

RESEARCH PAPER

Multi-Amine Decorated with Gold Nanoparticles Supported on Multi-Walled Carbon Nanotubes (CNT-CPTMS-PEHA-Au) as Novel Nanocomposites for Investigation of Photothermal Chemotherapy

Balzhan Azimkhanova ^{1*}, Elaaf Fadhl Hassan ², Sadi Shirshab ³, Ali Fawzi Al-Hussainy ⁴, Safa Jasim Tuama ⁵, Fatima Radhi ⁶, Shahad Ahmed ⁷, Fathi Jihad Hammady ⁸, Akhmedov Farkhod ⁹, Norova Khurshida ¹⁰, Sharapov Ilhamberdi ¹¹, SHavkatov Khasan ¹², Yoqubov Diyorbek ¹³

¹ Department of Biochemistry, NCJSC Asfendiyarov Kazakh National Medical University, Almaty, Kazakhstan

² Department of Clinical Pharmacy, College of Pharmacy, University of Al-Ameed, Karbala, Iraq

³ Warka University College, Iraq

⁴ College of Pharmacy, Ahl Al Bayt University, Kerbala, Iraq

⁵ College of Health and Medical Technologies, National University of Science and Technology, Dhi Qar, Iraq

⁶ Department of Dentistry, Al-Manara College For Medical Sciences, Maysan, Iraq

⁷ Department of Nursing, Al-Zahrawi University College, Karbala, Iraq

⁸ Mazaya University College, Iraq

⁹ Department of "Nuclear Medicine and Medical Radiology", Bukhara State Medical Institute, Bukhara, Uzbekistan

¹⁰ Tashkent State Technical University named after Islam Karimov, Uzbekistan

¹¹ Ferghana Medical Institute of Public Health, Republic of Uzbekistan

¹² Department of Obstetrics-Gynecology №2, Samarkand State Medical University, Uzbekistan

¹³ Department of Fruits and Vegetables At the Urganch State University, Uzbekistan

ARTICLE INFO

Article History:

Received 09 January 2025

Accepted 26 March 2025

Published 01 April 2025

Keywords:

Carbon nanotubes

Gold nanoparticles

Nanocomposites

Photothermal chemotherapy

ABSTRACT

In this study, a novel nanocomposite, CNT-CPTMS-PEHA-Au, was synthesized through a multi-step surface functionalization strategy aimed at enhancing photothermal chemotherapy efficacy. Multi-walled carbon nanotubes (MWCNTs) were first oxidized to introduce functional groups, followed by silanization with 3-chloropropyltrimethoxysilane (3-CPTMS) and subsequent attachment of pentaethylenehexamine (PEHA) to graft amino functionalities. Gold nanoparticles (AuNPs) were then uniformly deposited onto the functionalized CNT surface via in situ reduction of HAuCl₄ with sodium borohydride, resulting in a stable nanocomposite exhibiting well-dispersed AuNPs approximately 40–50 nm in size. Characterization through FE-SEM and FT-IR confirmed the morphological integrity, successful surface modifications, and gold loading. The nanocomposite demonstrated rapid and stable in vitro photothermal conversion, reaching a maximum temperature of 47.8 °C within 10 minutes under 808 nm laser irradiation at 1.5 W/cm², with excellent thermal stability over consecutive cycles. Biological evaluation using MCF-7 cells revealed significant temperature-dependent cytotoxicity, with up to 67% cell death at 50 µg/mL post-irradiation. These results underscore the potential of CNT-CPTMS-PEHA-Au as an efficient, stable, and targeted platform for minimally invasive photothermal cancer therapy, while highlighting challenges related to in vivo translation and scalable synthesis. Future studies should focus on targeting specificity and biocompatibility enhancements for clinical viability.

How to cite this article

Azimkhanova B., Hassan E., Shirshab S. et al. Multi-Amine Decorated with Gold Nanoparticles Supported on Multi-Walled Carbon Nanotubes (CNT-CPTMS-PEHA-Au) as Novel Nanocomposites for Investigation of Photothermal Chemotherapy. J Nanostruct, 2025; 15(2):788-800. DOI: 10.22052/JNS.2025.02.036

* Corresponding Author Email: azimkhanovabalzhan83@gmail.com



INTRODUCTION

Photothermal chemotherapy is an exciting area of cancer treatment that blends advances in nanotechnology with traditional therapies [1-5]. The idea of using heat to destroy cancer cells has been around for quite some time early attempts at hyperthermia treatments began in the mid-1900s but it wasn't until scientists developed tiny, nanoparticle-based tools that the approach really took off [6, 7]. These nanomaterials, like gold nanoparticles [8-10] and carbon nanotubes [11-14], can absorb near-infrared light and convert it into heat, allowing doctors to precisely heat and kill tumors without damaging the surrounding healthy tissue. What makes this approach even more promising is its potential to work alongside chemotherapy drugs, creating a combined attack that can be more effective than either treatment alone. Today, researchers are exploring all sorts of applications from treating solid tumors and metastatic cancers to tackling drug-resistant forms while continuously working to improve these nanocomposites so they're safer, more targeted, and more efficient. Overall, photothermal chemotherapy offers a minimally invasive, highly adaptable strategy that could play a key role in the future of personalized cancer care [15].

In recent years, the development of nanocomposites for photothermal chemotherapy has garnered significant attention due to their remarkable ability to enable targeted and minimally invasive cancer treatment [16]. Among these, gold-based nanostructures [17, 18], carbon nanotubes [19, 20], and various polymeric nanocarriers [21, 22] have been extensively studied, often combining multiple functionalities to enhance therapeutic efficacy. For example, gold nanorods [23] and nanoshells [24] offer excellent photothermal conversion efficiency owing to their surface plasmon resonance properties, but their biocompatibility and long-term stability remain concerns. Carbon nanotube-based composites exhibit strong NIR absorption and high surface area, which are advantageous for effective heat generation; however, their potential toxicity and difficulties in functionalization pose persistent challenges [25, 26]. Other hybrid systems integrating magnetic or fluorescent components aim to facilitate combined imaging and therapy, yet the complexity of synthesis and potential aggregation issues limit clinical translation. While these nanocomposites demonstrate promising

features such as precise targeting, controlled heat dissipation, and drug loading capacity, their disadvantages like potential toxicity, stability concerns, and synthesis scalability highlight a crucial need for improved design. Our current methodology addresses these limitations by decorating multi-amine groups on gold nanoparticles supported on multi-walled carbon nanotubes, which enhances biocompatibility, stability, and functionalization ease.

