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# A BRIEF STATEMENT: DEVELOPMENTS IN NANOCARRIER DELIVERY SYSTEMS USING GRAPHENE

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**Keywords:**

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**ABSTRACT:** One of the most important difficulties in contemporary medical biology is the creation of a cutting-edge, effective drug delivery system with a considerable improvement in its efficacy and increased therapeutic value. The advancement and development of innovative drug-delivery nanocarrier systems with accuracy and reduced side effects have been made possible by merging nanomaterial science with molecular and cellular biology. Over the past few years, the design and manufacture of nanocarriers using graphene oxide (GO) have grown quickly. Because of its amazing physicochemical features, GO has been widely exploited in efforts to create nanocarriers with high specificity, selectivity, biocompatibility, and minimal cytotoxicity. This review focuses on recent applications, synthesis, of GO-based nanocarriers, and advancements in enceinte drug delivery systems. Following a brief explanation of the principles and difficulties surrounding nanocarrier systems, we offer important illustrations of the transport of drugs and genes using GO. The assessment concludes with a few succinct observations on what is known currently and what the future may hold for nanocarrier delivery technologies.

**INTRODUCTION: A** Nanocarrier, a kind of submicron drug delivery system belonging to the nanoscale scope, can adjust the drug release rate, enhance the permeability of the biological membrane, change the drug distribution *in-vivo*, and improve its efficiency through encapsulation, absorption, and even covalent cross-linking [1]. In view of these advantages, Nanocarriers, including Nanoparticles, vesicles, Nano-sheets, and Nano-capsules have been intensively investigated in the biomedical field [2]. Graphene is a two-dimensional atomic crystal, and since its development, it has been applied in many novel ways in both research and industry.

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Graphene possesses unique properties and it has been used in many applications including sensors, batteries, fuel cells, super capacitors, transistors, components of high-strength machinery and display screens in mobile devices. In the past decade, the biomedical applications of graphene have attracted much interest. Graphene has been reported to have antibacterial, antiplatelet, and anticancer activities.

**Several Salient Features:** Graphene makes it a potential candidate for biological and biomedical applications. graphene has some following silent features which are given below.

- Synthesis
- Toxicity
- Biocompatibility
- Nano-scaled materials

- Promising Nanocarriers
- member of the Nanocarrier

**Structure of Graphene:** Strictly speaking, the term graphene refers to a single layer of graphite. More generally, it refers to fewer than ten layers of graphite.

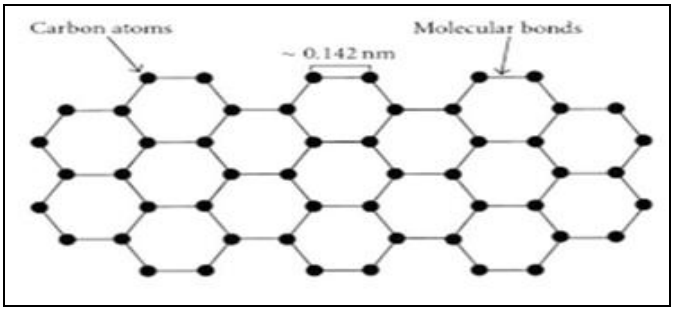


**FIG. 1: GRAPHENE STRUCTURE**

**Like:** carbon nanotubes, fullerene, graphite, and diamond

It is an allotrope of elemental carbon with sp2-hybridized carbon atoms, with partially filled p-orbitals above and below the plane of the sheet. The thinnest honeycomb lattice structure is composed of carbon atoms [3-4].

