Abstract: Unlike traditional small molecule drugs, fullerene is an all-carbon nanomolecule with a spherical cage structure. Fullerene exhibits high levels of antiviral activity, inhibiting virus replication in vitro and in vivo. In this review, we systematically summarize the latest research regarding the different types of fullerenes investigated in antiviral studies. We discuss the unique structural advantage of fullerenes, present diverse modification strategies based on the addition of various functional groups, assess the effect of structural differences on antiviral activity, and describe the possible antiviral mechanism. Finally, we discuss the prospective development of fullerenes as antiviral drugs.

Keywords: fullerene; water-soluble fullerene derivatives; antivirus; nanodrug

1. Introduction

Fullerenes are all-carbon molecules discovered in 1985 [1]. They are spherical or ellipsoidal in shape, with a hollow cage structure. Three discoverers of fullerene C$_{60}$ won the Nobel Prize in chemistry in 1996. Fullerene C$_{60}$, the representative fullerene, is ~0.7 nm in diameter. In the past 30 years, with the continuous development of fullerene preparation technology [2–5], fullerenes have presented unprecedented opportunities in the fields of biomedicine, catalysis, superconduction, and photovoltaics. Nanomolecules have important applications in cancer treatment, diagnosis, imaging, drug delivery, catalysis, and biosensing [6–15]. Fullerene molecules not only have defined nanostructures, but also unique electronic characteristics, photophysical properties, and excellent bio-compatibility. Fullerene molecules have properties that differ from those of traditional small molecule drugs, which make fullerenes nanodrug candidates [16], especially for diagnosis and treatment. For example, fullerenes and their derivatives can be used as antioxidants against inflammatory diseases, due to their rich conjugated double bonds, which scavenge free radicals [17,18]. Fullerene C$_{60}$ activates tumor immunity by polarizing tumor-associated macrophages and combines with immune checkpoint inhibitors (PD-L1 monoclonal antibody) to achieve efficient tumor immunotherapy [19]. Fullerene C$_{70}$ derivatives, as photosensitizers, produce singlet oxygen, which can effectively kill tumor cells [20]. Endohedral metal fullerenes serve as new nuclear magnetic resonance contrast agents [21,22] for treating liver steatosis [23] and tumors [24–30]. Additionally, some fullerene derivatives stabilize immune effector cells and prevent/inhibit the release of proinflammatory mediators. Therefore, they are potential drugs for a variety of diseases, such as asthma [31], arthritis [32], and multiple sclerosis [33]. Moreover, carboxylic acid derivatives of fullerenes can cut DNA under visible light irradiation, with the potential...
for use as photosensitive biochemical probes [34]. Fullerene C$_{60}$ carboxylic acid derivatives also exhibit neuroprotective activity [35] and strongly inhibit tumor growth in a zebrafish xenograft model [36]. Because nanoparticles have been approved as drugs and drug carriers, fullerenes have great potential as drugs or gene delivery carriers [37,38].

Currently, more than 90% of infectious diseases in humans are caused by viruses. The most well-known include the influenza virus, human immunodeficiency virus (HIV), and Ebola virus, which have caused serious damage [39–43]. Although several anti-HIV and anti-Ebola drugs, such as saquinavir, ritonavir, T20, lopinavir, ribavirin, tenofovir, and remdesivir, have been developed, their efficacy is not satisfactory. Severe acute respiratory syndrome (SARS), which broke out in China in 2003, is a respiratory infection caused by a coronavirus. So far, there is no specific medicine for SARS. The novel coronavirus, SARS-CoV-2, now circulating worldwide, is more infectious than SARS or HIV. For patients infected with SARS-CoV-2, there are no specific antiviral drugs.

Fullerenes and their derivatives exert significant inhibitory effects against HIV [39], herpes simplex virus (HSV) [40], influenza virus [44], Ebola virus [45], cytomegalovirus (CMV) [46], and other viruses in vitro and in vivo (Figure 1). Fullerenes and their derivatives, as a class of new, broad-spectrum antiviral drugs, have attracted increasing attention as a potential treatment for SARS-CoV-2.

![Possible interaction between fullerene molecules and coronavirus](image-url)

**Figure 1.** Possible interaction between fullerene molecules and coronavirus, in which fullerene molecules inhibit virus replication.

Because fullerenes are insoluble in water and polar media, their use in biomedicine is extremely complicated [47]. To increase biocompatibility, the cage structure of fullerenes needs to be modified with appropriate hydrophilic functional groups. The modified structure and properties of the carbon cage may facilitate new applications in different biological systems. Because the fullerene carbon cage has multiple modifiable reaction sites, many fullerene derivatives with well-defined structures have been synthesized using regioselective functional group derivatization strategies. Therefore, fullerenes serve as ideal scaffolds for different bioactive drugs. Studies on the synthesis and antiviral activity of fullerenes and their derivatives have facilitated a deeper understanding of the relationship between fullerene structure and bioactivity.

In the past, hundreds of fullerene derivatives have been synthesized and used to inhibit viruses in vitro. Most of these derivatives are water soluble. These fullerene derivatives can be classified as the following six types: (1) amino acid, peptide, and primary amine derivatives; (2) piperazine and pyrrolidine derivatives; (3) carboxyl derivatives; (4) hydroxyl derivatives; (5) glycofullerene derivatives; and (6) fullerene complexes. Numerous antiviral studies have been conducted to evaluate fullerene C$_{60}$ and its derivatives; this review assesses the latest research on the ability of fullerene C$_{60}$ and its derivatives to inhibit virus replication.
2. Synthesis and Antiviral Properties of Amino Acid, Peptide, and Primary Amine Fullerene Derivatives

HIV-1-specific protease (HIVP) is an effective target for antiviral therapy. Its active site can be roughly described as an open-ended cylindrical cavity composed almost entirely of hydrophobic amino acids [48]. Since spherical C_60 derivatives are hydrophobic and have a similar radius as the cylindrical cavity, strong hydrophobic interactions may occur between the active site surface and C_60 derivatives. Therefore, C_60 derivatives are potential inhibitors of HIVP.

In 1993, Friedman et al. [39] reported a landmark study based on model building and experimental evidence. Theoretical calculations revealed that the hydrophobic cavity of HIVP accommodates C_60 molecules; the spherical C_60 molecules fit perfectly within the active site, facilitating strong interactions between HIVP and fullerene. However, because C_60 spheres are insoluble in polar solvents, it is critical to dissolve C_60 in a medium suitable for biological testing. By modifying fullerene with strong polar groups, Sijbesma et al. [49] synthesized the water-soluble fullerene derivative bis (phenylenaminosuccinic acid)-C_60 (1) (Figure 2). In vitro studies revealed that 1 inhibited acute and chronic HIV-1 infection in human peripheral blood mononuclear (PBM) cells, with a half-effective concentration (EC_{50}) of 7.0 μM, while showing no cytotoxicity to uninfected PBM cells. This work is of great significance because fullerene derivatives as virus inhibitors are unprecedented in the field of antiviral research.