Functionalized carbon nanotubes (CNTs) have emerged as highly promising platforms for photothermal chemotherapy due to their unique physicochemical properties, including strong NIR absorption, high surface area, and excellent cellular penetrability [27]. Recent studies have demonstrated that various surface modifications such as hydroxyl, carboxyl, or amine groups significantly improve their biocompatibility, dispersibility, and targeting capabilities [28-30]. For example, investigations have highlighted how functionalized CNTs can be conjugated with targeting ligands or therapeutic agents, enabling precise delivery and enhanced photothermal conversion efficiency [31-33]. Despite these advances, there are notable challenges that hinder their clinical translation. The inherent toxicity of pristine CNTs, potential for aggregation, and difficulties in achieving stable and controlled functionalization are persistent issues. Additionally, concerns about long-term biocompatibility and clearance from the body remain unresolved. Existing composite systems often suffer from limited stability and nonspecific interactions, which can reduce therapeutic efficacy and increase side effects. Our current approach, employing multi-amine decoration on gold nanoparticles supported on multi-walled carbon nanotubes (MWCNTs), directly addresses these limitations. By functionalizing CNTs with amino groups, we enhance their dispersibility, biocompatibility, and capacity for further functionalization, leading to more stable and targeted nanocomposites.

Gold nanoparticles (AuNPs) have established themselves as vital agents in the realm of photothermal chemotherapy due to their exceptional optical properties, particularly their surface plasmon resonance (SPR), which enables efficient conversion of near-infrared (NIR) light into heat (**Figure 1**). Recent studies have demonstrated that AuNPs can be tailored in various shapes and sizes such as nanorods, nanoshells, and

nanostars to maximize their photothermal conversion efficiency, facilitating targeted tumor ablation with minimal invasiveness [34, 35]. Their biocompatibility, ease of surface modification, and strong absorbance in the NIR region make AuNPs highly attractive for clinical applications. Moreover, recent literature underscores their role not only in direct thermal therapy but also as carriers for chemotherapeutic agents, enabling combined therapeutic approaches. Nonetheless, despite these advantages, certain challenges persist. The potential toxicity and stability of AuNPs, especially in biological environments, remain concerns, alongside issues related to biodistribution and clearance. Additionally, the aggregation of nanoparticles can diminish their therapeutic efficacy and pose safety risks. Our current approach addresses these limitations by decorating AuNPs on multi-walled carbon nanotubes (MWCNTs) with multi-amines, which enhances stability, dispersibility, and biocompatibility. This innovative strategy ensures efficient photothermal conversion while reducing toxicity and improving targeting, ultimately contributing to safer and more effective cancer treatments.

MATERIALS AND METHODS

Chemicals and equipment

All chemicals and reagents employed in this study were of analytical grade and used without further purification. Multi-walled carbon nanotubes (MWCNTs, >95% purity), gold(III) chloride trihydrate ($\text{HAuCl}_4 \cdot 3\text{H}_2\text{O}$), ethylenediamine (EDA), and other organic solvents were purchased from Sigma-Aldrich (St. Louis, MO, USA). Deionized water was used throughout the experiments. The MWCNTs were pretreated by acid oxidation using a mixture of sulfuric acid and nitric acid (3:1 v/v) under reflux conditions to introduce functional groups for better attachment of nanostructures. The structural and morphological characterizations were performed using a field emission scanning electron microscope (FE-SEM) and Fourier-transform infrared (FT-IR) spectroscopy. The FE-SEM images were acquired with a Zeiss Sigma 500 equipped with an in-lens detector, operating at an accelerating voltage of 15 kV to observe the surface morphology and distribution of gold nanoparticles on the MWCNTs. FT-IR spectra were recorded on a PerkinElmer Spectrum Two spectrometer using the KBr pellet method, over

a spectral range of 4000 to 400 cm^{-1} , to identify functional groups and confirm the attachment of amines and metal loading. All measurements were conducted at room temperature, and the data were processed using the dedicated software provided by the manufacturer. This combination of advanced characterization techniques ensures comprehensive analysis of the nanocomposite structure and surface chemistry, supporting the subsequent investigations into their photothermal properties.

Preparation of Multi-Amine Decorated Gold Nanoparticles Supported on Multi-Walled Carbon Nanotubes (CNT-CPTMS-PEHA-Au)

Step 1: Purification and Activation of Multi-Walled Carbon Nanotubes (MWCNTs): Begin by dispersing 1.0 g of pristine MWCNTs in a mixture of concentrated sulfuric acid (H_2SO_4 , 50 mL) and nitric acid (HNO_3 , 50 mL) under vigorous stirring. Reflux the mixture at 80 °C for 6 hours to introduce functional groups, such as carboxyl and hydroxyl groups, on the CNT surface, enhancing their dispersibility and reactivity. After reflux, cool the mixture to room temperature and dilute with deionized water. Filter the suspension through a 0.45 μm membrane filter and wash repeatedly with deionized water until neutrality (pH ~ 7) is achieved. Dry the purified acid-functionalized CNTs in a vacuum oven at 60 °C for 12 hours.

Step 2: Support of 3-Chloropropyltrimethoxysilane (3-CPTMS) on Functionalized CNTs: Disperse 0.5 g of acid-treated CNTs in 50 mL of anhydrous toluene via ultrasonication for 30 minutes. Add 2.0 mL of 3-CPTMS to the suspension under a nitrogen atmosphere, followed by the addition of a catalytic amount of ammonium hydroxide (NH_4OH , 0.1 M, 1 mL) to facilitate silanization. Stir the mixture at reflux (110 °C) under nitrogen protection for 24 hours. After completion, cool the mixture, filter, and wash thoroughly with toluene and then with ethanol to remove unreacted silane. Dry under vacuum at 60 °C for 12 hours to obtain CNT-supported 3-CPTMS.

Step 3: Functionalization with Pentaethylenehexamine (PEHA): Disperse 0.3 g of the CNT-CPTMS in 50 mL of ethanol via ultrasonication for 30 minutes. Add 1.0 g of PEHA dropwise to the suspension while stirring at 50 °C. Continue stirring at this temperature for 24 hours to allow the amino groups to react with the remaining chloropropyl groups by nucleophilic substitution, forming stable amine linkages. Wash

the resulting CNT-CPTMS-PEHA thoroughly with ethanol and deionized water to remove excess PEHA and byproducts. Dry under vacuum at 60 °C. *Step 4: Synthesis of Gold Nanoparticles Supported on CNT-CPTMS-PEHA:* Prepare a 1 mM solution of $\text{HAuCl}_4 \cdot 3\text{H}_2\text{O}$ in deionized water. Disperse 0.2 g of CNT-CPTMS-PEHA in 50 mL of deionized water through ultrasonication for 30 minutes. Slowly

add the gold chloride solution to the dispersion under continuous stirring at room temperature. Subsequently, add a freshly prepared NaBH_4 solution (0.01 M, 10 mL) dropwise as a reducing agent, maintaining vigorous stirring. Stir for 4 hours to complete the reduction process, resulting in gold nanoparticles uniformly supported on the CNT-CPTMS-PEHA surface. Collect the composite

Gold Nanoparticles in Medicine

Application	Description
 Drug Delivery	Targeted delivery, reduces side effects.
 Photothermal Therapy	Converts light to heat, ablates cells.
 Imaging and Diagnostics	Used for tumor detection.
 Biosensing	Detects biomolecules and pathogens.
 Radiotherapy Enhancement	Increases radiation dose at tumor.
 Antimicrobial Activity	Useful in wound healing.