**Classification of Graphene:** Graphene materials can be classified in the following ways which are given below.

1. Based on their structure
2. Based on stacking arrangement
3. Based on edge type

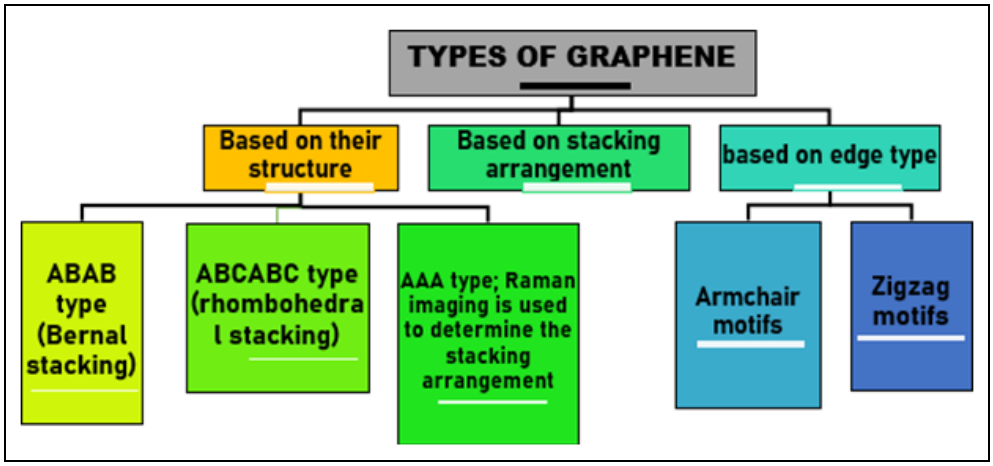


**FIG. 2: CLASSIFICATION OF GRAPHENE**

**Properties of Graphene:**


**FIG. 3: PROPERTIES OF GRAPHENE**

1. Graphene has acarbon–carbon bond length of 0.142 nm, 44 and it is the thinnest known material with good strength. The high strength of graphene facilitates its application in flexible electronics [5, 6].

2. The planar structure of graphene makes it a suitable factor for the high loading of different substances, such as bio-molecules and metals. On the other hand, GO can deliver small drug molecules (such as anticancer and antibacterial agents) and macromolecules, as well as its bipolar groups (hydrophilic and hydrophobic), allow it to carry both hydrophilic and hydrophobic substances.

3. Altogether, these excellent properties, their small size, and high bio-compatibility make GO a promising candidate for medical and biological applications [6].

4. The functionalization of GO with oxygenated groups such as carboxyl, increases its biocompatibility and solubility.

5. Some studies have shown the higher biocompatibility, safety and efficacy of drug loaded in GO-COOH rather than GO.

However, pure GO lacks a bioactive site to support cell growth, which restricts its application in the biomedical field.

Recently, researchers synthesized the carboxymethyl cellulose- modified graphene oxide (GO-CMC) complex as a drug carrier matrix, which further bonded to small molecule doxorubicin hydrochloride (DOX) by $\pi - \pi$ bond interaction and hydrogen bonding [7].

Nanocarrier exhibited a controllable drug release capacity without obvious cytotoxicity [16]. Another GO Nanocarrier for the diagnosis of renal dysfunction was.

**Synthesis of Graphene:** This review focuses on summarizing the synthesis of graphene using biological systems.

Generally, graphene synthesis is classified into two categories

1. Top-down
2. Bottom-up.

**Top-down:** Top-down approaches separate the stacked sheets by disrupting the van der Waals forces that hold the sheets together.

**Disadvantages:**

- Damaging of the sheets during the exfoliation process
- Re-agglomeration of the separated sheets, graphite, the precursor, is scarce.

**Bottom-up:** The bottom-up approach is simple but produces material with relatively more defects than the top-down approach.

Substrate-free method and carbonization.

Micromechanical exfoliation is a very simple and commonly used technique for investigating the fundamental properties of graphene.

**Disadvantages:**

- It is a slow and labor-intensive technique
- It’s not suited for commercial applications
- It requires a very high temperature

So, this technique may not be ideal for the synthesis of graphene for biomedical applications because, in addition, the surfactants are very difficult to remove.

The availability of a biocompatible, nontoxic surfactant for use in this technique will greatly aid in developing better methodologies for synthesis.

**Synthesis of Graphene using Biomolecules:** Recently, the usage of biological materials for the synthesis of nanoparticles (NPs) has garnered much attention due to their low energy requirements, environmentally friendly nature, dependability, cost-effectiveness, scalability, stability, and availability of the required solutions at high densities, compared with chemical synthesis.

Similar approaches have been exploited for synthesizing graphene using proteins, peptides, bacteria, fungi, plants, and others.