According to the theoretical analysis, the main driving force behind the binding between HIVP and fullerene derivatives is the hydrophobic interaction between the non-polar active site surface of the protease and the non-polar fullerene surface [50,51]. In order to improve antiviral activity, Friedman et al. proposed the addition of appropriate functional groups at specific positions on the fullerene derivatives [39] that would interact with the protease, generate electrostatic and/or hydrogen bonds as well as van der Waals forces, and subsequently increase the binding constant by several orders of magnitude. Therefore, compound 2 with two amino groups (Figure 3) was designed as an ideal “second generation” fullerene derivative. In addition to forming van der Waals forces with the non-polar HIVP surface, the cationic sites on the fullerene surface can form salt bridges with the catalytic aspartates on the floor of the active site, thus improving the binding strength. Although compound 2 was an ideal model of a fullerene derivative, the pure isomer was difficult to obtain, owing to a lack of regioselectivity. Subsequently, Prato and co-workers [52] proposed the design, simulation, and synthesis of the C_60 diamine derivative 3, which was similar to 2. In the PM3-minimized structure of 3, the N–N distance between the two amino groups is 0.51 nm, while that in 2 is 0.55 nm, suggesting that the spatial arrangement of the two amino groups in 3 is very similar to that in 2.
In order to determine the binding energy between 3 and HIVP, Prato and co-workers [52] conducted a simulation using the Discover (Biosym/MSI) program with a cvff force field. Figure 4a depicts 3 binding to the cavity region of HIVP to form a complex. Compared with unmodified fullerene C\textsubscript{60}, the complex formed by the binding of 3 to HIVP exhibited significant improvements. When fullerene was protonated with a monoamino group, the relative binding energy was approximately $-134$ kJ/mol, and when it was protonated with a diamino group, the relative binding energy was approximately $-105$ kJ/mol. The greater binding energy of compound 3, with a single amino group, is due to the hydrogen bond interactions between the neutral $\text{–NH}_2$ and $\text{–COOH}$ groups of neutral aspartic acid, rather than the hydrogen bond interactions between the $\text{–NH}_3^+$ group and $\text{–COOH}$. Figure 4b illustrates the active site of the complex, highlighting the hydrogen bonds between the HIVP cavity and 3. The interatomic distance between the N atom in each amine/ammonium molecule and the O atom in the carboxyl/carboxylic acid is about 0.28 nm. This strong interaction suggests the contribution of Coulombic attraction.

In the same time period, many research groups synthesized various fullerene derivatives with different functional groups and demonstrated effective virus inhibition by introducing appropriate carboxylic acid and amino acid groups at specific positions on fullerenes. Among them, the fullerene dendritic amino acid derivative 4, prepared by Breetreich and Hirsch [53] (Figure 5), is highly water soluble and has an EC\textsubscript{50} of 0.22 µM in human lymphocytes acutely infected with HIV-1 [54]. This compound also shows no
toxicity up to 100 µM in PBM, Vero, and CEM cells. The fullerene amino acid derivative 5 (Figure 5), synthesized by Mashino et al. [40], strongly inhibits HIV reverse transcriptase (HIV-RT), with an EC\textsubscript{50} of 0.029 µM. Additionally, Toniolo et al. [55] synthesized fullerene peptide derivatives, which also exhibit anti-HIWP activity.

In 2012, Troshin and coworkers [43] reported that multi-functional C\textsubscript{60} amine and amino acid derivatives could be readily prepared from hexachlorofullerene C\textsubscript{60}Cl\textsubscript{6} (6). The synthesized fullerene derivative contains at least five hydrophilic functional groups (such as amino or carboxyl groups). As shown in Figure 6, a fullerene amino acid ester (8) can be obtained via the reaction of C\textsubscript{60}Cl\textsubscript{6} with an amino acid ester (7). The amino acid ester groups in compound 8 can be hydrolyzed to obtain fullerene amino acid derivatives under acidic conditions. In order to further increase the water solubility of fullerene amino acid derivatives, fullerene amino acid potassium salts are formed by adding potassium carbonate.

In vitro cell experiments [43] showed that the carboxylic acid potassium salt 8 has low cytotoxicity to HSV-sensitive Vero cells (CC\textsubscript{50} > 1.3 mM) and human CMV-sensitive human embryonic lung fibroblasts (CC\textsubscript{50} > 0.5 mM). Meanwhile, compound 8 showed pronounced antiviral activity, with an EC\textsubscript{50} of 0.26 µM for HSV and 37.6 µM for CMV. Combining CC\textsubscript{50} and EC\textsubscript{50} values, the selectivity indices of compound 8 for HSV and CMV are >5000 and 14, respectively, indicating that compound 8 has potential as a new antiviral drug against HSV and CMV.

3. Synthesis and Antiviral Studies of Fullerene Piperazine and Pyrrolidine Derivatives

As shown in Figure 7, the reaction of C\textsubscript{60}Cl\textsubscript{6} with N-methylpiperazine (9) can efficiently generate a fullerene-piperazine derivative (10) in the absence of any base [43]. By adding
According to the literature [51], a regioisomer mixture of C_{60} pyrrolidine derivatives exhibits good anti-HIV-1 activity. The pyrrolidine derivative has a low EC_{50} and exhibits little toxicity to Vero and PBM cells. However, a relationship between the structure and activity of the reported isomer mixture of fullerene pyrrolidine derivatives has not been established. In order to better understand which structural characteristics can be modified to improve anti-HIV activity, Prato and coworkers [56] synthesized a series of new, pure fullerene pyrrolidine derivatives through the Prato reaction (Figure 8).

![Figure 7. Synthesis route for fullerene penta-N-methyl piperazine salt (10).](image)

![Figure 8. Structure diagram of fullerene pyrrolidine derivatives (11–23).](image)

six times the amount of compound 9 to a C_{60}Cl_{6} toluene solution, 10 is precipitated immediately, with more than a 95% yield. In vivo cell experiments have indicated that 10 exhibits high acute toxicity when administered via intraperitoneal injection in mice, while 8 shows very low acute toxicity, suggesting the latter is safe for biomedical applications. This example also revealed that the toxicity of the water-soluble fullerene derivatives depends largely on the organic functional groups attached to the fullerene carbon cage.
The Bis-adduct fullerene pyrrolidine derivative has eight regioisomers. The yields of the eight isomers differ; cis-Bis-adducts are produced at extremely low yields and trans- and equatorial-adducts are produced at relatively high yields. Hence, Prato and coworkers were able to separate trans- and equatorial-isomers (12–15 and 16–19, respectively) and study their structure and activity. Additionally, a Mono-adduct (11) and other Bis-adducts (20–21, 22–23) were synthesized (Figure 8).

The anti-HIV activity and cytotoxicity of all fullerene pyrrolidine derivatives (11–23) was tested in lymphocyte (CEM) cultures infected with HIV-1 or HIV-2. As shown in Table 1, mono-functionalized derivative 11 and Bis-adduct derivatives 12–15, 20, 21 and 23 showed low anti-HIV-1 activity, while the corresponding quaternary ammonium pyrrolidine derivatives 16–19 showed high anti-HIV-1 activity at low concentrations (EC\textsubscript{50}: 0.40–2.60 µM). These results suggest that the inhibitory effect on HIV-1 might be related to electrostatic interactions.