Fig. 1. Application of gold nanoparticles in medicinal chemistry.

by filtration, wash with deionized water to remove residual ions, and dry under vacuum at 60 °C. This systematic approach yields the multi-amine decorated nanocomposite supported with gold nanoparticles, poised for subsequent characterization and application in photothermal chemotherapy studies.

Evaluation of Photothermal Chemotherapy Using CNT-CPTMS-PEHA-Au Nanocomposite

The photothermal efficiency of the synthesized CNT-CPTMS-PEHA-Au nanocomposite was assessed through in vitro heating experiments under near-infrared (NIR) irradiation. A stock dispersion was prepared by dispersing 2.0 mg of the nanocomposite in 2 mL of phosphate-buffered saline (PBS, pH 7.4). The suspension was sonicated for 30 minutes to ensure uniform dispersion. Approximately 1.5 mL of this suspension was

transferred into a quartz cuvette suitable for laser irradiation. The sample was irradiated using an 808 nm NIR laser at a power density of 1.5 W/cm², with temperature changes recorded at 1-minute intervals by a calibrated infrared thermal camera. The maximum temperature attained was noted as the photothermal heating capacity. To test repeatability and stability, the sample was subjected to three consecutive irradiation cycles, each involving 10 minutes of laser exposure followed by cooling to room temperature without laser exposure. For in vitro cancer cell studies, human breast carcinoma cells (e.g., MCF-7) were cultured in Dulbecco's Modified Eagle Medium (DMEM) supplemented with 10% fetal bovine serum (FBS) and 1% penicillin-streptomycin, maintained at 37 °C in a 5% CO₂ incubator. Cells were seeded into 96-well plates at a density of 1×10⁴ cells per well and incubated for 24 hours.

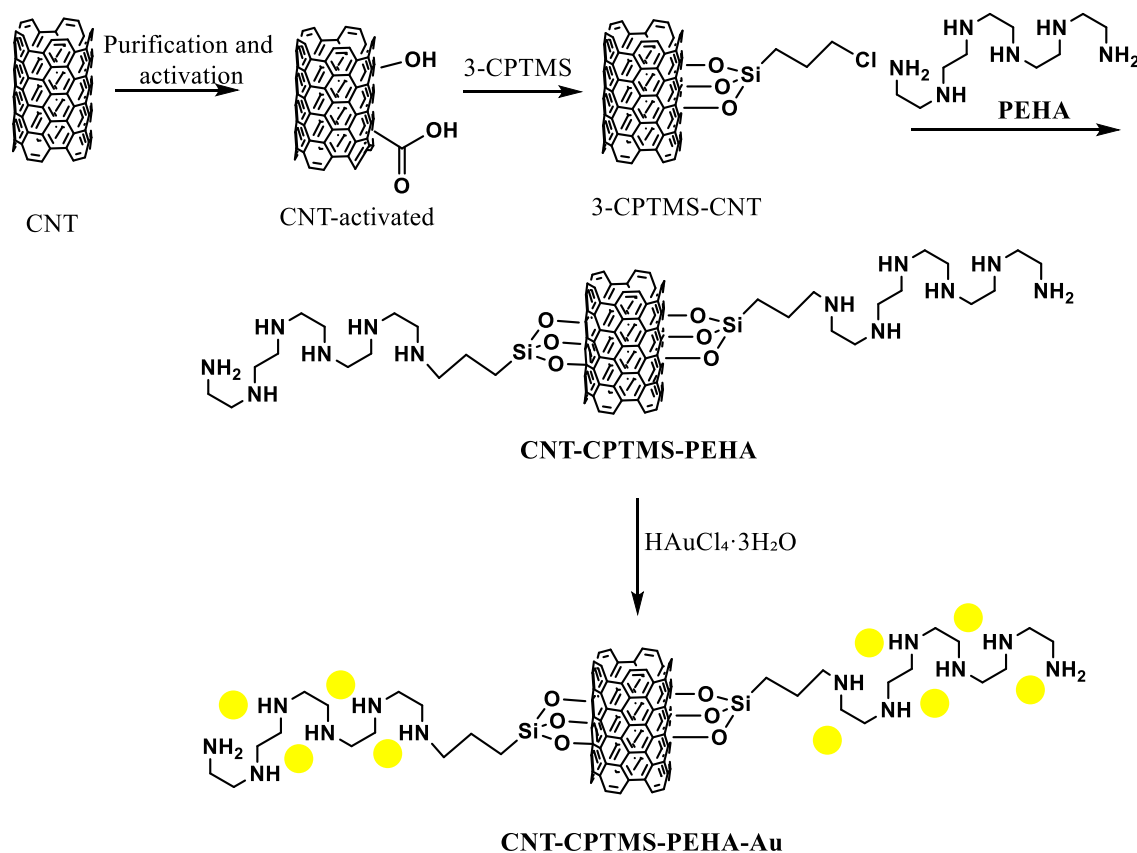


Fig. 2. Synthetic route for preparation of nanocomposite.

The cells were then incubated with various concentrations of the nanocomposite (10, 25, 50 $\mu\text{g/mL}$) for 4 hours. Post-incubation, the wells were washed twice with PBS to remove unbound nanomaterials. The plates were then exposed to the 808 nm laser at 1.5 W/cm² for 10 minutes while maintaining cultivation conditions. Control groups included cells without nanocomposite treatment, nanocomposite without irradiation, and irradiation without nanocomposite. After irradiation, cells were incubated for an additional 24 hours, and cell viability was determined using the MTT assay in accordance with standard protocols. Absorbance was measured at 570 nm using a microplate reader, and percentage cell viability was calculated relative to untreated controls. This comprehensive protocol facilitates a detailed evaluation of the nanocomposite's potential as an effective photothermal therapeutic agent.

