; Pramanik, A.; Chavva, S.R.; Tchounwou, C.; Shi, Y.; Vangara, A.; et al. Antimicrobial Peptide-Conjugated Graphene Oxide Membrane for Efficient Removal and Effective Killing of Multiple Drug Resistant Bacteria. *RSC Adv.* **2015**, *5*, 18881–18887. [CrossRef]
30. Xie, J.; Ming, Z.; Li, H.; Yang, H.; Yu, B.; Wu, R.; Liu, X.; Bai, Y.; Yang, S.T. Toxicity of graphene oxide to white rot fungus Phanerochaete chrysosporium. *Chemosphere* **2016**, *151*, 324–331. [CrossRef]
31. Yu, X.; Cheng, H.; Zhang, M.; Zhao, Y.; Qu, L.; Shi, G. Graphene-based smart materials. *Nat. Rev. Mater.* **2017**, *2*, 17046. [CrossRef]
32. Ma, Y.; Zhi, L. Graphene-Based Transparent Conductive Films: Material Systems, Preparation and Applications. *Small Methods* **2019**, *3*, 1800199. [CrossRef]
33. Song, N.; Gao, X.; Ma, Z.; Wang, X.; Wei, Y.; Gao, C. A review of graphene-based separation membrane: Materials, characteristics, preparation and applications. *Desalination* **2018**, *437*, 59–72. [CrossRef]
34. Cranford, S.W.; Buehler, M.J. Packing efficiency and accessible surface area of crumpled graphene. *Phys. Rev. B* **2011**, *84*, 205451. [CrossRef]
35. Li, X.; Chen, Y.; Cheng, Z.; Jia, L.; Mo, S.; Liu, Z. Ultrahigh specific surface area of graphene for eliminating subcooling of water. *Appl. Energy* **2014**, *130*, 824–829. [CrossRef]
36. Wang, S.; Zhang, Y.; Abidi, N.; Cabrales, L. Wettability and surface free energy of graphene films. *Langmuir* **2009**, *25*, 11078–11081. [CrossRef]
37. 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. [CrossRef]
38. Papageorgiou, D.G.; Kinloch, I.A.; Young, R.J. Mechanical properties of graphene and graphene-based nanocomposites. *Prog. Mater. Sci.* **2017**, *90*, 75–127. [CrossRef]
39. Sinclair, R.C.; Suter, J.L.; Coveney, P.V. Micromechanical exfoliation of graphene on the atomistic scale. *Phys. Chem. Chem. Phys.* **2019**, *21*, 5716–5722. [CrossRef]
40. Xu, X.; Zhang, Z.; Dong, J.; Yi, D.; Niu, J.; Wu, M.; Lin, L.; Yin, R.; Li, M.; Zhou, J.; et al. Ultrafast epitaxial growth of metre-sized single-crystal graphene on industrial Cu foil. *Sci. Bull.* **2017**, *62*, 1074–1080. [CrossRef]
41. Stankovich, S.; Dikin, D.A.; Piner, R.D.; Kohlhaas, K.A.; Kleinhammes, A.; Jia, Y.; Wu, Y.; Nguyen, S.T.; Ruoff, R.S. Synthesis of graphene-based nanosheets via chemical reduction of exfoliated graphite oxide. *Carbon* **2007**, *45*, 1558–1565. [CrossRef]
42. De Silva, K.K.H.; Huang, H.H.; Joshi, R.K.; Yoshimura, M. Chemical reduction of graphene oxide using green reductants. *Carbon* **2017**, *119*, 190–199. [CrossRef]
43. Guex, L.G.; Sacchi, B.; Peuvot, K.F.; Andersson, R.L.; Pourrahimi, A.M.; Strom, V.; Farris, S.; Olsson, R.T. Experimental review: Chemical reduction of graphene oxide (GO) to reduced graphene oxide (rGO) by aqueous chemistry. *Nanoscale* **2017**, *9*, 9562–9571. [CrossRef] [PubMed]
44. Bhol, P.; Yadav, S.; Altaee, A.; Saxena, M.; Misra, P.K.; Samal, A.K. Graphene-Based Membranes for Water and Wastewater Treatment: A Review. *ACS Appl. Nano Mater.* **2021**, *4*, 3274–3293. [CrossRef]
45. Sun, C.; Liu, M.; Bai, B. Molecular simulations on graphene-based membranes. *Carbon* **2019**, *153*, 481–494. [CrossRef]
