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Research on Magnetically Targeted Drug Delivery System Based on Fullerene Derivatives

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A carboxyl-terminated fullerene pyrrolidine derivative was synthesized by 1, 3-dipolar cycloaddition of imine ylide (FP-COOH). UV-Vis, FT-IR and MALDI-TOF respectively verified the effective synthesis of compounds. The compound (FP-COOH) was used as an intermediate, and then the hydrothermal chemical bonding method was used to load ferric oxide on the compound (FP-COOH). Its purpose was to form a magnetic targeting carrier system (FP-IONP-COOH). Then use the non-covalent method to combine FP-IONP-COOH with doxorubicin. The ultimate goal was to improve the side effects of doxorubicin. The solubility experiments showed that both FP-IONP-COOH and FP-IONP-COOH/DOX had good water solubility. The investigation of magnetism showed that FP-IONP-COOH has good magnetism. Finally, in vitro release experiments further verified the targeting of FP-IONP-COOH/DOX. The cumulative release of DOX at 48 h could be as high as 82 %, whereas the accumulated release of FP-IONP-COOH/DOX at 48 h was only 48 %, and was able to continuously release for more than 120 h, demonstrating its good sustained release in vivo.
Keywords: carboxyl-terminated fullerene pyrrolidine, iron oxide, magnetic targeting, doxorubicin.

1. INTRODUCTION

With the discovery of the third allotrope of carbon-fullerene [C_{60}], and the generation and establishment of its ton-scale method [1], numerous experts and scholars from various fields have scrambled to conduct a variety of research on C_{60}. The study found that C_{60} has many excellent features that can be used in many applications. However, due to the strong lipophilicity of the C_{60} cage structure, it exhibits strong hydrophobic properties, which severely limits its biological effects and its application to anti-tumor, drug carrier, anti-microbial, anti-oxidative stress, etc. Therefore, how to introduce C_{60} into an aqueous system is an effective way to break through this dilemma. One of the methods is to chemically modify the surface of C_{60} to introduce a hydrophilic group to form a C_{60} derivative. With the deepening and maturity of the chemical modification of C_{60}, there are more and more functional groups attached to C_{60}, and the successful synthesis of these fullerene derivatives with functional groups is often attributed to some key fullerene intermediates [2]. Fullerene intermediates generally have active chemical properties and can react with a variety of chemical groups to give a wider variety of fullerene derivatives. Fullerene pyrrolidine is the most common fullerene intermediate. Since Maggini, Prato synthesized N-methylfullerene pyrrolidine and C_{60}-TTF by the 1,3-dipolar cycloaddition reaction of imine ylide [3 – 4], the study of synthesizing fullerene pyrrolidine by this type of reaction was initiated, many reports have emerged in recent studies. Peyghan A.A. et al. showed that fullerene pyrrolidine not only has higher conductivity and better water solubility than raw fullerenes, but also improves its electrophilicity by 23 – 37 % [5]. Grinholc found that mono-N-methylpyrrolidinium fullerene iodide has a killing effect on Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus and Candida albicans under light irradiation. In vitro studies have found that the C_{60} derivative does not inhibit the toxicity of Escherichia coli and Candida albicans under dark conditions, and produces reactive oxygen species to destroy bacterial cell membranes under light, inhibiting all bacteria and highly inhibiting Staphylococcus [6]. Guan et al reported a UCNP-PEG-FA/PC_{70} nanocomposite (UCNP: upconversion nanoparticles, PC_{70}: trimethylpyridyl porphyrin-fullerene C_{70}), which can overcome the lack of light penetration depth and microenvironment hypoxia in PDT, and can accumulate in the tumor site under the active targeting of FA. Later, a TF C_{70}-OEG2-Ce6 nanovesicle that can be used for tumor imaging and PDT has been reported, and it has the following advantages: 1) Ce6 has a high loading rate (mass fraction of 57 %); 2) can be effectively absorbed in NIR; 3) improve cell uptake rate in vitro and in vivo; 4) enhance tumor imaging and PDT efficiency; 5) good biocompatibility and systemic clearance [7].