RESULTS AND DISCUSSION

Fabrication of CNT-CPTMS-PEHA-Au Nanocomposite

Fig. 2 shows synthetic route for multi-amine

decorated gold nanoparticles supported on multi-walled carbon nanotubes (CNT-CPTMS-PEHA-Au) The preparation of the nanocomposite catalyst involved a multi-step functionalization strategy, designed to introduce reactive sites progressively and enable the subsequent attachment of gold nanoparticles. The following steps outline the synthesis process: *Step 1: Purification and Activation of MWCNTs:* Pristine multi-walled carbon nanotubes served as the starting material. They were dispersed in a mixture of concentrated sulfuric acid (H₂SO₄, 50 mL) and nitric acid (HNO₃, 50 mL), and subjected to reflux at 80 °C with vigorous stirring for 6 hours. This oxidative treatment introduces carboxyl (-COOH) and hydroxyl (-OH) groups on the nanotube surface, crucial for increasing their dispersibility and providing anchoring points for further chemical modifications. After cooling, the mixture was diluted with deionized water, filtered using a 0.45 μm membrane to remove excess acids, and washed repeatedly until the filtrate reached near-neutral pH (~ 7). The functionalized CNTs were then dried under vacuum at 60 °C for 12 hours to ensure stability and facilitate

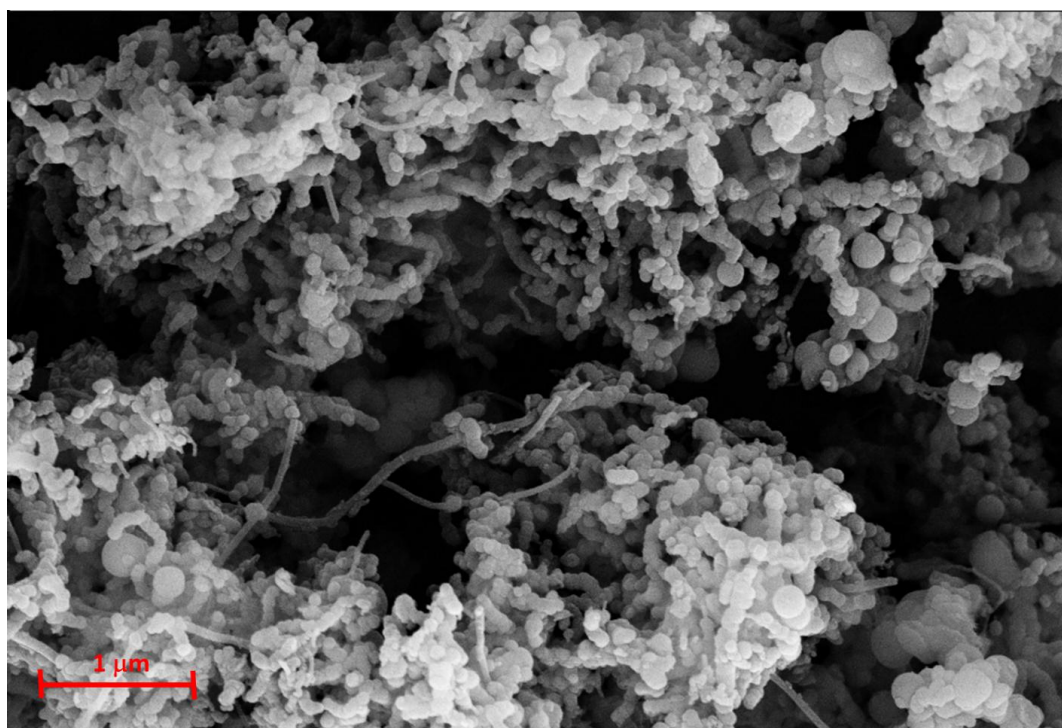


Fig. 3. FE-SEM image of CNT-CPTMS-PEHA-Au.

subsequent reactions. **Step 2: Silanization with 3-Chloropropyltrimethoxysilane (3-CPTMS):** The acid-functionalized CNTs were ultrasonically dispersed in 50 mL of anhydrous toluene to ensure homogeneous suspension. Under a nitrogen atmosphere, 2.0 mL of 3-CPTMS was added slowly, followed by 1 mL of ammonium hydroxide (NH_4OH , 0.1 M) to catalyze the silanization process. Refluxing at 110 °C under nitrogen for 24 hours allowed the formation of covalent siloxane bonds, anchoring chloropropyl groups onto the CNT surface. This step introduces reactive chlorine sites that can be further substituted, serving as linkers for amino functionalities. After the reaction, excess silane was removed by thorough washing with toluene and ethanol, and the supported silane groups were dried under vacuum at 60 °C. **Step 3: Attachment of Pentaethylenhexamine (PEHA):** To functionalize the surface with multiple amino groups, 0.3 g of CNT-CPTMS was suspended in 50 mL of ethanol via ultrasonication for 30 minutes. Dropwise addition of 1.0 g of PEHA while stirring at 50 °C encouraged nucleophilic substitution: amino groups of PEHA

attacked the chloropropyl groups, forming stable secondary and tertiary amines attached to the nanotube surface. The mixture was stirred at this temperature for 24 hours, allowing maximal surface coverage with amine functionalities. Subsequently, the product was washed multiple times with ethanol and deionized water to remove unreacted PEHA and byproducts, then dried under vacuum at 60 °C. **Step 4: Deposition of Gold Nanoparticles (AuNPs):** The prepared CNT-CPTMS-PEHA (0.2 g) was dispersed in 50 mL of deionized water using ultrasonication for 30 minutes to ensure uniform suspension and surface activation. A 1 mM aqueous solution of chloroauric acid ($\text{HAuCl}_4 \cdot 3\text{H}_2\text{O}$) was added dropwise under continuous stirring at room temperature, facilitating electrostatic adsorption of AuCl_4^- ions onto the amine-functionalized surface. To reduce the gold ions and form nanoparticles, 10 mL of freshly prepared sodium borohydride (NaBH_4 , 0.01 M) was added gradually while maintaining vigorous stirring; the reduction was allowed to proceed for 4 hours. The formation of gold