46. Ali, A.; Aamir, M.; Thebo, K.H.; Akhtar, J. Laminar Graphene Oxide Membranes Towards Selective Ionic and Molecular Separations: Challenges and Progress. *Chem. Rec.* **2020**, *20*, 344–354. [CrossRef]
47. Liu, L.; Cheng, Y.; Zhang, X.; Shan, Y.; Zhang, X.; Wang, W.; Li, D. Graphene-based transparent conductive films with enhanced transmittance and conductivity by introducing antireflection nanostructure. *Surf. Coat. Technol.* **2017**, *325*, 611–616. [CrossRef]
48. Chen, Y.; Yang, G.Z.; Liu, B.; Kong, H.; Xiong, Z.; Guo, L.; Wei, G. Biomineralization of ZrO_2 nanoparticles on graphene oxide-supported peptide/cellulose binary nanofibrous membranes for high-performance removal of fluoride ions. *Chem. Eng. J.* **2022**, *430*, 132721. [CrossRef]
49. Baskoro, F.; Wong, C.-B.; Kumar, S.R.; Chang, C.-W.; Chen, C.-H.; Chen, D.W.; Lue, S.J. Graphene oxide-cation interaction: Inter-layer spacing and zeta potential changes in response to various salt solutions. *J. Membr. Sci.* **2018**, *554*, 253–263. [CrossRef]
50. Zhang, Y.; Zhang, S.; Gao, J.; Chung, T.-S. Layer-by-layer construction of graphene oxide (GO) framework composite membranes for highly efficient heavy metal removal. *J. Membr. Sci.* **2016**, *515*, 230–237. [CrossRef]
51. Alayande, A.B.; Obaid, M.; Kim, I.S. Antimicrobial mechanism of reduced graphene oxide-copper oxide (rGO-CuO) nanocomposite films: The case of Pseudomonas aeruginosa PAO1. *Mater. Sci. Eng. C* **2020**, *109*, 110596. [CrossRef] [PubMed]
52. Guan, K.; Liu, G.; Matsuyama, H.; Jin, W. Graphene-based membranes for pervaporation processes. *Chin. J. Chem. Eng.* **2020**, *28*, 1755–1766. [CrossRef]
53. Kim, H.W.; Yoon, H.W.; Yoon, S.M.; Yoo, B.M.; Ahn, B.K.; Cho, Y.H.; Shin, H.J.; Yang, H.; Paik, U.; Kwon, S.; et al. Selective gas transport through few-layered graphene and graphene oxide membranes. *Science* **2013**, *342*, 91–95. [CrossRef] [PubMed]
54. Chi, C.; Wang, X.; Peng, Y.; Qian, Y.; Hu, Z.; Dong, J.; Zhao, D. Facile Preparation of Graphene Oxide Membranes for Gas Separation. *Chem. Mater.* **2016**, *28*, 2921–2927. [CrossRef]

55. Cobos, M.; De-La-Pinta, I.; Quindos, G.; Fernandez, M.J.; Fernandez, M.D. Synthesis, Physical, Mechanical and Antibacterial Properties of Nanocomposites Based on Poly(vinyl alcohol)/Graphene Oxide-Silver Nanoparticles. *Polymers* **2020**, *12*, 723. [CrossRef]
56. Akbari, A.; Sheath, P.; Martin, S.T.; Shinde, D.B.; Shaibani, M.; Banerjee, P.C.; Tkacz, R.; Bhattacharyya, D.; Majumder, M. Large-area graphene-based nanofiltration membranes by shear alignment of discotic nematic liquid crystals of graphene oxide. *Nat. Commun.* **2016**, *7*, 10891. [CrossRef]
57. Jia, T.; Shen, S.; Xiao, L.; Jin, J.; Zhao, J.; Che, Q. Constructing multilayered membranes with layer-by-layer self-assembly technique based on graphene oxide for anhydrous proton exchange membranes. *Eur. Polym. J.* **2020**, *122*, 109362. [CrossRef]
58. Shi, Y.; Wan, D.; Huang, J.; Liu, Y.; Li, J. Stable LBL self-assembly coating porous membrane with 3D heterostructure for enhanced water treatment under visible light irradiation. *Chemosphere* **2020**, *252*, 126581. [CrossRef]