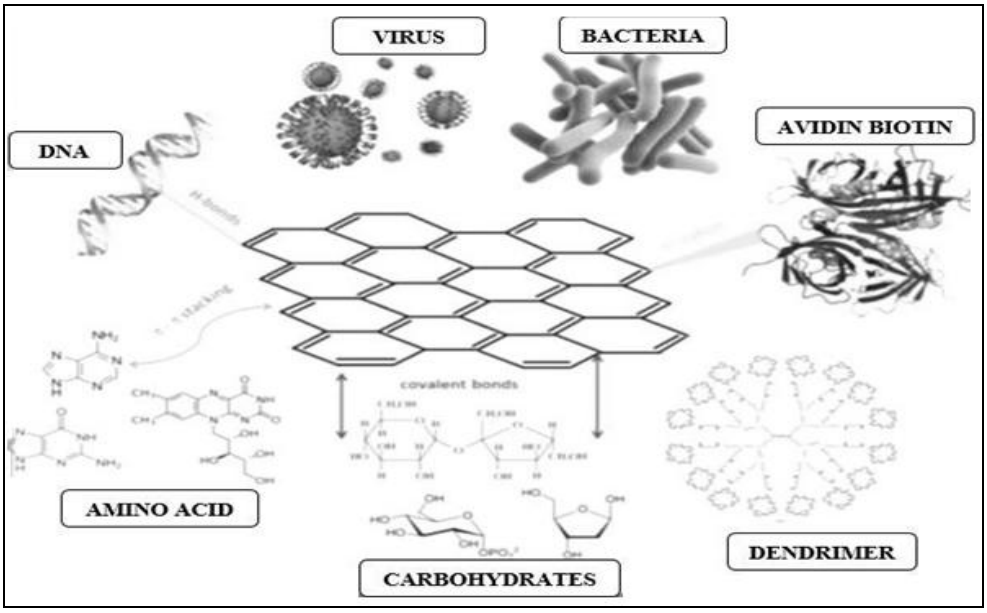


**FIG. 4: GRAPHENE SYNTHESIS BIO-FUNCTIONALIZATION THROUGH PHYSICAL ADSORPTION, CHEMICAL BONDING, AND BIO-CONJUGATION**

**For Example:**

1. Initially proposed a "green" reduction of GO via bacterial respiration of *Shewanella* cells, *Escherichia coli*, *Escherichia fergusonii*, and *Pseudomonas*. microbially reduced graphene exhibits excellent electrochemical properties.
2. Plant extracts have received much attention for the reduction of GO as a suitable alternative to chemical procedures and physical methods. leaf extracts of *Colocasia esculenta* and *Mesua ferrea*, *Ginkgo biloba* extract, leaf extracts of cherry, *Magnolia*, *Platanus*, persimmon, pine, maple, and *Ginkgo* Extracts from plants may act as both reducing and capping agents in NP synthesis [8, 9, 10, 11].

**Characterization of Graphene or Graphene Material:** Graphene and other nanocarrier material-related products can be studied by the following techniques for better therapeutic result, reduced toxicity, and enhanced knowledge of nanocarrier, which are given below.

1. Raman spectroscopy
2. FTIR (Fourier transform infrared spectroscopy)
3. XRD (X-ray diffraction)
4. TGA (Thermo-gravimetric analysis)
5. XRF (X-ray Fluoresence)
6. surface area & pore volume
7. TEM (Transmission electron microscopy)
8. Effect of pH on graphene by High-Performance Liquid Chromatography (HPLC)
9. drug loading assay 10.UV-Visible spectroscopy
11. Zeta potential by Dynamic Light Scattering (DLS)
12. AFM (Atomic force microscopy)
13. Binding Kinetics. UV−Vis Kinetic Measurements
14. Fluorescence Lifetime-Based FLIM-FRET
15. Microscale Thermophoresis (MST)
16. Statistical Analysis

**Raman Spectroscopy:** Raman Spectroscopy is a simple, fast, and non-destructive method that uses monochromatic excitation laser to verify the structure of the material. In Raman Spectroscopy of graphene, there are three response peaks of interest. The in-plane vibrations of the conjugated $\pi$-bonds exhibit characteristic Raman spectra: 1327, 1584, and 2643 $cm^{-1}$ for D, G, and 2D, respectively [11, 12, 13, 14, 15].

**Fourier-Transform Infrared Spectroscopy (FTIR):** FTIR Spectroscopy is widely employed to

detect functional groups and to characterize graphene nanocomposites, however, is not the most recommended technique to detect the presence of metallic species on the surface of graphene or graphene oxide [11, 16, 17, 18, 19].

**X-ray Diffraction:** Although XRD is not a perfect tool for identifying single-layer graphene, the analysis may help to differentiate between graphite and graphene samples. Pristine graphite exhibits a basal reflection (002) peak at $2\theta = 26.6°$ (d spacing = 0.335 nm) in the XRD pattern [11, 19, 20, 21, 22].