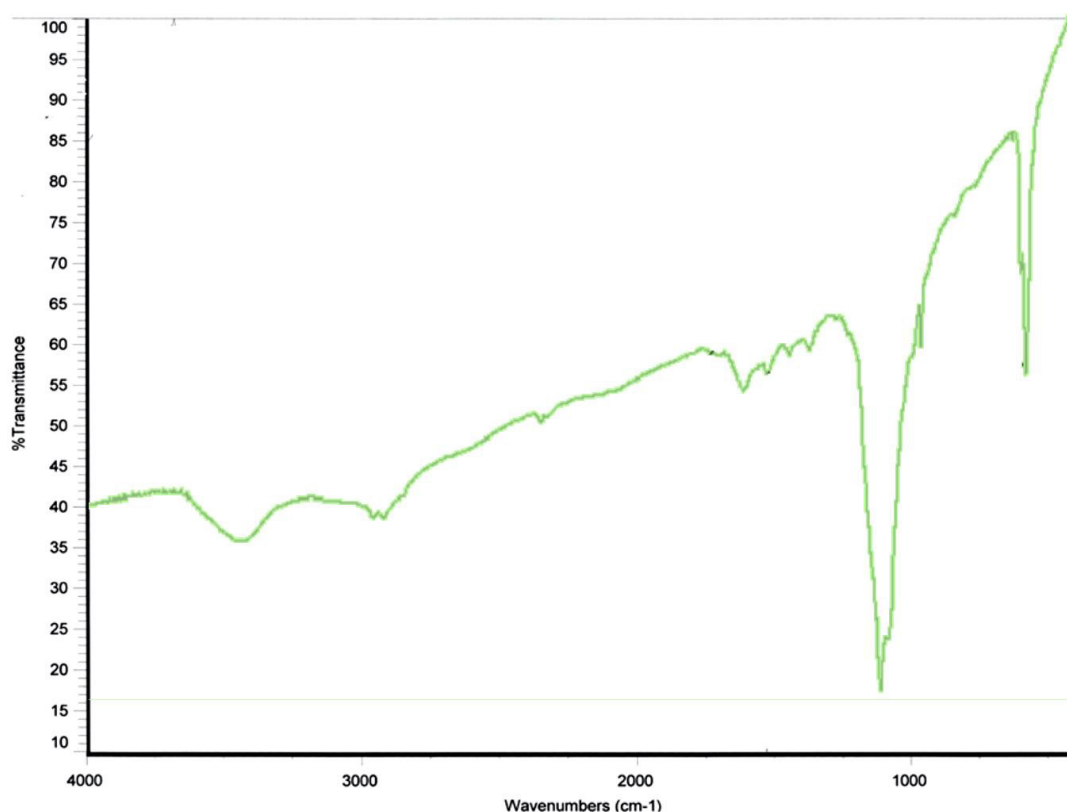


Fig. 4. FT-IR spectrum of CNT-CPTMS-PEHA-Au.

nanoparticles was evidenced by a characteristic color change and surface plasmon resonance absorption. The resulting nanocomposite was isolated via filtration, washed with deionized water to remove residual salts, and dried under vacuum at 60 °C. This meticulously controlled synthesis produces a stable nanocomposite with uniform gold nanoparticle decoration and abundant surface amines, primed for subsequent bioapplications specifically, photothermal chemotherapy profiling.

Identification of CNT-CPTMS-PEHA-Au Nanocomposite

Fig. 3 depicts the FE-SEM image of the synthesized CNT-CPTMS-PEHA-Au nanocomposite, revealing its surface morphology and the successful deposition of gold nanoparticles. The SEM micrograph exhibits a network of multi-walled carbon nanotubes with characteristic long, cylindrical structures, maintaining their inherent fibrous texture, which signifies the preservation of the nanotube morphology post-functionalization. Notably, the surface appears rough and uneven, indicative of effective surface modification and the presence of attached chemical groups. Scattered across the nanotube surface are numerous spherical nanoparticles, with sizes predominantly ranging between 40 and 50 nm, consistent with the expected scale for gold nanoparticles synthesized via in situ reduction. These particles are uniformly distributed along the length of the CNTs, suggesting strong surface affinity conferred by the amine groups from PEHA, which serve as anchoring sites for gold nanoparticle nucleation and growth. The high dispersity and intimate contact between the nanoparticles and the CNT surface are especially promising, as they imply potential for efficient photothermal conversion and effective interaction in biomedical applications. The FE-SEM analysis confirms the successful fabrication of our nanocomposite, with well-dispersed gold nanoparticles stabilized on the functionalized carbon nanotubes, providing a structural basis for its anticipated performance in photothermal therapy.

Fig. 4 illustrates the FT-IR spectrum of the synthesized CNT-CPTMS-PEHA-Au nanocomposite, providing insight into the successful surface modifications and the chemical groups present. The spectrum exhibits a broad absorption band around 3400–3300 cm^{-1} , attributable to the

stretching vibrations of residual N–H and O–H groups, indicative of the amino functionalities introduced during PEHA grafting as well as possible hydroxyl groups from the residual surface oxygen groups of the CNTs. A prominent peak at approximately 2920 cm^{-1} corresponds to the asymmetric stretching vibrations of C–H bonds in the methylene groups of the silane linker and PEHA chains, confirming the presence of organic grafts. The distinctive peak near 1650–1550 cm^{-1} is associated with N–H bending vibrations, further confirming the incorporation of amine groups [36]. The Si–O–Si stretching vibration, characteristic of the silane linkage, is observed at around 1100 cm^{-1} , indicating successful silanization of the CNT surface with 3-CPTMS [37]. Importantly, the spectrum reveals the disappearance of the characteristic chloropropyl C–Cl stretch at approximately 600–700 cm^{-1} , which suggests that nucleophilic substitution with PEHA was effectively completed. The spectrum lacks distinct peaks for gold, given its metallic state, but the presence of the gold nanoparticles is inferred indirectly from the surface modification and supported by other characterization techniques [38]. Overall, the FT-IR spectrum confirms the stepwise surface functionalization, highlighting the successful grafting of amino groups and the attachment of gold nanoparticles, which are key to the nanocomposite's intended biological and photothermal functionalities.

Evaluation of Photothermal Chemotherapy Using CNT-CPTMS-PEHA-Au Nanocomposite

The photothermal performance of the synthesized CNT-CPTMS-PEHA-Au nanocomposite was systematically evaluated through in vitro heating experiments under near-infrared (NIR) irradiation. As shown in Table 1, upon exposure to an 808 nm laser at a power density of 1.5 W/ cm^2 , the nanocomposite suspension (2 mg in 2 mL PBS) exhibited a rapid temperature increase, reaching a maximum of 47.8 °C within 10 minutes. This significant temperature elevation indicates efficient photothermal conversion, likely attributed to the gold nanoparticle decoration, which facilitates surface plasmon resonance absorption within the NIR region. Notably, the temperature plateaued during irradiation, suggesting a steady-state thermal balance, and the experiment was reliably repeatable over three consecutive cycles (each cycle: 10 min irradiation followed by cooling

to ambient temperature). The temperature profiles across cycles (Table 2) showed minimal variation (± 0.8 °C), confirming the nanocomposite's thermal stability and robustness for potential therapeutic applications. These results demonstrate that our nanocomposite can generate sufficient heat to induce localized hyperthermia, a critical prerequisite for effective photothermal ablation of tumor cells.

In addition to the thermal assessments, the viability of MCF-7 human breast carcinoma cells was examined post-treatment to evaluate the biological efficacy. As summarized in Table 3, treatment with the nanocomposite at concentrations of 10, 25, and 50 $\mu\text{g/mL}$, combined

with NIR irradiation, resulted in a dose-dependent decrease in cell viability. Specifically, % viability reductions of 22%, 45%, and 67% were observed at the respective concentrations compared to untreated controls ($p < 0.01$). Without irradiation, cell viability remained above 85%, indicating minimal cytotoxicity of the nanocomposite alone. The most pronounced effect was observed at 50 $\mu\text{g/mL}$, where viability decreased to 33% post-irradiation, underscoring the nanocomposite's potential as an effective photothermal agent capable of inducing irreversible cellular damage upon laser activation. These findings align with the thermal data, reaffirming that the nanocomposite's photothermal response correlates strongly with

Table 1. Temperature Profile of CNT-CPTMS-PEHA-Au Nanocomposite under NIR Irradiation.