Doxorubicin is a broad-spectrum anti-tumor drug. It not only has lipid-soluble anthracycline ligand and water-soluble daunorubicin, but also has acid phenolic hydroxyl group and basic amino group. It is easy to penetrate the cell membrane into cancer cells and therefore has strong antitumor activity and pharmacological activity. Doxorubicin kills cells in different growth cycles, so it is suitable for the treatment of many common cancers such as breast cancer, stomach cancer, bladder cancer, liver cancer,

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lung cancer and malignant lymphoma. However, doxorubicin also produces free radical-mediated toxic side effects on liver, kidney, heart, brain, lung, blood and testis during administration [8 – 10]. Fullerene has excellent free radical scavenging function and can also act as drug carrier. The results show that this not only improves the administration efficiency, but also reduces the side effects and enhances the anti-tumor effect.

In this experiment, a carboxyl-terminated fullerene pyrrolidine (FP-COOH) was synthesized by the experimental method of Bjelaković et al. [11 – 14], by a 1, 3-dipolar cycloaddition method of imine ylide. The fullerene pyrrolidine intermediate has a free carboxyl structure, which not only improves the hydrophobicity of the fullerene, but also has an amidation reaction, an esterification reaction and the like due to its active chemical properties.

At the same time, in order to solve the problem of drug targeting, this experiment also synthesized a ferroferric oxide-loaded magnetic targeting carrier system (FP-IONP-COOH) by hydrothermal bonding reaction on carboxyl-terminated fullerene pyrrolidine (FP-COOH), and based on this, non-covalent binding of doxorubicin to solve the cytotoxicity problem of doxorubicin.

2. MATERIALS AND METHODS

2.1. Instruments

ZF-2 Triple Ultraviolet (Shanghai Anting Electronic Instrument Factory), RE-2000B Rotary Evaporator (Gongyi Yuhua Co., Ltd.), UV-2450 Ultraviolet Visible Spectrophotometer (SHIMADIU Company of Japan), IR-2000 Fourier Infrared Spectrometer (Nikkoli Company of USA), 5800 MALDI-TOF Mass Spectrometer (AB Sciex Company of USA), Hydrothermal kettle (Gongyi City Station Street integrity instrument distribution department).

2.2. Materials

C_{60}, purity 99.9 %, purchased from Henan Yongyang Yongxin Reagent Co., Ltd.; benzyl chloroformate, γ-aminobutyric acid, benzyl bromoacetate, trifluoroacetic acid purchased from Aladdin; tert-butanol, petroleum ether, Ethyl acetate, dichloromethane, triethylamine, toluene, diethyl ether, chloroform, anhydrous methanol, methanol, anhydrous calcium chloride, anhydrous sodium sulfate, phosphorus pentoxide, ammonium sulfate, sodium hydroxide, hydrochloric acid, carbonic acid Sodium hydrogen was purchased from Komi Ore Reagent Co., Ltd.; 4-dimethylaminopyridine and dicyclohexylcarbodiimide were purchased from Beijing Bomaijie Co., Ltd.; Ethylene glycol was purchased from Sinopharm Chemical Reagent Co., Ltd.; From Ron Reagent Co., Ltd.; sodium acetate was purchased from Tianjin Sailboat Chemical Reagent Technology Co., Ltd.; sodium acrylate was purchased from Aiwang Chemical Technology Co., Ltd.; FeCl3.6H2O was purchased from Tianjin Yongda Chemical Reagent Co., Ltd.; 10 % Pd/ C was purchased from Alfa Aesar Company; Paraformaldehyde was purchased from Tianjin Kaitong Chemical Reagent Co., Ltd.; GF254 silica gel plate and column chromatography silica gel were purchased from Qingdao Ocean Chemical Co., Ltd.

2.3. Synthesis procedures

The synthesis route is shown in Fig. 1, compound 5 is synthesized by a series of reactions. The target product FP-COOH can be obtained by 1,3-dipole cycloaddition reaction of compound 5 and C_{60} and removal of OtBu protective group.