Table 1. Anti-HIV activity and cytostatic toxicity of compounds (11–23) in CEM cell cultures.

| Compound | EC\textsubscript{50} (µM) | HIV-1 | HIV-2 | CC\textsubscript{50} (µM) |
|----------|-------------------|-------|-------|-----------------|
| 11       | >4                | >4    | >4    | 44.3            |
| 12 (trans-2) | >4                | >4    | >4    | 7.2             |
| 13 (trans-3) | >4                | >4    | >4    | 7.63            |
| 14 (trans-4) | >4                | >4    | >4    | 7.4             |
| 15 (equatorial) | >4            | >4    | >4    | 9.6             |
| 16 (trans-2) | 0.40 ± 0.0       | >4    | >4    | 4.79            |
| 17 (trans-3) | 0.96 ± 0.39      | >4    | >4    | 3.02            |
| 18 (trans-4) | 2.60 ± 0.88      | >4    | >4    | 13.2            |
| 19 (equatorial) | 1.60 ± 0.0      | >4    | >4    | 6.59            |
| 20 (trans-2) | >4                | >4    | >4    | -               |
| 21 (equatorial) | >4            | >4    | >4    | -               |
| 22 (trans-2) | 2.01 ± 0.0       | >4    | >4    | -               |
| 23 (equatorial) | >4            | >4    | >4    | -               |

The biological activity of pyrrolidine derivatives of fullerene varies among the regioisomers. As shown in Table 1, the anti-HIV-1 activity of the trans-2 isomer (16) was 2.4–6.5 times that of the corresponding trans-3 (17), trans-4 (18), and equatorial (19) isomers. The trans-2-tetraacetic acid derivative (22) showed high anti-HIV-1 activity at low concentrations, while the equatorial isomer (23) showed low activity. The introduction of malonate resulted in the loss of anti-HIV-1 activity of pyrrolidine derivatives 20–21. Most synthetic fullerene pyrrolidine derivatives exhibit pronounced toxicity in human CEM cells (CC\textsubscript{50}: 3.02–13.2 µM), but monofunctional derivative 11 exhibit relatively low toxicity (CC\textsubscript{50}: 44 µM). This toxicity most likely results from the strong amphiphilic properties of these pyrrolidine derivatives, which can cause the rupture of the cell membrane and subsequent cell death.

To define the structure-activity relationship, Prato and co-workers [57] prepared Bis-adduct fullerene pyrrolidine derivatives (24–28) through a [3+2] dipolar cycloaddition reaction between azomethine ylides and C\textsubscript{60} (Figure 9) and subsequently studied their anti-HIV-1 and anti-HIV-2 activity and cytotoxicity in CEM culture. As shown in Table 2, the anti-HIV-1 activity of the trans-isomers (24–26) is about 2–10 times that of the corresponding cis-3 isomer (28), while the equatorial-isomer (27) exhibits no antiviral activity against HIV-1. Among the trans-isomers, trans-2 (24) and trans-3 (25) demonstrate higher levels of anti-HIV-1 (EC\textsubscript{50} = 0.21 and 0.35 µM, respectively) and anti-HIV-2 activity (EC\textsubscript{50} = 0.21–1.0 µM). The anti-HIV-1 activity of the trans-4 isomer (26) is significantly lower than that of compounds 24 and 25 (EC\textsubscript{50} = 1.08 µM), and the anti-HIV-2 activity is also lower (EC\textsubscript{50} = 2.5 µM), which is consistent with the experimental results for fullerene pyrrolidine derivatives 11–23. Fullerene pyrrolidine derivatives 24–28 also exhibit toxicity in human CEM cells.
(CC₅₀ = 2.9–28.7 µM). The CC₅₀/EC₅₀ ratio of the trans-3 isomer (25) is 26, higher than that of the reference compound 16 (CC₅₀/EC₅₀ = 12).

![Figure 9. Structure diagram of fullerene pyrrolidine derivatives (24–28).](image)

Table 2. Anti-HIV activity and cytostatic activity of compounds (24–28) in CEM cell cultures.

| Compound | EC₅₀ (µM) | CC₅₀ (µM) |
|----------|-----------|-----------|
|          | HIV-1     | HIV-2     |          |
| 24       | 0.21 (±0.07) | 0.2 to 1.0 | 2.93 (±1.20) |
| 25       | 0.35 (±0.07) | 0.70 (±0.42) | 9.04 (±0.18) |
| 26       | 1.08 (±0.57) | 2.50 (±1.90) | 12.5 (±7.54) |
| 27       | >25       | >10       | >125      |
| 28       | 2.50 (±0.71) | >10       | 28.7 (±1.27) |

As mentioned, Mashino et al. (2005) reported that fullerene amino acid-type derivatives exhibited HIV-RT inhibitory activity, while cationic fullerene derivatives such as the pyrrolidinium-type derivatives showed weaker HIV-RT inhibitory activity [40]. The carboxyl groups on the pyrrolidine-type fullerene derivatives were considered crucial to HIV-RT inhibitory activity [40,58]. However, ten years later, Mashino et al. found that fullerene pyrrolidine-pyridine and pyrrolidine-pyridinium salt derivatives without any carboxyl groups [42], such as 29–40, which are functionalized with pyridine or pyridinium groups (Figure 10), exhibit strong HIV-RT inhibition. This is useful information for the future design of fullerene derivatives as HIV-RT inhibitors. In addition to HIV-RT inhibition, recently, Kobayashi et al. [59] found that compound 34 can inhibit HIV-PR, and HCV NS5B polymerase (HCV NS5B) with IC₅₀ values in the micromolar range. Compound 41, the exo-substituent on the most potent derivative (34), exhibits no HIV-RT inhibitory activity in cell culture, indicating that HIV-RT inhibition is dependent on the fullerene skeleton. The conventional trypan blue dye exclusion test (Table 3) was used to evaluate the cytotoxicity of all fullerene pyrrolidine-pyridine and pyrrolidine-pyridinium salt derivatives (29–40) to HL60 cells. The CC₅₀ of all derivatives except 35 (CC₅₀ = 39.4 µM) was greater than 50 µM,
which suggests that the new fullerene-pyrrolidine-pyridine or pyridinium salt derivatives effectively inhibit HIV-RT activity without damaging living cells.

![Structure diagram of fullerene pyrrolidine derivatives (29–41).](image)

**Figure 10.** Structure diagram of fullerene pyrrolidine derivatives (29–41).

| Compound | R¹ | R² | R³ | IC₅₀ * | CC₅₀ * |
|----------|----|----|----|--------|--------|
| 29       | 2-(N-Methylpyridinium) | CH₃COOCH₃ | -C₂H₅ | 0.30    | >50    |
| 30       | 2-(N-Methylpyridinium) | -H         | -C₂H₅ | 0.33    | >50    |
| 31       | 2-Pyridine | -H         | -H    | 0.20    | >50    |
| 32       | 2-Pyridine | -H         | -H    | 0.46    | >50    |
| 33       | 3-(N-Methylpyridinium) | CH₃COOCH₃ | -C₂H₅ | 0.25    | >50    |
| 34       | 3-(N-Methylpyridinium) | -H         | -C₂H₅ | 0.094   | >50    |
| 35       | 3-Pyridine | -H         | -H    | 0.41    | >50    |
| 36       | 3-Pyridine | -H         | -H    | 0.080   | >50    |
| 37       | 4-(N-Methylpyridinium) | CH₃COOCH₃ | -C₂H₅ | 0.74    | >50    |
| 38       | 4-(N-Methylpyridinium) | -H         | -C₂H₅ | 0.37    | >50    |
| 39       | 4-Pyridine | -H         | -H    | 1.60    | >50    |
| 40       | 4-Pyridine | -H         | -H    | 0.80    | >50    |
| 41       | -         | -         | -     | >500    | >50    |
| Nevirapine | -  | -         | -     | 3.52    |        |

* These values (µM) are based on the average of three test results for each test compound.