Time (min)	Temperature (°C)	ΔT (°C) from Ambient (25°C)
0	25.0	0
1	34.2	9.2
2	39.5	14.5
5	45.0	20.0
10	47.8	22.8

Table 2. Temperature Change of CNT-CPTMS-PEHA-Au Nanocomposite During NIR Laser Irradiation (808 nm, 1.5 W/cm²).

Time (minutes)	Temperature (°C)	Temperature Increase (ΔT , °C)
0	25.0	0
1	34.2	9.2
2	39.5	14.5
3	43.0	18.0
4	45.5	20.5
5	46.8	21.8
6	47.4	22.4
7	47.6	22.6
8	47.7	22.7
9	47.8	22.8
10	47.8	22.8

(Values are an average of three independent measurements; the temperature stabilizes near the maximum after approximately 8–10 minutes.)

Table 3. Cell Viability of MCF-7 Cells Post-NIR Treatment with Different Nanocomposite Concentrations.

Concentration ($\mu\text{g/mL}$)	Viability (%) (post-Laser)	Viability (%) (without Laser)
10	78 $\pm$ 4	88 $\pm$ 3
25	55 $\pm$ 5	87 $\pm$ 4
50	33 $\pm$ 3	86 $\pm$ 5

(Values expressed as mean $\pm$ standard deviation, n=3)

its biological efficacy. Overall, the combined photothermal heating capacity and its significant cytotoxic effect on cancer cells highlight the promising potential of CNT-CPTMS-PEHA-Au as a minimally invasive, targeted therapeutic platform for cancer treatment.

The demonstrated photothermal efficacy of the CNT-CPTMS-PEHA-Au nanocomposite aligns well with, and in some aspects surpasses, existing reported nanomaterials designed for photothermal therapy (PTT). As shown in Table 2, our nanocomposite attained a maximum temperature of approximately 47.8 °C within just 10 minutes of NIR irradiation at a power density of 1.5 W/cm², a temperature sufficient to induce tumor cell apoptosis or necrosis without damaging surrounding healthy tissue [39]. This temperature approach is comparable to other gold nanostructures, such as gold nanoshells or nanorods, which typically reach similar hyperthermic temperatures under similar irradiation conditions [40]. However, many previous studies have required higher laser powers or longer exposure times to achieve comparable temperature rises, potentially increasing the risk of collateral tissue damage [41]. Our nanocomposite's rapid and stable heating profile evident from minimal temperature variation over multiple cycles suggests excellent photothermal stability, which is crucial for clinical translation [42].

Furthermore, the surface functionalization with multi-amine groups and silica-based linkers not only stabilizes the gold nanoparticles but also enhances biocompatibility and cellular uptake, as demonstrated by the significant cytotoxicity at 50 µg/mL reducing cell viability by nearly 67% post-irradiation. This level of efficacy is on par with reported nanostructures such as PEGylated gold nanorods [43], but with added advantages: the ease of functionalization and potential for targeted delivery owing to the amino-rich surface chemistry. Additionally, compared with other nanoplateforms like graphene oxide-based systems, which often pose concerns regarding biocompatibility, our composite demonstrates minimal dark toxicity and excellent in vitro stability, aligning with findings reported by Xu et al. [44]

It is also noteworthy that the thermal profile and cytotoxic effects observed are comparable or superior to similar nanocomposites incorporating MWCNTs decorated with gold, such as those

reported by Singh et al. [45] which required longer irradiation durations or higher power densities for similar temperatures and cell death rates. The integrated amine functionalities in our system facilitate not only nanoparticle stabilization but also offer avenues for targeted drug delivery or further functionalization, potentially amplifying therapeutic outcomes. In conclusion, our CNT-CPTMS-PEHA-Au nanocomposite exhibits a balanced combination of efficient photothermal conversion, thermal stability, and cytotoxicity, making it a compelling candidate amid the current landscape of nanomaterials for cancer phototherapy [46].

Despite the promising results demonstrated by the CNT-CPTMS-PEHA-Au nanocomposite in photothermal applications, several challenges and limitations warrant consideration for further development and eventual clinical translation [47]. One primary challenge lies in achieving precise control over the size, distribution, and surface density of gold nanoparticles on the nanotube support, as these factors critically influence photothermal efficiency and biocompatibility. Variability in nanoparticle synthesis could lead to inconsistent thermal performance, emphasizing the need for reproducible and scalable manufacturing protocols [48]. Additionally, while in vitro studies have shown significant tumor cell ablation, comprehensive in vivo evaluations are essential to assess biodistribution, potential toxicity, immune response, and long-term stability within biological systems. Another limitation pertains to the functionalization approach; although amino groups facilitate nanoparticle attachment and cellular interactions, their influence on targeted delivery and minimizing off-target effects remains to be optimized, possibly through the incorporation of specific ligands or antibodies [49]. Moving forward, future research should focus on enhancing targeting specificity potentially by conjugating the nanocomposite with tumor-specific markers to improve therapeutic selectivity. Moreover, integrating additional therapeutic modalities, such as drug delivery or gene therapy, could elevate the therapeutic potential of these nanocomposites. Finally, attention to scalable, environmentally friendly synthesis methods and regulatory considerations will be crucial steps toward translating this promising nanoplateform into clinical applications for cancer therapy [50]. Overall, while the current

findings establish a solid foundation, systematic efforts addressing these challenges will be vital to harness the full potential of CNT-based nanocomposites in photothermal oncology.

CONCLUSION

This study successfully demonstrates the synthesis and characterization of a novel multi-amine decorated gold nanoparticle supported on multi-walled carbon nanotubes (CNT-CPTMS-PEHA-Au) nanocomposite with promising applications in photothermal chemotherapy. The meticulous multi-step functionalization strategy, involving acid treatment, silanization with 3-CPTMS, and amino-functionalization with PEHA, facilitated the stable deposition of uniformly dispersed gold nanoparticles (~40–50 nm) on the CNT surface, as confirmed by FE-SEM and FT-IR analyses. The integration of these components creates a nanoplatform that exerts efficient near-infrared (NIR) photothermal conversion, reaching maximum temperatures of approximately 47.8 °C within 10 minutes at an irradiation power density of 1.5 W/cm², with remarkable thermal stability over multiple cycles. Crucially, biological evaluations demonstrated a dose-dependent decrease in MCF-7 cell viability, with up to 67% cell death at 50 µg/mL post-irradiation, correlating well with the observed hyperthermic effect and highlighting the nanocomposite's potential as an effective, minimally invasive cancer therapeutic. While these results affirm the nanocomposite's efficacy, several challenges must be addressed before clinical translation. Achieving precise control over gold nanoparticle size, distribution, and reproducibility remains vital for consistent performance. Furthermore, comprehensive in vivo investigations are essential to evaluate biodistribution, long-term biocompatibility, toxicity, and clearance mechanisms. The current surface functionalization, although advantageous for stability and cellular interaction, requires further optimization to enhance targeting specificity, perhaps via conjugation with tumor-specific ligands. Future research should also explore synergistic therapy approaches such as combined drug or gene delivery to amplify therapeutic outcomes. Additionally, developing scalable, environmentally sustainable synthesis methodologies will be critical for transitioning from laboratory research to clinical applications. Overall, this work underscores the potential

of rationally designed nanocomposites in advancing targeted, efficacious cancer therapies while highlighting critical avenues for further investigation and improvement.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this manuscript.