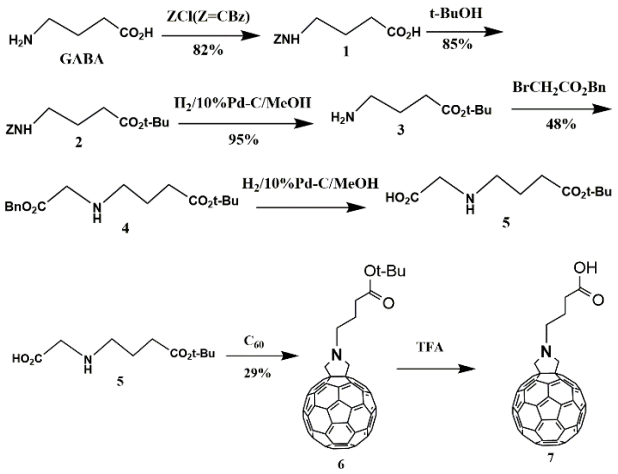


Fig. 1. Synthesis route of FP-COOH

Synthesis of compound 1:

The 2 M NaOH (60 mL) solution dissolved with γ-aminobutyric acid (9 g) and benzyl chloroformate (16.5 g) were stirred at 0 – 10 ℃ for 30 minutes and then stirred for 5 hours at room temperature. The mixture was extracted by AcOEt, the water layer was retained and the water layer was adjusted to pH ≤ 3 with 1 M HCl solution, then the water layer was extracted by CH_2Cl_2. The organic layer was combined and washed with water and NH_4SO_4 brine in turn. Compound 1 (15.12 g, R_f = 0.12, PE/AcOEt = 4:1) was obtained by rotary evaporation and vacuum drying. The yield was about 82 %. ^{1}H-NMR(400MHz, CDCl3): δ7.41 ~ 7.32 (m, 5H), 5.13 (s, 2H), 4.92 (br s, 1H), 3.30 (q, 2H), 2.44 (t, 2H), 1.88 (quint, 2H), 1.28 (s, 1H).

Synthesis of compound 2:

At 0 ℃, compound 1 (12 g), 4-dimethylpyridine (494.4 mg), and t-butanol (11.6 mL) were sequentially added into dry CH_2Cl_2 (80 mL), then dicyclohexylcarbodiimide (10.016 g) was added to stir for about 30 min. After 30 min, the reaction was continued at room temperature for 12 h, DCU was removed by filtration the next day, and the PH of the filtrate was adjusted by 1 M HCl solution to be less than 3. After that, the organic phase was washed repeatedly with NH_4SO_4 brine, 5 % $NaHCO_3$ solution, and NH_4SO_4 brine. After washing, it is rotary evaporated and vacuum dried to obtain a crude yellow product. The crude product is separated by silica gel column chromatography, and purified by using PE/AcOEt = 9:1 as eluent to obtain compound 2 (12.64 g, R_f = 0.48, PE/AcOEt = 4:1, yield about 85 %). ^{1}H-NMR(400 MHz, $CDCl_3$): δ7.38 ~ 7.32 (m, 5H), 5.11 (s, 2H), 5.00 (br s, 1H), 3.25 (q, 2H), 2.29 (t, 2H), 1.81 (quint, 2H), 1.46 (s, 9H).

Synthesis of compound 3:

At room temperature, compound 2 (10 g) was dissolved in 30 mL methanol, then 10 %Pd/C (600 mg) was added and catalytic hydrogenolysis was carried out for 10 h through H_2. After completion of the reaction, the palladium carbon was

removed by suction filtration, and the filtrate was evaporated to dryness to give compound 3 (5.63 g, R_f = 0.33, PE/AcOEt = 4:1, yield: 95 %). ^{1}H-NMR(400MHz, $CDCl_3$): δ2.73 (q, 2H), 2.28 (t, 2H), 1.74 (quint, 2H), 1.46 (s, 9H).