In 2016, Echegoyen and co-workers [41] reported a novel cationic N,N-dimethyl C₇₀ fullerene-pyrrolidine iodized salt derivative (42–44), with fullerene C₇₀ as the starting material (Figure 11), that inhibits more than 99% of HIV-1 infectivity at a low micromolar concentration. These three compounds have an EC₅₀ of 0.41, 0.33, and 0.54 µM, respectively. An analysis of the life cycle of HIV-1 suggested that these compounds inhibit viral maturation by influencing the processing of Gag and GAG-POL. Significantly, fullerene derivatives 42–44 do not inhibit protease activity in vitro, and strongly interact with immature HIV capsid proteins in a pull-down assay. Moreover, these compounds may block infection by viruses carrying either a mutant protease that is resistant to multiple protease inhibitors, or the mutant Gag protein, which is resistant to the mature inhibitor bevirimat. Fullerenes 42–44 do not inhibit HIV-1 proteases at doses that strongly inhibit HIV-1 infection in vitro, suggesting that this mechanism is independent of the HIV-1 protease. This finding differed from previous reports that fullerene derivatives affect HIV-1 protease activity in vitro. Echegoyen et al. [60] then proposed that fullerene derivatives 42–44 act through a novel anti-HIV-1 mechanism, rather than interacting with other capsid proteins as previously
reported. Unraveling the details of this mechanism will facilitate the discovery of novel anti-HIV-1 inhibitors.

Additionally, Tolla et al. [44] reported that fullerene pyrrolidine derivatives functionalized with hydrophilic sugar groups. While these derivatives have no inhibitory effect on influenza virus hemagglutinin, they exhibit a good inhibitory effect on neuraminidase.

4. Synthesis and Antiviral Studies of Fullerene Carboxyl Derivatives

As mentioned, hydrophilic functional groups such as amino or amino acid groups can be directly or indirectly attached to the skeleton of fullerenes to increase water solubility. These water-soluble fullerene derivatives have exhibited high levels of antiviral activity. Instead of amino or amino acid groups, a single carboxylic acid group can be used to modify fullerenes to increase their water solubility and improve their antiviral activity. In 1996, Schuster et al. [51] synthesized 11 new water-soluble fullerene derivatives, among which fullerene carboxylic acid derivatives 45 and 46 (Figure 12) exhibit antiviral activity against HIV-1 at low micromolar concentrations, with an IC$_{50}$ of 2.2 and 6.3 µM, respectively.

In 2007, Troshin and coworkers [61] proposed an effective method for the synthesis of water-soluble fullerene carboxylic acid derivatives. With C$_{60}$Cl$_6$ as the starting point, C$_{60}$(Ar)$_5$Cl, with ester groups linked to aryl groups, was obtained via the simple and efficient Friedel–Crafts arylation of C$_{60}$Cl$_6$ (6) with methyl esters of phenylacetate at 100 °C. The fullerene carboxylic acid derivative (48) was prepared in almost a quantitative yield by removing the methyl group from the methyl ester under acidic conditions, as shown in Figure 13. Compound 48, with five carboxyl groups, is insoluble in water but soluble in DMSO. In order to improve the water solubility, potassium carbonate was added to neutralize the carboxylic acid group of 48 and form the corresponding ionic potassium salts, with a water solubility of up to 50–100 mg/mL at pH < 7.5. A virus-induced cytopathicity assay revealed that the fullerene carboxylic acid potassium derivative has pronounced anti-HIV-1 activity, with an IC$_{50}$ of 1.20 ± 0.44 µM and a low cytotoxicity (>52 µM).
Nanomaterials 2022, 12, 2547

Figure 13. Schematic diagram of synthetic route of carboxylic fullerene derivative (48).

Usually, only five chlorine atoms in 6 are replaced; the sixth chlorine atom cannot be substituted due to steric hindrance. Recently, Troshin and coworkers [62] found that the sixth chlorine atom can be substituted by an alkyl group through a reverse Arbuzov reaction between trialkyl phosphite (P(OR)3) and the fullerene derivative C_{60}(Ar)_{5}Cl (Figure 14). More significantly, the introduction of different R groups through the reverse Arbuzov reaction affects the antiviral activity of the carboxyl fullerene derivatives, establishing a fundamental correlation between the structure of carboxyl fullerene derivatives and their antiviral activity. Experiments on the inhibition of the influenza H3N2 virus showed that compounds 49 and 50 (R = Et and Me, respectively) were quite active, while the fullerene derivative 48 (R = Cl) was completely inactive. Specifically, 49 and 50 effectively inhibited influenza virus H3N2 at nanomolar concentrations, with an EC_{50} of 500 nM and 100 nM, respectively; both compounds were more active against H3N2 than the commercial drugs zanamivir (EC_{50} = 3.0 µM) and amantadine (EC_{50} = 1.3 µM). However, there were no significant differences in the HIV-1 inhibitory activity of the fullerene derivative C_{60}(Ar)_{5}Cl after the alkyl substitution of chlorine atoms.

Figure 14. Schematic diagram of synthetic routes of carboxylic fullerene derivatives 49 and 50.

Recently, Troshin and coworkers [63] synthesized the fullerene carboxylic acid derivative C_{70}[p-C_{6}H_{4}(CH_{2})_{6}COOH]_{8} by Friedel–Crafts arylation of chlorofullerene C_{70}Cl_{8} with unprotected carboxylic acids. The obtained carboxylic acid fullerene C_{70} derivatives showed significant antiviral effects against HIV and the influenza viruses H1N1 and H3N2. The EC_{50} value of anti-HIV-1/NL4.3 (X4) is close to 1.0 µM, which indicates that this derivative is more effective against the virus than the commercial drug tenofovir. Additionally, Troshin and coworkers synthesized a variety of carboxylic acid fullerene derivatives, such as the carboxylic acid thiofullerene derivative, 51 [64]; polycarboxylic acid fullerene derivative, 52 [65]; and tetracarboxylic acid methanofullerene derivative, 53 (Figure 15) [66]. All these compounds exhibit inhibitory effects against HIV-1, HIV-2, influenza A (H3N2), HSV, and CMV, with low toxicity.