59. Wang, L.; Wang, N.; Li, J.; Li, J.; Bian, W.; Ji, S. Layer-by-layer self-assembly of polycation/GO nanofiltration membrane with enhanced stability and fouling resistance. *Sep. Purif. Technol.* **2016**, *160*, 123–131. [CrossRef]
60. Mohan, V.B.; Lau, K.-t.; Hui, D.; Bhattacharyya, D. Graphene-based materials and their composites: A review on production, applications and product limitations. *Compos. Part B* **2018**, *142*, 200–220. [CrossRef]
61. Chen, Y.; Li, J.; Li, T.; Zhang, L.; Meng, F. Recent advances in graphene-based films for electromagnetic interference shielding: Review and future prospects. *Carbon* **2021**, *180*, 163–184. [CrossRef]
62. Han, W.; Wu, Z.; Li, Y.; Wang, Y. Graphene family nanomaterials (GFNs)—promising materials for antimicrobial coating and film: A review. *Chem. Eng. J.* **2019**, *358*, 1022–1037. [CrossRef]
63. Li, J.; Liu, X.; Lu, J.; Wang, Y.; Li, G.; Zhao, F. Anti-bacterial properties of ultrafiltration membrane modified by graphene oxide with nano-silver particles. *J. Colloid Interface Sci.* **2016**, *484*, 107–115. [CrossRef] [PubMed]
64. Li, S.; Gao, B.; Wang, Y.; Jin, B.; Yue, Q.; Wang, Z. Antibacterial thin film nanocomposite reverse osmosis membrane by doping silver phosphate loaded graphene oxide quantum dots in polyamide layer. *Desalination* **2019**, *464*, 94–104. [CrossRef]
65. Zhu, J.; Wang, J.; Uliana, A.A.; Tian, M.; Zhang, Y.; Zhang, Y.; Volodin, A.; Simoens, K.; Yuan, S.; Li, J.; et al. Mussel-Inspired Architecture of High-Flux Loose Nanofiltration Membrane Functionalized with Antibacterial Reduced Graphene Oxide-Copper Nanocomposites. *ACS Appl. Mater. Interfaces* **2017**, *9*, 28990–29001. [CrossRef]
66. Abadikhah, H.; Naderi Kalali, E.; Khodi, S.; Xu, X.; Agathopoulos, S. Multifunctional Thin-Film Nanofiltration Membrane Incorporated with Reduced Graphene Oxide@TiO_2@Ag Nanocomposites for High Desalination Performance, Dye Retention, and Antibacterial Properties. *ACS Appl. Mater. Interfaces* **2019**, *11*, 23535–23545. [CrossRef]
67. Zhao, C.; Lv, J.; Xu, X.; Zhang, G.; Yang, Y.; Yang, F. Highly antifouling and antibacterial performance of poly (vinylidene fluoride) ultrafiltration membranes blending with copper oxide and graphene oxide nanofillers for effective wastewater treatment. *J. Colloid Interface Sci.* **2017**, *505*, 341–351. [CrossRef]
68. Chen, B.; Liu, M.; Zhang, L.; Huang, J.; Yao, J.; Zhang, Z. Polyethylenimine-functionalized graphene oxide as an efficient gene delivery vector. *J. Mater. Chem.* **2011**, *21*, 7736. [CrossRef]
69. Feng, L.; Yang, X.; Shi, X.; Tan, X.; Peng, R.; Wang, J.; Liu, Z. Polyethylene glycol and polyethylenimine dual-functionalized nano-graphene oxide for photothermally enhanced gene delivery. *Small* **2013**, *9*, 1989–1997. [CrossRef]
70. Wen, H.; Dong, C.; Dong, H.; Shen, A.; Xia, W.; Cai, X.; Song, Y.; Li, X.; Li, Y.; Shi, D. Engineered redox-responsive PEG detachment mechanism in PEGylated nano-graphene oxide for intracellular drug delivery. *Small* **2012**, *8*, 760–769. [CrossRef]
71. Zhang, W.; Guo, Z.; Huang, D.; Liu, Z.; Guo, X.; Zhong, H. Synergistic effect of chemo-photothermal therapy using PEGylated graphene oxide. *Biomaterials* **2011**, *32*, 8555–8561. [CrossRef] [PubMed]