Synthesis of compound 6:

In a 100 mL, flame dried, two necked round-bottomed flask, equipped with a dropping funnel, and a magnetic stirring bar, under N_2, were placed compound 3 (4 g) and TEA (2.5 mL) with 30 mL of dry CH_2Cl_2. The mixture was cooled at 0 ℃, and 2.5 mL of benzyl 2-bromoacetate (as a solution in 5 mL of dry CH_2Cl_2) were added over a period of 1 h. The reaction mixture was left at room temperature with stirring for 24 h. The next day, dry Et_2O (200 mL) was added to the product system, a white precipitate was formed in the process, and the precipitate was removed by suction filtration. The organic phase was washed with water, 1 M HCl, water, and then dried over anhydrous Na_2SO_4 overnight. The solvent was removed with a rotary evaporator and the remaining material was chromatographed on a silica gel column (SiO_2) with an PE/AcOEt = 3:1 as eluent, then dried in vacuo to give 3.17 g of a yellow oil. (R_f = 0.83, PE/AcOEt = 1:1, yield: 41 %).

2 g of this yellow oil was dissolved in 20 mL of anhydrous methanol, and 80 mg of 10 % Pd/C was added as a catalyst, followed by catalytic reaction at room temperature for 10 h under H_2 atmosphere. After completion of the reaction, the palladium carbon was removed by suction filtration, and the filtrate was evaporated to dryness to give a colorless oil. A large amount of dry Et_2O was added to the oil to give a white solid. After washing, centrifugation and vacuum drying, 1.27 g white powder product was obtained. (R_f = 0.83, PE/AcOEt = 1:1, yield: 41 %).

C_{60} (200 mg) was dissolved in dry PhMe (200 mL) by ultrasound, after dissolution, 300 mg of white powdery product and 100 mg of $(CH_2O)_n$ were added in turn, then refluxed at 130 °C under N_2 for 24 h. After 24 h, the crude black-brown product was obtained by spin evaporation and vacuum drying, then purified by column chromatography. The unreacted C_{60} was separated with PhMe as the eluent, and then the compound 6 was separated with PhMe:AcOEt = 4:1 as eluent. (73 mg, yield: 29 %).

Synthesis of compound 7 (FP-COOH):

20 mg of compound 6 was dissolved in 2 mL of CH_2Cl_2, then 4 mL of TFA was added and stirred at room temperature for 2 h in the dark. After the reaction was completed, 20 mL of Et_2O was added and frozen overnight. The next day, the precipitate was centrifuged and dried under vacuum to obtain the brown compound 7, which is abbreviated as FP-COOH.

Construction of magnetic targeting carrier system supported by ferroferric oxide:

37.6 mg of compound 7 was placed in a round bottom flask, followed by the addition of 0.5 ml of ethylene glycol and 9.5 ml of diethylene glycol, and the mixture was stirred to fully disperse them. Under stirring, 750 sodium acetate, 750 mg of sodium acrylate and 270 mg of $FeCl_3{\cdot}6H_2O$ were slowly added. Stirring is continued until a uniform yellow dispersion is formed. The dispersion was transferred to a hydrothermal reaction vessel, sealed, and heated at 200 °C for 10 h. After the reaction was completed, the reaction solution was centrifuged at 12000 r/min for 5 min, and the precipitate was repeatedly washed with ethanol, then collect the precipitate by centrifugation, and finally vacuum dry at 30 °C for 10 h to obtain a magnetic targeting system supported by ferroferric oxide (FP-IONP-COOH).

Preparation of FP-IONP-COOH/DOX:

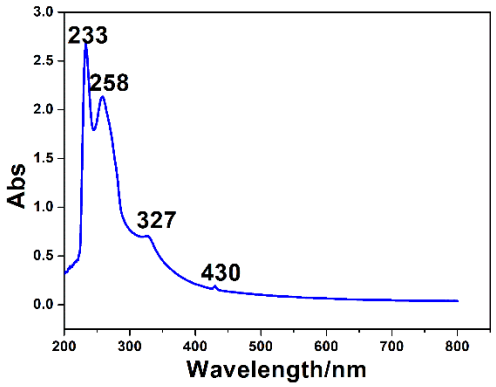
Weigh out the prescribed amount of FP-IONP-COOH and then fully dissolve it in the appropriate amount of distilled water. According to the mass ratio (DOX:FP-IONP-COOH = 2:1), add the appropriate amount of DOX, and under the protection of nitrogen, react in the dark for 48 hours. After the reaction, remove the free DOX on the surface by centrifugation.