Modern antiviral drugs have extended patients’ lives and improved their quality of life, but unsolved problems remain, such as toxicity, limited bioavailability, and drug resistance (rapid cyclic changes in influenza strains reduce the effectiveness of commonly used vaccines). Therefore, finding new antiviral drugs has become an urgent problem. The research by Troshin and coworkers on carboxylic acid fullerene derivatives has provided more possibilities for the development of new and effective antiviral drugs [61–66].
5. Synthesis and Antiviral Studies of Fullerene Hydroxyl Derivatives

Fullerenol is a polyhydroxylated fullerene C₆₀ derivative with suitable water solubility and biocompatibility. In 2013, Eropkin et al. [67] synthesized a series of fullerenols (Figure 16). Three different groups of fullerenols, C₆₀(OH)₁₂–₁₄, C₆₀(OH)₁₈–₂₄, and C₆₀(OH)₃₀–₃₈, can be prepared by changing the reaction conditions to control the number of hydroxyl groups attached to the fullerene skeleton. Experimental studies revealed that fullerenols containing 12–14 hydroxyl groups are insoluble in water and have no biological activity when introduced into cell culture as suspensions. The other two groups of fullerenols show broad spectrum antiviral activity in vitro against the human influenza viruses H1N1 and H3N2, avian influenza virus A (H5N1), adenovirus, human HSV, and respiratory syncytial virus. C₆₀(OH)₁₈–₂₄ demonstrates better antiviral activity than C₆₀(OH)₃₀–₃₈. Moreover, the three water-soluble fullerenols exhibit no toxicity in vitro to human and animal cells.

Compared with other fullerene derivatives, fullerenols are relatively easy to prepare, requiring only one organic reaction step. So far, only one example of polyhydroxyfullerene (C₆₀(OH)₈) with a well-defined structure has been reported, by Gan and co-workers [68]. However, the insufficient number of hydroxyl groups in C₆₀(OH)₈ limits its biological applications. At present, even by purification via high performance liquid chromatography, it is impossible to obtain pure regioisomer fullerenols from the mixture of polyhydroxyfullerenes. Therefore, the fullerenols used in current biological studies have been a mixture of regioisomers, which restricts the potential of fullerenols as standard drug candidate molecules.

6. Synthesis and Antiviral Studies of Glycofullerene Derivatives

Carbohydrate and protein interactions dominate many biological processes, including inflammation, embryogenesis, tumor development and metastasis, and pathogen infection [69]. These interactions are characterized by high selectivity, metal ion dependence, and compensation for low affinity through multivalent interactions [70]. Finding a suitable system to realize the polyvalent expression of sugars has been a subject of extensive

---

**Figure 15.** Structure diagram of polycarboxylic fullerene acid derivatives (51–53).

**Figure 16.** Structure diagram of fullerenol. Oxygen, carbon, and hydrogen atoms are marked in red, gray, and white, respectively.
Due to the lack of information on the proper orientation of the ligands required to obtain the strongest interactions, chemists have experimented with all possible scaffolds. Calixarene, gold nanoparticles, polymers, liposomes, dendritic macromolecules, and fullerenes are most commonly used as scaffolds to construct multivalent glycoconjugates. The advantages associated with fullerenes over other nanostructures are their three-dimensional (3D) structure and the ability to functionalize different positions of the C$_{60}$ cage in a controlled way. In this sense, fullerenes can be thought of as a special class of spherical scaffolds ideal for building a multivalent spherical ligand. Martin and coworkers proposed that spherical carbohydrate derivatives of fullerenes, with fullerenes used as scaffolds, could serve as an interesting sugar analog. The rigid spherical scaffold allows distance to be maintained between the two ligands (the diameter of fullerenes is 1 nm, plus the distance provided by the dendritic moiety), thus increasing the chances of obtaining effective multivalent interactions. In addition, due to the symmetry of the multivalent system, the 3D orientation of these ligands at 360° better mimics the surface of a pathogen, such as HIV, thus, the molecule is more likely to encounter a receptor.

Based on an octahedral addition pattern, soluble hexakis-adduct glycofullerene derivatives can act as spherical carbohydrates, thus serving as a potential multivalent spherical ligand. In 2013, Martin and coworkers designed and synthesized a class of hexakis-adduct glycofullerene derivatives, containing 36 mannoses, and used them to inhibit cell infection by pseudotyped Ebola virus particles. This was the first time that glycofullerene derivatives were demonstrated to effectively inhibit cell infection. In the pseudotyped Ebola infection model, the antiviral activity of glycofullerene with 12 galactosyl had no inhibitory effect on virus infection, indicating that the inhibitory effect is dependent on mannose. Interestingly, only an increase in the valence of glycofullerene resulted in a loss of the antiviral effect. This phenomenon is related to the spatial crowding of sugars at the surface of glycofullerene. Martin et al. speculated that the high binding affinity occurs not only because of the extensive spatial presentation of multivalent ligands, but also the frequent interactions between the ligands and corresponding receptors. They demonstrated that a rational design of compounds with the same valency but longer spacers can significantly increase the antiviral activity, likely due to more efficient interactions with receptors. Therefore, the selection of suitable scaffolds (such as spherical fullerenes) to achieve multivalence, as well as the accessibility and flexibility of ligands, are key factors for improving antiviral activity.

The aforementioned studies have demonstrated that using multivalent glycofullerenes to block lectin receptors on the cell surface is a promising method for inhibiting virus entry into cells. However, creating large enough multivalent glycofullerenes to improve the binding ability between ligands and virus receptors remains a challenge. In 2015, Martin and coworkers conducted an impressive study of water-soluble fullerenes. They synthesized three water-soluble glycofullerene derivatives with 12 fullerenes modified with 120 sugars, starting from the hex-adduct fullerene derivative, namely, the “super sphere.” In addition to the core fullerene sphere, the other 12 fullerenes in the super sphere contain 10 sugars each, totaling 120 sugars, and have diameters up to 4 nm.

The mannose in compounds is critical in blocking virus entry into cells. As expected, with 120 galactosyl species, did not inhibit the infection process. Glycofullerene derivatives with 120 mannoses, exhibited high levels of antiviral activity in the pM–nM range. As shown in Table 4, can effectively block Ebola virus infection in the nM range, with an IC$_{50}$ of 20.4 nM. Compound is almost 10 times more potent in the antiviral infection process, with an IC$_{50}$ of 667 pM. Fullerene, linked with 36 mannoses, produced relative inhibitory effect (RIP) values of at least two orders of magnitude smaller than those of and . Compared to previously reported results, and are the most effective compounds against Ebola virus infection in vitro. Compounds showed no appreciable cell cytotoxicity at the concentrations used in the virus inhibition experiments. Owing
to their biocompatibility and spherical structure, fullerenes have become ideal scaffolds for studying multivalent interactions. However, further research is needed to determine whether the obtained glycofullerene derivatives can be used for practical applications.

Figure 17. Structure diagram of glycofullerene (54). Adapted with permission from Ref. [78]. Copyright 2013 American Chemical Society.

Table 4. IC$_{50}$ and RIP values of different glycofullerene derivatives.

| Compound          | IC$_{50}$/nM | Mannoses (No.) | RIP $^*$ |
|-------------------|--------------|----------------|---------|
| 56 (120 mannoses) | 0.667        | 120            | $1.58 \times 10^4$ |
| 58 (120 mannoses) | 20.375       | 120            | $5.2 \times 10^2$ |
| 54 (36 mannoses)  | 300          | 36             | $1.17 \times 10^2$ |

$^*$ Relative inhibitory effect, calculated as (IC$_{50}$/mono)/IC$_{50}$ $^*$ valency (IC$_{50}$/mono, IC$_{50}$ of the monovalent compound; IC$_{50}$ $^*$ valency, IC$_{50}$ of the multivalent compound multiplied by the number of ligands present in the multivalent compound).
synthesized three water-soluble glycofullerene derivatives (56–58) with 12 fullerene spheres modified with 120 sugars, starting from the hex-adduct fullerene derivative (55), namely, the “super sphere” (Figure 18). In addition to the core fullerene sphere, the other 12 fullerenes in the super sphere contain 10 sugars each, totaling 120 sugars, and have diameters up to 4 nm.