72. Lim, M.-Y.; Choi, Y.-S.; Kim, J.; Kim, K.; Shin, H.; Kim, J.-J.; Shin, D.M.; Lee, J.-C. Cross-linked graphene oxide membrane having high ion selectivity and antibacterial activity prepared using tannic acid-functionalized graphene oxide and polyethyleneimine. *J. Membr. Sci.* **2017**, *521*, 1–9. [CrossRef]
73. Zhang, Y.; Chen, S.; An, J.; Fu, H.; Wu, X.; Pang, C.; Gao, H. Construction of an Antibacterial Membrane Based on Dopamine and Polyethylenimine Cross-Linked Graphene Oxide. *ACS Biomater. Sci. Eng.* **2019**, *5*, 2732–2739. [CrossRef] [PubMed]
74. Kaneda, M.; Lu, X.; Cheng, W.; Zhou, X.; Bernstein, R.; Zhang, W.; Kimura, K.; Elimelech, M. Photografting Graphene Oxide to Inert Membrane Materials to Impart Antibacterial Activity. *Environ. Sci. Technol. Lett.* **2019**, *6*, 141–147. [CrossRef]
75. He, C.; Shi, Z.Q.; Cheng, C.; Lu, H.Q.; Zhou, M.; Sun, S.D.; Zhao, C.S. Graphene oxide and sulfonated polyanion co-doped hydrogel films for dual-layered membranes with superior hemocompatibility and antibacterial activity. *Biomater. Sci.* **2016**, *4*, 1431–1440. [CrossRef]
76. Zhang, W.; Cheng, W.; Ziemann, E.; Be'er, A.; Lu, X.; Elimelech, M.; Bernstein, R. Functionalization of ultrafiltration membrane with polyampholyte hydrogel and graphene oxide to achieve dual antifouling and antibacterial properties. *J. Membr. Sci.* **2018**, *565*, 293–302. [CrossRef]
77. Liu, B.; Jiang, M.; Zhu, D.Z.; Zhang, J.M.; Wei, G. Metal-organic frameworks functionalized with nucleic acids and amino acids for structure- and function-specific applications: A tutorial review. *Chem. Eng. J.* **2022**, *428*, 131118. [CrossRef]
78. Grande, C.D.; Mangadlao, J.; Fan, J.; De Leon, A.; Delgado-Ospina, J.; Rojas, J.G.; Rodrigues, D.F.; Advincula, R. Chitosan Cross-Linked Graphene Oxide Nanocomposite Films with Antimicrobial Activity for Application in Food Industry. *Macromol. Symp.* **2017**, *374*, 1600114. [CrossRef]
79. Sanmugam, A.; Vikraman, D.; Park, H.J.; Kim, H.S. One-Pot Facile Methodology to Synthesize Chitosan-ZnO-Graphene Oxide Hybrid Composites for Better Dye Adsorption and Antibacterial Activity. *Nanomaterials* **2017**, *7*, 363. [CrossRef]

80. Sreeprasad, T.S.; Maliyekkal, M.S.; Deepti, K.; Chaudhari, K.; Xavier, P.L.; Pradeep, T. Transparent, luminescent, antibacterial and patternable film forming composites of graphene oxide/reduced graphene oxide. *ACS Appl. Mater. Interfaces* **2011**, *3*, 2643–2654. [CrossRef]
81. Xie, Y.Y.; Hu, X.H.; Zhang, Y.W.; Wahid, F.; Chu, L.Q.; Jia, S.R.; Zhong, C. Development and antibacterial activities of bacterial cellulose/graphene oxide-CuO nanocomposite films. *Carbohydr. Polym.* **2020**, *229*, 115456. [CrossRef] [PubMed]
82. Lin, C.-M.; Chang, Y.-C.; Cheng, L.-C.; Liu, C.-H.; Chang, S.C.; Hsien, T.-Y.; Wang, D.-M.; Hsieh, H.-J. Preparation of graphene-embedded hydroxypropyl cellulose/chitosan/polyethylene oxide nanofiber membranes as wound dressings with enhanced antibacterial properties. *Cellulose* **2020**, *27*, 2651–2667. [CrossRef]
83. Yan, X.; Li, F.; Hu, K.D.; Xue, J.; Pan, X.F.; He, T.; Dong, L.; Wang, X.Y.; Wu, Y.D.; Song, Y.H.; et al. Nacre-mimic Reinforced Ag@reduced Graphene Oxide-Sodium Alginate Composite Film for Wound Healing. *Sci. Rep.* **2017**, *7*, 13851. [CrossRef] [PubMed]