3. RESULTS AND DISCUSSION

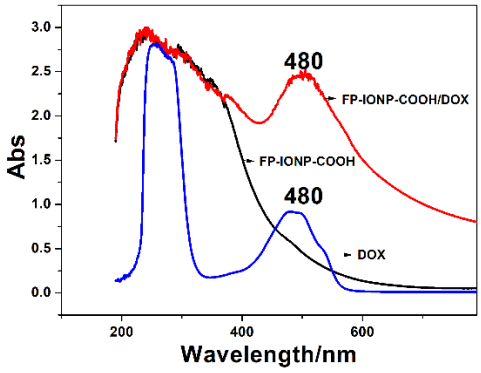
We mainly characterized the compounds 1, compound 2, compound 3, compound 6, compound 7, FP-IONP-COOH and FP-IONP-COOH/DOX, which used 1H-NMR, UV-Vis, FT-IR and MALDI-TOF-MS.

3.1. UV-Vis

DCM and distilled water were used as solvents to characterize the deprotected compound 7 (FP-COOH) and FP-IONP-COOH/DOX, respectively. The measurement wavelength was 200-800 nm, and the UV spectrum was recorded as shown in Fig. 2.



a



b

Fig. 2. a – UV-Vis of FP-COOH; b – UV of DOX, FP-IONP-COOH, FP-IONP-COOH/DOX

After removing the OtBu protecting group (Fig. 2 a), the peak becomes more fine and obvious. The absorption peak of C_{60} is 233 nm, 258 nm and 327 nm, which are in good agreement with the reported value [15], and the small peak at 430 nm is the characteristic absorption peak of the

fullerene pyrrole ring [16]. These indicate the successful synthesis of carboxy-terminated fullerene pyrrolidine. The characteristic absorption peak of DOX is recorded at 480 nm Fig. 2 b [17]. FP-IONP-COOH has no absorption peak at 480 nm, while FP-IONP-COOH/DOX has a strong absorption peak at 480 nm, indicating that FP-IONP-COOH successfully loaded doxorubicin.

3.2. FT-IR

The samples were prepared by potassium bromide tablet method and then determined by infrared spectroscopy. The FT-IR of FP-COOH are shown in Fig. 3. As can be seen from Fig. 3, the infrared spectrum of FP-COOH:vmax/(cm^{-1}):3431.78 is the stretching vibration absorption peak of -OH with only one absorption peak, 2922.48 and 2850.27 are the anti-symmetric and symmetric stretching vibration absorption peaks of C-H saturated alkanes. It is in the low frequency state. 723.13 is the stretching vibration peak of C-H, and the structural formulas $-CH_2-CH_2-CH_2-$, 576.49, 1189.15 and 1430.00 are the characteristic absorption peaks of fullerene C_{60}, and the absorption peak of 527.02 is the sign of addition to C_{60}. From the above analysis, FP-COOH was successfully synthesized, which are in good agreement with the reported value [14].

Hydrothermal reduction of iron ions to synthesize FP-IONP-COOH. During the reaction, a part of -COOH in FP-COOH is reduced due to the reduction of ethylene glycol and diethylene glycol, resulting in C=O (1684 cm^{-1}) and -OH (3431 cm^{-1}) vibration peak is weakened. These are similar to the points of view of the report [18].