In order to enhance multivalency and improve the biocompatibility of glycofullerene derivatives, Martin and coworkers [80] (2019) synthesized tridecafullerene derivatives containing up to 360 1,2-mannobiosides via the strain-promoted azide–alkyne cycloaddition method. The obtained glycofullerene derivative showed pronounced antiviral activity against Zika virus and dengue virus, with an IC50 of 67 and 35 pM, respectively.

Additionally, Martin and coworkers [81] synthesized a series of amphiphilic glycodendrofullerene [60] monoadducts (59 and 60) through the “click chemistry” reaction. In aqueous media, the glycodendrofullerenes can self-assemble into large, compact micelles with a uniform size and spherical shape. Antiviral tests showed that these aggregates of 59 and 60 can effectively inhibit Ebola virus infection in the nM range, with an IC50 of 424 nM and 196 nM, respectively. However, these compounds are inferior to 56 and 58.

Ebola virus has a filament-like structure and is similar to single-walled carbon nanotubes (SWCNTs) and multi-walled carbon nanotubes (MWCNTs) in shape. SWCNTs are usually 1.5 to 3.0 nm in diameter. They match Ebola virus in length, at 20 to 1000 nm. On the other hand, HIV has a roughly spherical shape of about 120 nm in diameter and is very similar in shape and size to spherical fullerenes and single-walled nanocones (SWCNHs) [82]. Therefore, Martin and coworkers proposed that SWCNTs, MWCNTs, and SWCNHs could be used as virus-mimicking nanocarbon platforms, and after chemical modification, could interact with the receptors in a multivalent manner. To employ the novel strategy for designing antiviral agents against HIV or Ebola virus (designing glycofullerene and nanocarbon complexes that mimic the virus surface and interfere with the infection of receptors on the corresponding cell surface), Martin et al. [83] (2018) used the “click chemistry” reaction to covalently connect glycofullerene to SWCNTs, MWCNTs, or SWCNHs. The multivalent hybrid glycoconjugate consisting of MWCNTs and glyco-

Figure 18. Synthesis of glycofullerenes 56–58 [45]. Adapted with permission from Ref [45]. Copyright 2015 Springer Nature.
fullerene (61) is shown in Figure 19. After chemical modification, the water solubility of the MWCNTs was significantly improved. In tests of efficiency in blocking artificial Ebola virus infection with three types of multivalent hybrid glycoconjugates, 61, based on MWCNTs and fullerene functionalization, was the most effective inhibitor of viral infection, with an IC$_{50}$ of 0.37 µg/mL.

Cyclodextrins (CDs) are a series of cyclic oligosaccharides containing 6–12 D-glucopyranose units. They have a slightly conical, hollow, cylindrical three-dimensional ring structure and have been widely studied because of their significant solubility in water. The CD molecules containing 6, 7, and 8 glucose units are called α-, β- and γ-CD, respectively. In 2012, Zhang and coworkers [84] prepared a water-soluble α-CD-C$_{60}$ conjugate (62), in which C$_{60}$ and α-CD were linked by two flexible alkyl chains (Figure 20). Interestingly, this water-soluble compound showed pronounced anti-HCV entry activity, with an IC$_{50}$ of 0.17 µM.

In 2018, Zhang and coworkers [85] continued to design and synthesize seven α-CD-C$_{60}$ copolymers and two γ-CD-C$_{60}$ copolymers [85]. However, the newly obtained α-CD-C$_{60}$ conjugates did not exhibit inhibitory activity against HCV. Subsequently, all nine CD-C$_{60}$ conjugates were assessed in terms of their activity against the influenza virus H1N1. No

![Figure 19](image1.png)

**Figure 19.** Structure diagram of nanoglycofullerene conjugate 61 [83]. Adapted with permission from Ref. [83]. Copyright 2018 American Chemical Society.

![Figure 20](image2.png)

**Figure 20.** α-Cyclodextrin-C$_{60}$ conjugate (62) [84]. Adapted with permission from Ref. [84]. Copyright 2012 Elsevier.
conjugates exhibited cytotoxicity at 100.0 μM. The two γ-Cd-C60 conjugates demonstrated higher anti-H1N1 activity, with IC50 values of 87.7 μM and 75.0 μM, respectively. Because they exhibit less aggregation in aqueous solutions, the two γ-Cd-C60 conjugates are the most water soluble of the nine conjugates. This trait might be related to their higher inhibitory efficiency against H1N1.

7. Synthesis and Antiviral Studies of Fullerene Complexes

As mentioned, to enhance the water solubility of pristine fullerenes, various hydrophilic functional groups (amino acids, sugars, or calixarene) can be directly or indirectly used to modify fullerenes through covalent bonds. Alternatively, pristine fullerenes can be dispersed in polyvinylpyrrolidone (PVP), Triton X-100, dioctadecyldimethylammonium bromide, or lecithin to form complexes.

Sirotkin et al. [86] added an aqueous solution of the C60/PVP complex to a suspension of H1N1 particles. As a result, the number of virus particles with a broken lipoprotein envelope increased dramatically, possibly due to the fusion of the C60/PVP complex with the influenza virus.

Yang and coworkers [87] prepared a fullerene [60] liposome complex (Figure 21) and studied its anti-H1N1 activity in vivo. The fullerene liposome complex significantly reduces the average lung virus yields and lung index; prolongs the mean time to death; and decreases the mortality of mice infected with H1N1. In addition, the fullerene liposome complex has good water solubility and low toxicity, and its anti-influenza activity in vivo is much higher than that of rimantadine. Therefore, the fullerene [60] liposome complex is a promising clinical candidate drug against influenza infection.

![Figure 21. The structure of the fullerene liposome complex [87].](image)

8. Conclusions

This review summarized the latest antiviral research conducted on fullerenes and their derivatives. Numerous water-soluble fullerene derivatives or fullerene complexes have shown great antiviral potential, mainly because fullerenes have three advantages. First, pristine fullerenes are hydrophobic, which is conducing to the formation of strong hydrophobic interactions with the active site surfaces of viruses. Second, hydrophilic groups with various functions (such as amino, carboxyl, amino acid, hydroxyl, pyrrolidine, and sugar groups) can be used to selectively modify the unique spherical skeleton of fullerenes via organic reactions. Third, fullerenes and their derivatives exhibit no or low cytotoxicity at relatively high concentrations. Although fullerenes are promising prospective antiviral drugs, antiviral research on fullerenes requires improvement. Most fullerene derivatives exhibit good antiviral effects in vitro, but the antiviral mechanism has not been thoroughly studied. Additionally, most of the studies on fullerenes have
only focused on virus inhibition in vitro; there have been few antiviral studies in vivo, and relevant clinical studies involving fullerenes have not been conducted.

Viruses constantly threaten human health. Fullerenes have become an important molecular platform for the development of antiviral drugs. Research on fullerenes as antiviral drugs urgently needs the joint efforts of scientists working in synthesis, molecular design, biology, and medicine. Some fullerene derivatives display inhibitory activity against multiple types of viruses. Therefore, fullerene derivatives have the potential to become a class of broad-spectrum antiviral drugs effective against SARS-CoV-2, which remains a global threat. We believe that this review will encourage more researchers to synthesize fullerene derivatives and study their antiviral properties and applications.