84. Geng, H.; Dai, J.; Li, J.; Di, Z.; Liu, X. Antibacterial ability and hemocompatibility of graphene functionalized germanium. *Sci. Rep.* **2016**, *6*, 37474. [CrossRef] [PubMed]
85. Dhanasekar, M.; Jenefer, V.; Nambiar, R.B.; Babu, S.G.; Selvam, S.P.; Neppolian, B.; Bhat, S.V. Ambient light antimicrobial activity of reduced graphene oxide supported metal doped TiO_2 nanoparticles and their PVA based polymer nanocomposite films. *Mater. Res. Bull.* **2018**, *97*, 238–243. [CrossRef]
86. Zhu, J.; Wang, J.; Hou, J.; Zhang, Y.; Liu, J.; Van der Bruggen, B. Graphene-based antimicrobial polymeric membranes: A review. *J. Mater. Chem. A* **2017**, *5*, 6776–6793. [CrossRef]
87. Pang, J.; Kang, Z.; Wang, R.; Xu, B.; Nie, X.; Fan, L.; Zhang, F.; Du, X.; Feng, S.; Sun, D. Exploring the sandwich antibacterial membranes based on UiO-66/graphene oxide for forward osmosis performance. *Carbon* **2019**, *144*, 321–332. [CrossRef]
88. Zahid, M.; Akram, S.; Rashid, A.; Rehan, Z.A.; Javed, T.; Shabbir, R.; Hessien, M.M.; El-Sayed, M.E. Investigating the Antibacterial Activity of Polymeric Membranes Fabricated with Aminated Graphene Oxide. *Membranes* **2021**, *11*, 510. [CrossRef] [PubMed]
89. Zhang, Y.; Ruan, H.; Guo, C.; Liao, J.; Shen, J.; Gao, C. Thin-film nanocomposite reverse osmosis membranes with enhanced antibacterial resistance by incorporating p-aminophenol-modified graphene oxide. *Sep. Purif. Technol.* **2020**, *234*, 116017. [CrossRef]
90. Li, J.; Wang, G.; Zhu, H.; Zhang, M.; Zheng, X.; Di, Z.; Liu, X.; Wang, X. Antibacterial activity of large-area monolayer graphene film manipulated by charge transfer. *Sci. Rep.* **2014**, *4*, 4359. [CrossRef]
91. Xia, M.Y.; Xie, Y.; Yu, C.H.; Chen, G.Y.; Li, Y.H.; Zhang, T.; Peng, Q. Graphene-based nanomaterials: The promising active agents for antibiotics-independent antibacterial applications. *J. Control. Release* **2019**, *307*, 16–31. [CrossRef] [PubMed]
92. Pham, V.T.H.; Truong, V.K.; Quinn, M.D.J.; Notley, S.M.; Guo, Y.C.; Baulin, V.A.; Al Kobaisi, M.; Crawford, R.J.; Ivanova, E.P. Graphene Induces Formation of Pores That Kill Spherical and Rod-Shaped Bacteria. *Acs Nano* **2015**, *9*, 8458–8467. [CrossRef] [PubMed]
93. Akhavan, O.; Ghaderi, E. Toxicity of Graphene and Graphene Oxide Nanowalls Against Bacteria. *Acs Nano* **2010**, *4*, 5731–5736. [CrossRef] [PubMed]
94. Marković, Z.M.; Jovanović, S.P.; Mašković, P.Z.; Danko, M.; Mičušík, M.; Pavlović, V.B.; Milivojević, D.D.; Kleinová, A.; Špitalský, Z.; Todorović Marković, B.M. Photo-induced antibacterial activity of four graphene based nanomaterials on a wide range of bacteria. *RSC Adv.* **2018**, *8*, 31337–31347. [CrossRef]
95. Gurunathan, S.; Han, J.W.; Dayem, A.A.; Eppakayala, V.; Kim, J.H. Oxidative stress-mediated antibacterial activity of graphene oxide and reduced graphene oxide in Pseudomonas aeruginosa. *Int. J. Nanomed.* **2012**, *7*, 5901–5914. [CrossRef]
96. Liu, S.B.; Zeng, T.H.; Hofmann, M.; Burcombe, E.; Wei, J.; Jiang, R.R.; Kong, J.; Chen, Y. Antibacterial Activity of Graphite, Graphite Oxide, Graphene Oxide, and Reduced Graphene Oxide: Membrane and Oxidative Stress. *Acs Nano* **2011**, *5*, 6971–6980. [CrossRef]