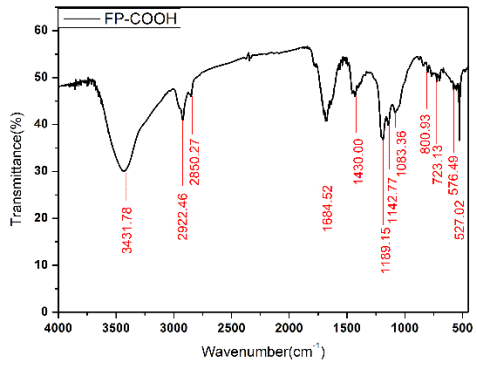


Fig. 3. IR spectrums of FP-COOH

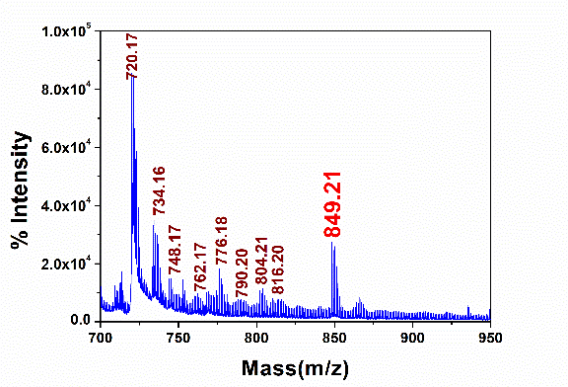
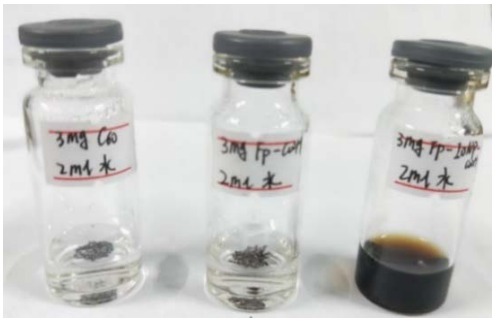


Fig. 4. Mass spectrum of FP-COOH

3.3. MALDI-TOF-MS

Matrix-assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOF) was used to determine its molecular weight, and
CHCA was used as matrix. Its mass spectra are shown in Fig. 4.

The molecular formula of FP-COOH is $C_{66}H_{11}O_2N$, and the theoretical molecular weight is 849 g/mol. In Fig. 4, besides the C_{60} reference peak, there is a strong molecular ion peak, m/z 849.21, which is consistent with the theoretical value of the molecular weight of the product. There are also [M-1], [M+1], [M+2] peaks.



a b c

Fig. 5. a – C_{60}; b – FP-COOH; c – FP-IONP-COOH dispersion in water

3.4. Study on the water solubility and stability of FP-IONP-COOH

Weigh C_{60}, FP-COOH and FP-IONP-COOH 3mg in 2 ml water, and let them stand for 2 weeks at room temperature to check their stability.

The solubility of C_{60}, FP-COOH, and FP-IONP-COOH in water is shown in Fig. 5. C_{60} is insoluble in water, while the fullerene derivative (FP-COOH) synthesized with reference to Bjelaković's method is slightly soluble in water this is because it contains carboxyl hydrophilic groups. Finally, from the above figure, we can also clearly see that the carboxylated fullerene pyrrolidine (FP-IONP-COOH) supported by ferric oxide has better water solubility and is almost completely soluble in water. Compared with the compound FP-COOH, the hydrophobicity of fullerene is greatly improved, and the bioavailability of fullerene is also greatly improved. And the FP-IONP-COOH was left standing for a period of time, but no settlement phenomenon was found, indicating that the FP-IONP-COOH also has good stability.

3.5. Magnetic properties of FP-IONP-COOH nanosuspensions

5 mg of FP-IOP-COOH was weighed and dissolved in 1 ml of water. Several magnets were placed beside the FP-IONP-COOH nanosuspension to observe the behavior of FP-IONP-COOH under magnetic field.

The paramagnetic behavior of FP-IONP-COOH is shown in Fig. 6. When a magnetic field is present, the FP-IONP-COOH particles move quickly toward the magnetic field, indicating that FP-IONP-COOH has good magnetic

properties. This result is consistent with Jinjin Shi's [18] research on the magnetic properties of IONP shows similar performance, indicating that IONP is an important means to improve the magnetic targeting ability of tumor drugs. Future research on the magnetic targeting properties of fullerenes can move closer to the direction of loading magnetic substances such as IONP, enabling new achievements in the research of tumor drugs.