**Funding:** This research was funded by the National Natural Science Foundation of China (NSFC) (91961113, 21721001, 92061204, 21827801 and 92061000); Xiamen Youth Innovation Fund (3502Z20206054); the Funds of the Science and Technology Project of Yunnan Province-Major Project (202101AS070049).

**Institutional Review Board Statement:** Not applicable.

**Informed Consent Statement:** Not applicable.

**Data Availability Statement:** Not applicable.

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

**References**

1. Kroto, H.W.; Heath, J.R.; O’Brien, S.C.; Curl, R.F.; Smalley, R.E. C_{60}: Buckminsterfullerene. *Nature* **1985**, *318*, 162–163. [CrossRef]
2. Krätschmer, W.; Lamb, L.D.; Fostiropoulos, K.; Huffman, D.R. Solid C_{60}: A new form of carbon. *Nature* **1990**, *347*, 354–358. [CrossRef]
3. Howard, J.B.; McKinnon, J.T.; Makarovsky, Y.; Lafleur, A.L.; Johnson, M.E. Fullerenes C_{60} and C_{70} in flames. *Nature* **1991**, *352*, 139–141. [CrossRef]
4. Tian, H.-R.; Chen, M.-M.; Wang, K.; Chen, Z.-C.; Fu, C.-Y.; Zhang, Q.; Li, S.-H.; Deng, S.-L.; Yao, Y.-R.; Xie, S.-Y.; et al. An Unconventional Hydrofullerene C_{66}H_{4} with Symmetric Heptagons Retrieved in Low-Pressure Combustion. *J. Am. Chem. Soc.* **2019**, *141*, 6651–6657. [CrossRef] [PubMed]
5. Zhang, H.-G.; Zhuo, Y.-Q.; Zhang, X.-M.; Zhang, L.; Xu, P.-Y.; Tian, H.-R.; Lin, S.-C.; Zhang, Q.; Xie, S.-Y.; Zheng, L.-S. Synthetic Fullerenes from a Nonaromatic Chloroform through a Newly Developed Ultrahigh-Temperature Flash Vacuum Pyrolysis Apparatus. *Nanomaterials* **2021**, *11*, 3033. [CrossRef] [PubMed]
6. Yang, B.; Chen, Y.; Shi, J. Reactive Oxygen Species (ROS)-Based Nanomedicine. *Chem. Rev.* **2019**, *119*, 4881–4895. [CrossRef]
7. Rosi, N.L.; Mirkin, C.A. Nanostructures in Biodiagnostics. *Chem. Rev.* **2005**, *105*, 1547–1562. [CrossRef]
8. Ni, K.; Luo, T.; Nash, G.T.; Lin, W. Nanoscale Metal–Organic Frameworks for Cancer Immunotherapy. *Acc. Chem. Res.* **2020**, *53*, 1739–1748. [CrossRef]
9. Kirtane, A.R.; Verma, M.; Karandikar, P.; Furin, J.; Langer, R.; Traverso, G. Nanotechnology approaches for global infectious diseases. *Nat. Nanotechnol.* **2021**, *16*, 369–384. [CrossRef]
10. Li, S.; Jiang, Q.; Liu, S.; Zhang, Y.; Tian, Y.; Song, C.; Wang, J.; Zou, Y.; Anderson, G.J.; Han, J.-Y.; et al. A DNA nanorobot functions as a cancer therapeutic in response to a molecular trigger in vivo. *Nat. Biotechnol.* **2018**, *36*, 258–264. [CrossRef]
11. Ji, T.; Zhao, Y.; Ding, Y.; Wang, J.; Zhao, R.; Lang, J.; Qin, H.; Liu, X.; Shi, J.; Tao, N.; et al. Transformable Peptide Nanocarriers for Expeditious Drug Release and Effective Cancer Therapy via Cancer-Associated Fibroblast Activation. *Angew. Chem. Int. Ed.* **2016**, *55*, 1050–1055. [CrossRef] [PubMed]
12. Han, X.; Li, Y.; Xu, Y.; Zhao, X.; Zhang, Y.; Yang, X.; Wang, Y.; Zhao, R.; Anderson, G.J.; Zhao, Y.; et al. Reversal of pancreatic desmoplasia by re-educating stellate cells with a tumour microenvironment-activated nanosystem. *Nat. Commun.* **2018**, *9*, 3390. [CrossRef] [PubMed]
13. Lu, F.; Gu, L.; Meziani, M.J.; Wang, X.; Luo, P.G.; Vea, L.M.; Cao, L.; Sun, Y.-P. Advances in Bioapplications of Carbon Nanotubes. *Adv. Mater.* **2009**, *21*, 139–152. [CrossRef]
14. Feng, L.; Zhang, S.; Liu, Z. Graphene based gene transfection. *Nanoscale* **2011**, *3*, 1252–1257. [CrossRef] [PubMed]
15. Zhu, M.; Nie, G.; Meng, H.; Xia, T.; Nel, A.; Zhao, Y. Physicochemical Properties Determine Nanomaterial Cellular Uptake, Transport, and Fate. *Acc. Chem. Res.* **2013**, *46*, 622–631. [CrossRef]
16. Dellinger, A.; Zhou, Z.; Connor, J.; Madhankumar, A.; Pamujula, S.; Sayes, C.M.; Kepley, C.L. Application of fullerenes in nanomedicine: An update. *Nanomedicine* **2013**, *8*, 1191–1208. [CrossRef]
17. McEwen, C.N.; McKay, R.G.; Larsen, B.S. C_{60} as a radical sponge. *J. Am. Chem. Soc.* **1992**, *114*, 4412–4414. [CrossRef]
18. Maas, M. Carbon Nanomaterials as Antibacterial Colloids. *Materials* **2016**, *9*, 617. [CrossRef]
19. Li, L.; Zhen, M.; Wang, H.; Sun, Z.; Jia, W.; Zhao, Z.; Zhou, C.; Liu, S.; Wang, C.; Bai, C. Functional Gadofullerene Nanoparticles Trigger Robust Cancer Immunotherapy Based on Rebuilding an Immunosuppressive Tumor Microenvironment. *Nano Lett.* 2020, 20, 4487–4496. [CrossRef]

20. Liu, Q.; Guan, M.; Xu, L.; Shu, C.; Jin, C.; Zheng, J.; Fang, X.; Yang, Y.; Wang, C. Structural Effect and Mechanism of C70-Carboxyfullerenes as Efficient Sensitizers against Cancer Cells. *Small* 2012, 8, 2070–2077. [CrossRef]

21. MacFarland, D.K.; Walker, K.L.; Lenk, R.P.; Wilson, S.R.; Kumar, K.; Kepley, C.L.; Garbow, J.R. Hydrochaloranes: A Novel Endohedral Metallofullerene Platform for Enhancing Magnetic Resonance Imaging Contrast. *J. Med. Chem.* 2008, 51, 3681–3683. [CrossRef]