**X-ray Fluorescence (XRF):** To calculate the relative oxygen content, the intensity values from carbon and oxygen of blank filters were subtracted from the samples. To compensate for variations in the amount of PGNS and GO placed on the filters, the oxygen content of all filters was normalized based on the measured carbon content. The relative oxygen content was then calculated by dividing the resulting relative oxygen content of GO by PGNS.

**Thermogravimetric Analysis (TGA):** Thermogravimetric Analysis (TGA) is a useful tool for determining functionalized graphene or GO weight loss under aggressive temperatures. Many authors have applied it to trace changes in the structure of graphitic materials before and after the functionalization of graphene sheets [11, 20, 22, 23, 24].

**Specific Surface Area and Pores Volume:** BET Specific Surface Area and Pore Distribution is a technique widely used to estimate the maximum adsorption capacity of some materials. For adsorptive and catalytic processes, the surface area and pore size distribution considerably influence their efficiency, capacity, and yield. BET technique has not been extensively employed by researchers in works related to the use of functionalized graphene as an adsorbent of liquid pollutants; moreover, not always the Nano sorbents withgreater specific surface area (SBET, given in $m^2 \cdot g^{-1}$) show great maximum adsorptive capacity (qm, given in $mg \cdot g^{-1}$) [11, 21, 25, 26, 27, 28, 29].

**TEM (Transmission electron microscopy) [11, 30, 31, 32, 33]:** TEM is also a very useful technology for studying functional groups on the surface of graphene and all nanocarriers. Graphene oxide nanocarrier can be obtained by two-step functionalization. One is FA functionalization to obtain GO-FA; the other can be β-CD functionalization to obtain graphene oxide that can be identified by transmission electron microscopy. Company & model of TEM (TEM, Tecnai T12).

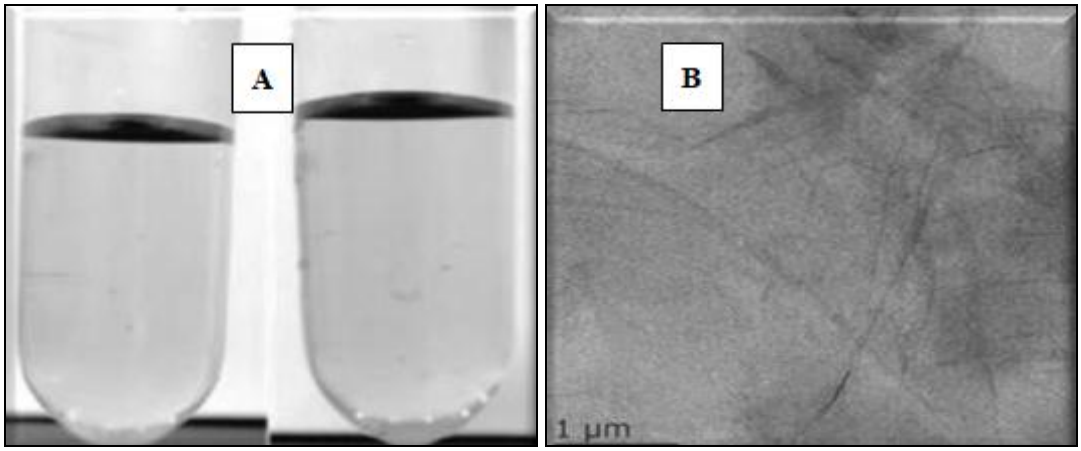


**FIG. 5: (A) NANOCARRIER SOLUTION AFTER (B) TEM IMAGE OF GRAPHENE OXIDE**

**Drug Loading Assay:** Our main goal was to achieve high concentrations of loaded drugs on GO-COOH. For this purpose, the maximum loading concentrations of each drug were measured. Then, simultaneous loading of two different drugs was conducted at the same concentration of maximum drug loading. pH plays an important role in the loading of drugs on GO-COOH. In order to achieve maximum loading, the pH of drugs must be close to the pH of GO-COOH (5.5-6); otherwise, GO-COOH is going to accumulate. Therefore, drug loading efficiency (% EE) will be reduced [34].

**Effect of pH on Graphene by High-Performance Liquid Chromatography (HPLC):** The binding of any drug at different pH levels is determined via HPLC. The drug (50 µM) was mixed with a series

dilution of PGNS and GO (15−500 μg/mL) in triplicate at 3 pH levels of 5, 6.8, and 7.4. After 24 h, 300 μL of the mixture was moved to a 10 kDa MWCO filtration plate and centrifuged over a collection plate at 500 rpm until complete filtration [35].