97. Perreault, F.; de Faria, A.F.; Nejati, S.; Elimelech, M. Antimicrobial Properties of Graphene Oxide Nanosheets: Why Size Matters. *Acs Nano* **2015**, *9*, 7226–7236. [CrossRef]
98. Chen, J.N.; Peng, H.; Wang, X.P.; Shao, F.; Yuan, Z.D.; Han, H.Y. Graphene oxide exhibits broad-spectrum antimicrobial activity against bacterial phytopathogens and fungal conidia by intertwining and membrane perturbation. *Nanoscale* **2014**, *6*, 1879–1889. [CrossRef]
99. Mellata, M. Human and avian extraintestinal pathogenic *Escherichia coli*: Infections, zoonotic risks, and antibiotic resistance trends. *Foodbrone Pathog. Dis.* **2013**, *10*, 916–932. [CrossRef]
100. Costalonga, M.; Herzberg, M.C. The oral microbiome and the immunobiology of periodontal disease and caries. *Immunol. Lett.* **2014**, *162*, 22–38. [CrossRef]
101. Krzysciak, W.; Jurczak, A.; Koscielniak, D.; Bystrowska, B.; Skalniak, A. The virulence of Streptococcus mutans and the ability to form biofilms. *Eur. J. Clin. Microbiol. Infect. Dis.* **2014**, *33*, 499–515. [CrossRef] [PubMed]
102. Hebecker, B.; Naglik, J.R.; Hube, B.; Jacobsen, I.D. Pathogenicity mechanisms and host response during oral Candida albicans infections. *Expert Rev. Anti-Infect.* **2014**, *12*, 867–879. [CrossRef] [PubMed]
103. De Bentzmann, S.; Plesiat, P. The Pseudomonas aeruginosa opportunistic pathogen and human infections. *Environ. Microbiol.* **2011**, *13*, 1655–1665. [CrossRef] [PubMed]
104. Murphy, C.N.; Clegg, S. Klebsiella pneumoniae and type 3 fimbriae: Nosocomial infection, regulation and biofilm formation. *Future Microbiol.* **2012**, *7*, 991–1002. [CrossRef]

105. Gart, E.V.; Suchodolski, J.S.; Welsh, T.H., Jr.; Alaniz, R.C.; Randel, R.D.; Lawhon, S.D. Salmonella Typhimurium and Multidirectional Communication in the Gut. *Front. Microbiol.* **2016**, *7*, 1827. [CrossRef]
106. Song, B.; Zhang, C.; Zeng, G.; Gong, J.; Chang, Y.; Jiang, Y. Antibacterial properties and mechanism of graphene oxide-silver nanocomposites as bactericidal agents for water disinfection. *Arch. Biochem. Biophys.* **2016**, *604*, 167–176. [CrossRef]
107. Karahan, H.E.; Wei, L.; Goh, K.; Liu, Z.; Birer, O.; Dehghani, F.; Xu, C.; Wei, J.; Chen, Y. Bacterial physiology is a key modulator of the antibacterial activity of graphene oxide. *Nanoscale* **2016**, *8*, 17181–17189. [CrossRef]
108. Bregnocchi, A.; Zanni, E.; Uccelletti, D.; Marra, F.; Cavallini, D.; De Angelis, F.; De Bellis, G.; Bossu, M.; Ierardo, G.; Polimeni, A.; et al. Graphene-based dental adhesive with anti-biofilm activity. *J. Nanobiotechnol.* **2017**, *15*, 89. [CrossRef]
109. Huang, X.; Marsh, K.L.; McVerry, B.T.; Hoek, E.M.; Kaner, R.B. Low-Fouling Antibacterial Reverse Osmosis Membranes via Surface Grafting of Graphene Oxide. *ACS Appl. Mater. Interfaces* **2016**, *8*, 14334–14338. [CrossRef]
110. Zhang, W.; Yang, Y.; Ziemann, E.; Be'er, A.; Bashouti, M.Y.; Elimelech, M.; Bernstein, R. One-step sonochemical synthesis of a reduced graphene oxide–ZnO nanocomposite with antibacterial and antibiofouling properties. *Environ. Sci. Nano* **2019**, *6*, 3080–3090. [CrossRef]