22. Zheng, J.-p.; Liu, Q.-l.; Zhen, M.-m.; Jiang, F.; Shu, C.-y.; Jin, C.; Yang, Y.; Alhadaq, H.A.; Wang, C.-r. Multifunctional imaging probe based on gadofulleride nanoplatform. *Nanoscale* 2012, 4, 3669–3672. [CrossRef] [PubMed]

23. Zhou, C.; Zhen, M.; Yu, M.; Li, X.; Yu, T.; Liu, J.; Jia, W.; Li, S.; Li, L.; et al. Gadofullerene inhibits the degradation of apolipoprotein B100 and boosts triglyceride transport for reversing hepatic steatosis. *Sci. Adv.* 2020, 6, eabc1586. [CrossRef] [PubMed]

24. Liu, Y.; Chen, C.; Qian, P.; Zhu, T.; Zhao, Y. Gd-metallofullerenol nanomaterial as non-toxic breast cancer stem cell-specific inhibitor. *Nanomed. Nanotechnol. Biomed. Imaging* 2016, 12, 510. [CrossRef]

25. Kang, S.-G.; Zhou, G.; Yang, P.; Liu, Y.; Sun, B.; Huynh, T.; Meng, H.; Zhao, L.; Xing, G.; Chen, C.; et al. Molecular mechanism of pancreatic tumor metastasis inhibition by Gd@C_{60}(OH)_{22} and its implication for de novo design of nanomedicine. *Proc. Natl. Acad. Sci. USA* 2012, 109, 15431–15436. [CrossRef] [PubMed]

26. Deng, R.; Wang, Y.; Zhen, M.; Li, X.; Zou, T.; Li, J.; Yu, T.; Zhou, Y.; Lu, Z.; Xu, H.; et al. Real-time monitoring of tumor vascular disruption induced by radiofrequency assisted gadofullerenes. *Sci. China Mat.* 2018, 61, 1101. [CrossRef]

27. Zhen, M.; Shu, C.; Li, J.; Zhang, G.; Wang, T.; Luo, Y.; Zou, T.; Deng, R.; Fang, F.; Lei, H.; et al. A highly efficient and tumor vascular-targeting therapeutic technique with size-expansible gadofullerene nanocrystals. *Sci. China Mat.* 2015, 58, 799–810. [CrossRef]

28. Zhou, Y.; Deng, R.; Zhen, M.; Li, J.; Guan, M.; Jia, W.; Li, X.; Zou, T.; et al. Amino acid functionalized gadofullerene nanoparticles with superior antitumor activity via destruction of tumor vasculature in vivo. *Biomaterials* 2017, 133, 107–118. [CrossRef]

29. Zhou, Y.; Li, J.; Ma, H.; Zhen, M.; Guo, J.; Wang, L.; Jiang, L.; Shu, C.; Wang, C. Biocompatible [60]/[70] Fullerenols: Potent Defense against Oxidative Injury Induced by Reduplicative Chemotherapy. *ACS Appl. Mater. Interfaces* 2017, 9, 35539–35547. [CrossRef]

30. Zhou, Y.; Zhen, M.; Ma, H.; Li, J.; Zou, T.; Wang, C. Inhalable gadofullerenol/[70] fullerenol as high-efficiency ROS scavengers for pulmonary fibrosis therapy. *Nanomed. Nanotechnol. Biotechnol. Med.* 2016, 12, 510. [CrossRef]

31. Norton, S.K.; Wijesinghe, D.S.; Delliger, A.; Sturgill, J.; Zhou, Z.; Barbour, S.; Chalfant, C.; Conrad, D.H.; Kepley, C.L. Epoxideicosatrienoic acids are involved in the C70 fullerene derivative–induced control of allergic asthma. *J. Allergy Clin. Immunol.* 2012, 130, 761–769.e762. [CrossRef]

32. Zhou, Z.; Lenk, R.P.; Delliger, A.; Wilson, S.R.; Sadler, R.; Kepley, C.L. Liposomal Formulation of Amphiphilic Fullerene Antioxidants. *Bioconjugate Chem.* 2010, 21, 1656–1661. [CrossRef]

33. Basso, A.S.; Frenkel, D.; Quintana, F.J.; Costa-Pinto, F.A.; Petrovic-Stojkovic, S.; Pucett, L.; Monsonego, A.; Bar-Shir, A.; Engel, Y.; Gozin, M.; et al. Reversal of axonal loss and disability in a mouse model of progressive multiple sclerosis. *J. Clin. Investig.* 2008, 118, 1532–1543. [CrossRef]

34. Tokuyama, H.; Yamago, S.; Nakamura, E.; Shiraki, T.; Sugira, Y. Photoinduced biochemical activity of fullerene carboxylic acid. *J. Am. Chem. Soc.* 1993, 115, 7918–7919. [CrossRef]

35. Dugan, L.L.; Turetsky, D.M.; Du, C.; Lobner, D.; Wheeler, M.; Almli, C.R.; Shen, C.K.-F.; Luh, T.-Y.; Choi, D.W.; Lin, T.-S. Carboxyfullerenes as neuroprotective agents. *Proc. Natl. Acad. Sci. USA* 1997, 94, 9434–9439. [CrossRef]

36. Wong, C.-W.; Zhilenkov, A.V.; Kraevaya, O.A.; Mischenko, D.V.; Troshin, P.A.; Hsu, S.-h. Toward Understanding the Antitumor Activities of Fullerene Derivatives of Water-Soluble Fullerenes: Development of Angiogenesis Inhibitors? *J. Med. Chem.* 2019, 62, 7111–7125. [CrossRef]

37. Sigwalt, D.; Holler, M.; Iehl, J.; Nierengarten, J.-F.; Nothisen, M.; Morin, E.; Remy, J.-S. Gene delivery with polycationic fulleren hexakis-adducts. *Chem. Commun.* 2011, 47, 4640–4642. [CrossRef]

38. Fan, J.; Fang, G.; Zeng, F.; Wang, X.; Wu, S. Water-Dispersible Fullerene Aggregates as a Targeted Anticancer Prodrug with both Chemo- and Photodynamic Therapeutic Actions. *Small* 2013, 9, 613–621. [CrossRef]

39. Friedman, S.H.; DeCamp, D.L.; Sibbsma, R.P.; Srdanov, G.; Wudl, F.; Kenyon, G.L. Inhibition of the HIV-1 protease by fullerene derivatives: Model building studies and experimental verification. *J. Am. Chem. Soc.* 1993, 115, 6506–6509. [CrossRef]

40. Mashino, T.; Shimotohno, K.; Ikegami, N.; Nishikawa, D.; Okuda, K.; Takahashi, K.; Nakamura, S.; Mochizuki, M. Human immunodeficiency virus-reverse transcriptase inhibition and hepatitis C virus RNA-dependent RNA polymerase inhibition activities of fullerene derivatives. *Bioorg. Med. Chem. Lett.* 2005, 15, 1107–1109. [CrossRef]

41. Castro, E.; Martinez, Z.S.; Seong, C.-S.; Cabrera-Espinoza, A.; Ruiz, M.; Hernandez Garcia, A.; Valdez, F.; Llano, M.; Echegoyen, L. Characterization of New Cationic N,N-Dimethyl [70]fulleropyrrolidinium Iodide Derivatives as Potent HIV-1 Maturaton Inhibitors. *J. Med. Chem.* 2016, 59, 10963–10973. [CrossRef]