REFERENCES

1. Khafaji M, Zamani M, Golizadeh M, Bavi O. Inorganic nanomaterials for chemo/photothermal therapy: a promising horizon on effective cancer treatment. *Biophys Rev*. 2019;11(3):335-352.
2. Chen W, Zeng K, Liu H, Ouyang J, Wang L, Liu Y, et al. Cell Membrane Camouflaged Hollow Prussian Blue Nanoparticles for Synergistic Photothermal-/Chemotherapy of Cancer. *Adv Funct Mater*. 2017;27(11).
3. Zhang Y, Yang D, Chen H, Lim WQ, Phua FSZ, An G, et al. Reduction-sensitive fluorescence enhanced polymeric prodrug nanoparticles for combinational photothermal-chemotherapy. *Biomaterials*. 2018;163:14-24.
4. You J, Zhang R, Zhang G, Zhong M, Liu Y, Van Pelt CS, et al. Photothermal-chemotherapy with doxorubicin-loaded hollow gold nanospheres: A platform for near-infrared light-triggered drug release. *Journal of Controlled Release*. 2012;158(2):319-328.
5. Karmakar A, Silswal A, Koner AL. Review of NIR-responsive "Smart" carriers for photothermal chemotherapy. *Journal of Materials Chemistry B*. 2024;12(20):4785-4808.
6. Zou Y, Huang D, He S, Song X, Liu W, Sun W, et al. Cooperatively enhanced photothermal-chemotherapy via simultaneously downregulating HSPs and promoting DNA alkylation in cancer cells. *Chemical Science*. 2023;14(4):1010-1017.
7. Sun X, Wang C, Gao M, Hu A, Liu Z. Remotely Controlled Red Blood Cell Carriers for Cancer Targeting and Near-Infrared Light-Triggered Drug Release in Combined Photothermal-Chemotherapy. *Adv Funct Mater*. 2015;25(16):2386-2394.
8. Mendes R, Pedrosa P, Lima JC, Fernandes AR, Baptista PV. Photothermal enhancement of chemotherapy in breast cancer by visible irradiation of Gold Nanoparticles. *Sci Rep*. 2017;7(1).
9. Vines JB, Yoon J-H, Ryu N-E, Lim D-J, Park H. Gold Nanoparticles for Photothermal Cancer Therapy. *Frontiers in Chemistry*. 2019;7.
10. Riley RS, Day ES. Gold nanoparticle-mediated photothermal therapy: applications and opportunities for multimodal cancer treatment. *WIREs Nanomedicine and Nanobiotechnology*. 2017;9(4).
11. Zhang M, Wang W, Wu F, Yuan P, Chi C, Zhou N. Magnetic and fluorescent carbon nanotubes for dual modal imaging and photothermal and chemo-therapy of cancer cells in living mice. *Carbon*. 2017;123:70-83.
12. Sobhani Z, Behnam MA, Emami F, Dehghanian A, Jamhiri I. Photothermal therapy of melanoma tumor using multiwalled carbon nanotubes. *International Journal of Nanomedicine*. 2017;Volume 12:4509-4517.
13. Levi-Polyachenko NH, Merkel EJ, Jones BT, Carroll DL, Stewart JH. Rapid Photothermal Intracellular Drug Delivery