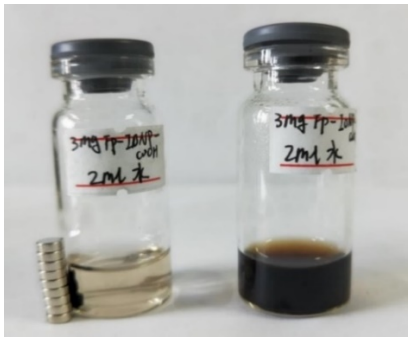


Fig. 6. Magnetic properties of FP-IONP-COOH

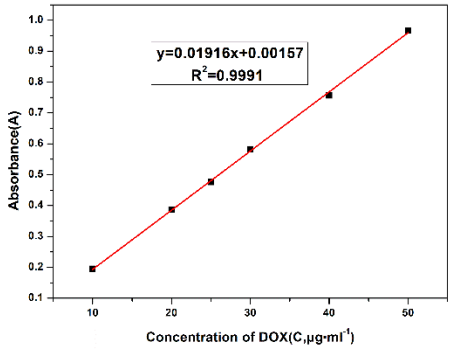


Fig. 7. Standard curve of DOX

3.6. Establishment of the standard curve

Weigh accurately DOX 10 mg, 0.5 % SDS to dissolve, volume the volumetric flask to 10 ml, shake well, and prepare a 1mg/ml stock solution. The stock solution was diluted to a standard solution having a concentration of 10, 20, 25, 30, 40, 50 μg/ml, respectively. The absorbance of DOX was measured at 479 nm using an ultraviolet spectrophotometer, and the data was recorded. Linear regression was performed with the DOX concentration (C) as the abscissa and the absorbance (A) as the ordinate.

The standard curve of DOX is shown in Fig. 7. The concentration range is 10 – 50 μg/ml, the linear regression equation is $y = 0.01916x + 0.00157$, and the correlation coefficient is $r = 0.9991$. The linear relationship is good.

3.7. Determination of FP-IONP-COOH/DOX saturated drug loading rate

To obtain the saturated drug loading rate of FP-IONP-COOH for DOX, select the mass ratio of different FP-IONP-COOH to DOX (1:0.5, 1:1, 1:2, 1:2.5, 1:3 and 1:4) was selected and the effect of different feed ratios on drug loading rate was investigated.

It can be seen from the drug load diagram in Fig. 8 above that more DOX is adsorbed onto FP-IONP-COOH as the DOX input increases, but when FP-IONP-COOH:DOX = 1:2, the drug loading rate has not changed much. Considering that excessive input of DOX will cause a lot of waste, this experiment selects FP-IONP-COOH: DOX = 1:2 as the final preparation ratio. When the ratio of FP-IONP-COOH to DOX was 1:2, FP-IONP-COOH/DOX was prepared with a drug loading of 66.8 %. But the non-covalent drug loading of doxorubicin on polymer microspheres prepared by Peng Liu [19] only was 35 %. By comparing the two, it can be concluded that the loading efficiency of doxorubicin is better with carboxylated fullerene pyrrolidine supported by ferric oxide. This is because that a lot of sp^2-carbons were found in C_{60} molecules, and DOX has delocalized π bonds, and this makes it easy to load onto C_{60} through π–π stacking. So FP-IONP-COOH can be used as a good carrier for doxorubicin.

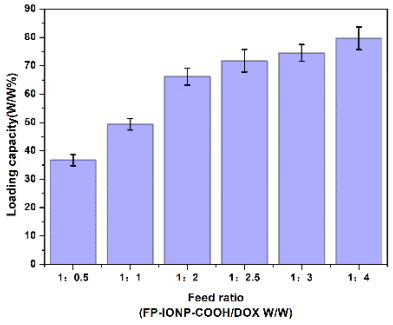


Fig. 8. Effect of different feed ratios on FP-IONP-COOH/DOX drug loading rate

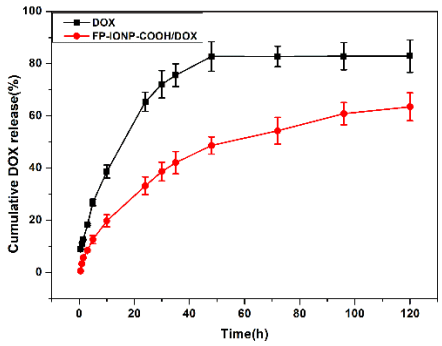


Fig. 9. Release of DOX from FP-IONP-COOH/DOX (n = 3)

3.8. In vitro drug release study of targeted preparation FP-IONP-COOH /DOX

Fullerenes act as versatile delivery systems with a wide range of biomedical applications, enabling anticancer drugs to be released from fullerenes in a controlled, targeted manner [20]. Based on this, the fullerene derivative FP-IONP-COOH was used as the drug carrier, and the doxorubicin was loaded in a non-covalent manner, then its targeting was investigated through in vitro release experiments.