111. Liu, H.; Liu, X.; Zhao, F.; Liu, Y.; Liu, L.; Wang, L.; Geng, C.; Huang, P. Preparation of a hydrophilic and antibacterial dual function ultrafiltration membrane with quaternized graphene oxide as a modifier. *J. Colloid Interface Sci.* **2020**, *562*, 182–192. [CrossRef] [PubMed]
112. Najjar, A.; Sabri, S.; Al-Gaashani, R.; Kochkodan, V.; Atieh, M. Enhanced Fouling Resistance and Antibacterial Properties of Novel Graphene Oxide-Arabic Gum Polyethersulfone Membranes. *Appl. Sci.* **2019**, *9*, 513. [CrossRef]
113. Zhang, G.; Zhou, M.; Xu, Z.; Jiang, C.; Shen, C.; Meng, Q. Guanidyl-functionalized graphene/polysulfone mixed matrix ultrafiltration membrane with superior permselective, antifouling and antibacterial properties for water treatment. *J. Colloid Interface Sci.* **2019**, *540*, 295–305. [CrossRef]
114. Yu, L.; Zhou, W.; Li, Y.; Zhou, Q.; Xu, H.; Gao, B.; Wang, Z. Antibacterial Thin-Film Nanocomposite Membranes Incorporated with Graphene Oxide Quantum Dot-Mediated Silver Nanoparticles for Reverse Osmosis Application. *ACS Sustain. Chem. Eng.* **2019**, *7*, 8724–8734. [CrossRef]
115. Goswami, M.; Yadav, A.K.; Chauhan, V.; Singh, N.; Kumar, S.; Das, A.; Yadav, V.; Mandal, A.; Tiwari, J.K.; Siddiqui, H.; et al. Facile development of graphene-based air filters mounted on a 3D printed mask for COVID-19. *J. Sci. Adv. Mater. Dev.* **2021**, *6*, 407–414. [CrossRef]
116. Valentini, F.; Cirone, M.; Relucenti, M.; Santarelli, R.; Gaeta, A.; Mussi, V.; De Simone, S.; Zicari, A.; Mardente, S. Antiviral Filtering Capacity of GO-Coated Textiles. *Appl. Sci.* **2021**, *11*, 7501. [CrossRef]
117. Gupta, A.; Sharma, C.P.; Thamaraiselvan, C.; Pisharody, L.; Powell, C.D.; Arnusch, C.J. Low-Voltage Bacterial and Viral Killing Using Laser-Induced Graphene-Coated Non-woven Air Filters. *ACS Appl. Mater. Interfaces* **2021**, *13*, 59373–59380. [CrossRef] [PubMed]
118. Das Jana, I.; Kumbhakar, P.; Banerjee, S.; Gowda, C.C.; Kedia, N.; Kuila, S.K.; Banerjee, S.; Das, N.C.; Das, A.K.; Manna, I.; et al. Copper Nanoparticle–Graphene Composite-Based Transparent Surface Coating with Antiviral Activity against Influenza Virus. *ACS Appl. Nano Mater.* **2020**, *4*, 352–362. [CrossRef]
119. Montes, S.; Etxeberria, A.; Mocholi, V.; Rekondo, A.; Grande, H.; Labidi, J. Effect of combining cellulose nanocrystals and graphene nanoplatelets on the properties of poly(lactic acid) based films. *Express Polym. Lett.* **2018**, *12*, 543–555. [CrossRef]
120. Huang, J.; Jacobsen, J.; Larsen, S.W.; Genina, N.; van de Weert, M.; Mullertz, A.; Nielsen, H.M.; Mu, H. Graphene oxide as a functional excipient in buccal films for delivery of clotrimazole: Effect of molecular interactions on drug release and antifungal activity in vitro. *Int. J. Pharm.* **2020**, *589*, 119811. [CrossRef]
121. Gontarek-Castro, E.; Rybarczyk, M.K.; Castro-Muñoz, R.; Morales-Jiménez, M.; Barragán-Huerta, B.; Lieder, M. Characterization of PVDF/Graphene Nanocomposite Membranes for Water Desalination with Enhanced Antifungal Activity. *Water* **2021**, *13*, 1279. [CrossRef]
122. Chen, J.; Sun, L.; Cheng, Y.; Lu, Z.; Shao, K.; Li, T.; Hu, C.; Han, H. Graphene Oxide-Silver Nanocomposite: Novel Agricultural Antifungal Agent against Fusarium graminearum for Crop Disease Prevention. *ACS Appl. Mater. Interfaces* **2016**, *8*, 24057–24070. [CrossRef] [PubMed]