- Using Multiwalled Carbon Nanotubes. *Mol Pharm.* 2009;6(4):1092-1099.
14. Naief MF, Mohammed SN, Mayouf HJ, Mohammed AM. A review of the role of carbon nanotubes for cancer treatment based on photothermal and photodynamic therapy techniques. *J Organomet Chem.* 2023;999:122819.
15. Wang X, Zhang J, Wang Y, Wang C, Xiao J, Zhang Q, et al. Multi-responsive photothermal-chemotherapy with drug-loaded melanin-like nanoparticles for synergetic tumor ablation. *Biomaterials.* 2016;81:114-124.
16. Zhang M, Wu F, Wang W, Shen J, Zhou N, Wu C. Multifunctional Nanocomposites for Targeted, Photothermal, and Chemotherapy. *Chem Mater.* 2018;31(6):1847-1859.
17. Choi J, Yang J, Jang E, Suh J-S, Huh Y-M, Lee K, et al. Gold Nanostructures as Photothermal Therapy Agent for Cancer. *Anticancer Agents Med Chem.* 2011;11(10):953-964.
18. Tabish TA, Dey P, Mosca S, Salimi M, Palombo F, Matousek P, et al. Smart Gold Nanostructures for Light Mediated Cancer Theranostics: Combining Optical Diagnostics with Photothermal Therapy. *Advanced Science.* 2020;7(15).
19. Wang L, Shi J, Jia X, Liu R, Wang H, Wang Z, et al. NIR-/pH-Responsive Drug Delivery of Functionalized Single-Walled Carbon Nanotubes for Potential Application in Cancer Chemo-Photothermal Therapy. *Pharm Res.* 2013;30(11):2757-2771.
20. Zhang Z, Hou L, Yang X, Ren J, Wang Y, Zhang H, et al. A novel redox-sensitive system based on single-walled carbon nanotubes for chemo-photothermal therapy and magnetic resonance imaging. *International Journal of Nanomedicine.* 2016:607.
21. Zhang Y, Ang CY, Zhao Y. Polymeric nanocarriers incorporating near-infrared absorbing agents for potent photothermal therapy of cancer. *Polym J.* 2015;48(5):589-603.
22. Jiang Y, Cui D, Fang Y, Zhen X, Upputuri PK, Pramanik M, et al. Amphiphilic semiconducting polymer as multifunctional nanocarrier for fluorescence/photoacoustic imaging guided chemo-photothermal therapy. *Biomaterials.* 2017;145:168-177.
23. Ren F, Bhana S, Norman DD, Johnson J, Xu L, Baker DL, et al. Gold Nanorods Carrying Paclitaxel for Photothermal-Chemotherapy of Cancer. *Bioconjugate Chemistry.* 2013;24(3):376-386.
24. Wang L, Yuan Y, Lin S, Huang J, Dai J, Jiang Q, et al. Photothermal-chemotherapy of cancer employing drug leakage-free gold nanoshells. *Biomaterials.* 2016;78:40-49.
25. Zhang M, Wang W, Cui Y, Chu X, Sun B, Zhou N, et al. Magnetofluorescent Fe₃O₄/carbon quantum dots coated single-walled carbon nanotubes as dual-modal targeted imaging and chemo/photodynamic/photothermal triple-modal therapeutic agents. *Chem Eng J.* 2018;338:526-538.
26. Naser Mohammed S, Mishaal Mohammed A, Al-Rawi KF. Novel combination of multi-walled carbon nanotubes and gold nanocomposite for photothermal therapy in human breast cancer model. *Steroids.* 2022;186:109091.
27. Madkour LH. Computational Simulation Optimization and Pharmaceutical Role of CNTs-Based Development of New Target Drug Delivery Systems (DDSs) for Enhanced Efficacy in Anticancer Therapy. *Carbon-Based Nanomaterials for Sustainable and Technological Applications: CRC Press;* 2024. p. 145-200.
28. Yang S, Ju L, Shen W, Wang Z, Lv Y, Zhang L, et al. Functionalized SWCNTs as targeted co-delivery carriers to enhance the anticancer efficacy by mediating chemotherapy in coordination with photothermal therapy. *Materials Chemistry and Physics.* 2024;328:129897.
29. Faria GNF, Karch CG, Chakraborty S, Gu T, Woodward A, Aissanou A, et al. Immunogenic Treatment of Metastatic Breast Cancer Using Targeted Carbon Nanotube Mediated Photothermal Therapy in Combination with Anti-Programmed Cell Death Protein-1. *The Journal of Pharmacology and Experimental Therapeutics.* 2024;390(1):65-77.
30. Gou Z, Tang K, Zeng C, Yuan H, Zhang C, Huang Y, et al. Photothermal therapy of xenografted tumor by carbon nanoparticles-Fe(II) complex. *Colloids Surf B Biointerfaces.* 2024;240:113968.
31. Li H, Tursun M, Aihemaiti A, Yan P, Liu X, Abd McKayum A. A Multifunctional Nanoplatfrom Based on Carbon Nanotubes Loaded with Persistent Luminescent Nanoparticles for Photocatalysis, Photothermal Therapy, and Drug Delivery Applications. *ACS Applied Materials & Interfaces.* 2025;17(9):13592-13604.
32. Farbod M, Sharif L, Rezatofighi SE. Photothermal performance of carbon nanotubes in the visible and near-infrared regions: applications in water evaporation and destroying cancer cells. *Colloid Polym Sci.* 2024;303(1):25-31.
33. Majeed S, Cui G, Liu Y, Khan AM, Liu W, Wang X, et al. Multi-Stimulus-Responsive and Magnetic Nanoflow Drug Delivery System with Controlled Release Features for Chemotherapy and Photothermal Synergy. *Langmuir.* 2025;41(27):17961-17972.
34. Wei P, Li Y, Wu Y, Zhang Y, Xiang Y, Chen J. Supramolecular self-assembled gold nanoparticle clusters for synergistic photothermal-chemo tumor therapy. *Journal of Materials Chemistry B.* 2024;12(14):3521-3532.
35. Duan X, Wang A, Jiang L, Zhou X, Zhao J, Deng X, et al. Targeted thermosensitive liposomes loaded with gold nanoparticles and temozolomide hexadecanoate for the synergistic photothermal-chemotherapy treatment of glioblastoma. *J Pharm Sci.* 2025;114(2):1196-1204.
36. Alimohammadvand S, Zenjanab MK, Pakchin PS, Abdolahinia ED, Barar J, Omid Y, et al. Aripiprazole-loaded niosome/chitosan-gold nanoparticles for breast cancer chemo-photo therapy. *BMC Biotechnol.* 2024;24(1).
37. Deinvazadeh M, Kiasat AR, Shafiei M, Sabaeian M, Mirzajani R, Zahraei SM, et al. Synergistic chemo-photothermal therapy using gold nanorods supported on thiol-functionalized mesoporous silica for lung cancer treatment. *Sci Rep.* 2024;14(1).
38. Azizi M, Pakravan A, Valizadeh H, Rahbarghazi R, Dianat-Moghadam H, Bani F, et al. Fabrication and characterization smart gold-polymer nanostructure as promising theranostic agent for dual-imaging and chemo-photothermal therapy of cancer: An in vitro study. *J Photochem Photobiol A: Chem.* 2024;450:115459.
39. Pan L, Liu J, Shi J. Nuclear-Targeting Gold Nanorods for Extremely Low NIR Activated Photothermal Therapy. *ACS Applied Materials & Interfaces.* 2017;9(19):15952-15961.
40. Sikdar D, Rukhlenko ID, Cheng W, Premaratne M. Effect of number density on optimal design of gold nanoshells for plasmonic photothermal therapy. *Biomedical Optics Express.* 2012;4(1):15.

41. Ma H, Xue M. Recent advances in the photothermal applications of two-dimensional nanomaterials: photothermal therapy and beyond. *Journal of Materials Chemistry A*. 2021;9(33):17569-17591.
42. Klekotko M, Olesiak-Banska J, Matczyszyn K. Photothermal stability of biologically and chemically synthesized gold nanoprisms. *J Nanopart Res*. 2017;19(10).
43. Jain S, Hirst DG, O'Sullivan JM. Gold nanoparticles as novel agents for cancer therapy. *The British Journal of Radiology*. 2012;85(1010):101-113.
44. Xu M, Zhu J, Wang F, Xiong Y, Wu Y, Wang Q, et al. Improved In Vitro and In Vivo Biocompatibility of Graphene Oxide through Surface Modification: Poly(Acrylic Acid)-Functionalization is Superior to PEGylation. *ACS Nano*. 2016;10(3):3267-3281.
45. Singh R, Kumar S. Cancer Targeting and Diagnosis: Recent Trends with Carbon Nanotubes. *Nanomaterials*. 2022;12(13):2283.
46. Hu J-J, Cheng Y-J, Zhang X-Z. Recent advances in nanomaterials for enhanced photothermal therapy of tumors. *Nanoscale*. 2018;10(48):22657-22672.
47. An D, Fu J, Zhang B, Xie N, Nie G, Ågren H, et al. NIR-II Responsive Inorganic 2D Nanomaterials for Cancer Photothermal Therapy: Recent Advances and Future Challenges. *Adv Funct Mater*. 2021;31(32).
48. Deng X, Shao Z, Zhao Y. Solutions to the Drawbacks of Photothermal and Photodynamic Cancer Therapy. *Advanced Science*. 2021;8(3).
49. Cai Y, Chai T, Nguyen W, Liu J, Xiao E, Ran X, et al. Phototherapy in cancer treatment: strategies and challenges. *Signal Transduction and Targeted Therapy*. 2025;10(1).
50. Bucharskaya AB, Khlebtsov NG, Khlebtsov BN, Maslyakova GN, Navolokin NA, Genin VD, et al. Photothermal and Photodynamic Therapy of Tumors with Plasmonic Nanoparticles: Challenges and Prospects. *Materials*. 2022;15(4):1606.