Measure the 4 ml of FP-IONP-COOH /DOX and DOX injection (same DOX concentration) into a dialysis bag, and place the above dialysis bags into 50 ml of 0.5 % SDS dodecyl group. In a sodium sulfate solution, placed on a shaker at 37 °C, 100 r/min. Sampling 0.2 ml at 0.5, 1, 1.5, 3, 5, 10, 24, 30, 35, 48, 60, 72, 84, 96, 108, 120, 130, 150 h, and supplementing 0.2 ml of SDS solution, A standard curve of DOX in a 0.5 % SDS solution, the concentration of DOX

was measured, and the cumulative release of DOX at each time of each sample was calculated.

Jović [21] combined non-covalently with fullerol and DOX. Its research results showed that the physical method of doxorubicin could indeed slow the release and reduce the side effects of doxorubicin. The carrier FP-IONP-COOH and DOX are physically loaded, and it is also hoped that the DOX can be slowly released. As can be seen from the results of the in vitro drug release characteristics of FP-IONP-COOH/DOX (Fig. 9), the release rate of DOX in the release medium SDS is very fast, and the cumulative release amount has reached 82 % by 48 hours. The cumulative release of FP-IONP-COOH/DOX to 48 h is only 48 %, which can be continuously released for more than 120 h. Compared with DOX, the release rate of FP-IONP-COOH/DOX is significantly slower.

Goodarzi [22] has expressed in his article that carboxylated fullerenes have antioxidant activity, antimicrobial, and as a target drug delivery system. And here we have studied the targeting of carboxylated fullerene FP-COOH as a drug carrier. The results of in vitro release indicate that this study is consistent with the expected results of Kazemzad, Jović and Goodarzi on the targeting of carboxylated fullerene as a drug carrier. The FP-IONP-COOH/DOX nano drug delivery system does have a significant slow-release effect on the release of DOX, and has great potential in cancer treatment.

4. CONCLUSIONS

In this paper, FP-COOH was synthesized by the method of Bjelaković [11] et al. In the future development, these fullerene derivatives can not only be used as carriers, such as loaded hematoporphyrin monomethyl ether for improving photodynamic efficacy, doxorubicin loaded for sustained release, but also have other application prospects. Such as in the cosmetics industry as an antioxidant, scavenging free radicals, anti-aging and other effects, and as a photosensitizer used in photodynamic therapy. These types of fullerene derivatives open up new application values for fullerenes and also lay a good foundation for the fuller development of fullerenes.

Compared with the carboxy-terminated fullerene pyrrolidine (FP-COOH) synthesized by Bjelaković et al., The carboxylated fullerene pyrrolidine (FP-IONP-COOH) supported by ferric oxide nanoparticles not only has better water solubility, but also has good magnetic properties, Thus, the fullerene magnetic targeting carrier system can be constructed, after non-covalent complexing with doxorubicin, can achieve targeted dosing of doxorubicin, which not only improves the efficiency of tumor treatment, but also reduces the side effects of doxorubicin. This study (FP-IONP-COOH) is similar to the magnetic fullerene tumor drugs studied by Jinjin Shi [18] et al. Both have magnetic targeting capabilities. The drug targeting research provides a new idea for the diagnosis and treatment of tumors. While compared with the two, the biggest difference is the intermediate. The intermediate (FP-COOH) in this study contains carboxyl groups and is more widely used. The advantage of this study was the use of carboxyl functional groups to improve the properties of fullerene. The surface charge of the carboxyl group changes based on environmental acidity. This key feature of carboxyl functional groups has enabled them to control drug release [23]. Carboxyl functional groups are also hydrophilic and reinforce the hydrophilicity of fullerene. These also prevent aggregation of fullerene into the bloodstream. These groups increase hydrophilicity by increasing hydrogen bonds. Thus, this study showed that using these functional groups can improve the properties of nanotubes for drug release. As a recommendation for further studies, fullerene can be used as drug carriers along with other compounds, such as polymers. It is necessary to take further steps to improve the properties of carbon fullerene so that they can be used on a commercial scale.

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