**UV-Vis Spectroscopy:** UV−Vis Spectroscopy techniques are important for identification. The Drug (Cayman Chemical, Michigan, and United States) was added in increasing concentrations (15 μM to 1 mm) to 1 mg/mL of PGNS and GO suspensions. After 24 h, 3000 RC.

Centrifugation for 10 min was performed to sediment larger agglomerates. The supernatants were collected, and their concentration was measured using UV-vis spectroscopy by integrating the area under the spectrum in the visible range between 400 and 900 nm. DLS. Size measurement of PE-CVD graphene and graphene oxide at 100 μg/mL concentration was carried out continuously over time using DLS (Malvern Analytical Ltd, UK) for 150 min with and without 1 mg drug.

**Zeta Potential by Dynamic Light Scattering (DLS):** The Zeta sizer is an important instrument for the determination of the size of all Nano range particles and it also measures to zeta potential of particles in Nano-grade formulation if anyone studies graphene oxide; this instrument plays a crucial role. Nowadays, a U.K.-based company Produces a DLS-based Zeta-sizer Nano ZSP (Malvern Analytical, Malvern, United Kingdom) [35, 36].

**Atomic Force Microscopy (AFM):** This technique also plays a very important role in the analysis of nano-grade formulation or nanocarrier, which takes 20 μL of the suspension samples. During this technique, the sample can placed on a mica substrate evaporated at 50 °C for >30 min and allowed to cool before analysis. The sample was analyzed in repulsive mode on an MFP-3D-BIO (Asylum Research, Oxford Instruments, and California). Noise filtration was performed using two-dimensional fast Fourier transform (2D-FFT) filtering in Gwydion 2.57 [35].

**Binding Kinetics by UV−Vis Kinetic Measurements:** Binding kinetic is an important parameter for studying any nano carrier because it play a crucial role in maintaining drug level in blood so during this process firstly quartz UV cuvettes, 1 mL of 50 μM of drug (the concentration based on a pilot experiment showing equilibrium reached within 24 h under the same parameters without graphene) then it can be separate from 400 mL of water suspensions of PGNS and graphene oxide at concentrations of 500 and 1000 μg/mL using a 20K MWCO RC membrane. Absorbance measurements on the specific nm that taken (15-minute intervals) use a UV-visible spectrophotometer such as Shimadzu UV-1800 UV-visible spectrophotometer or can use other spectrophotometer use for determination for binding kinetic properties of nano carriers. if decrease in absorbance correlated with the decrease of the drug in the lower compartment and its binding to graphene and nano carriers [37].

**Fluorescence Lifetime-Based FLIM-FRET:** Fluorescence life time-based FLIM-FRET is novel technology for study of dose changes related effect of carrier and their effectiveness and toxicity related parameter. During this techniques dose can be (15 μg/mL-100 μg/mL) fluoresce in in exposure of graphene with and without the addition 0.5 and 1 mg of the drug was recorded using a Leica TCS SP8 falcon platform (Leica Microsystems, Mannheim, Germany). The lifetime-based FILM FRET can collect and fitted to a two-exponential tail fit decay to calculate the intensity-based mean lifetimes and the FLIM-FRET changes corresponding to drug doses [35, 38].

**Microscale Thermophoresis (MST):** This technology Microscale thermophoresis (MST; Monolith NT.115, NanoTemper) can use for the identification study of types of graphene oxide & other nanocarrier determined via Serial concentrations of the drug (0-1 mM) in mixed with graphene oxide or other nanocarrier to a final concentration of 50 μg/mL. Afterward, each mixture can be pulled into a capillary and set into capillary compartment [35].

**Statistical Analysis:** All scientific data can be analyzed by using the *t*-test for differences. That can be reported as means ± standard deviation [38].

**Applications:** The excellent performance of Graphene oxide is based on the inseparable synergy

between hydrophobicity and p-conjugated structure in graphene sheets. Graphene has mechanical strength, electrical conductivity, adsorption, hygroscopicity, water retention, controlled-release, and biocompatibility together, and it will have broad application prospects in biomedical, supercapacitor, water treatment, dye absorption, catalyst carrier, and intelligent response for microfluidic system that are followings types which given below.

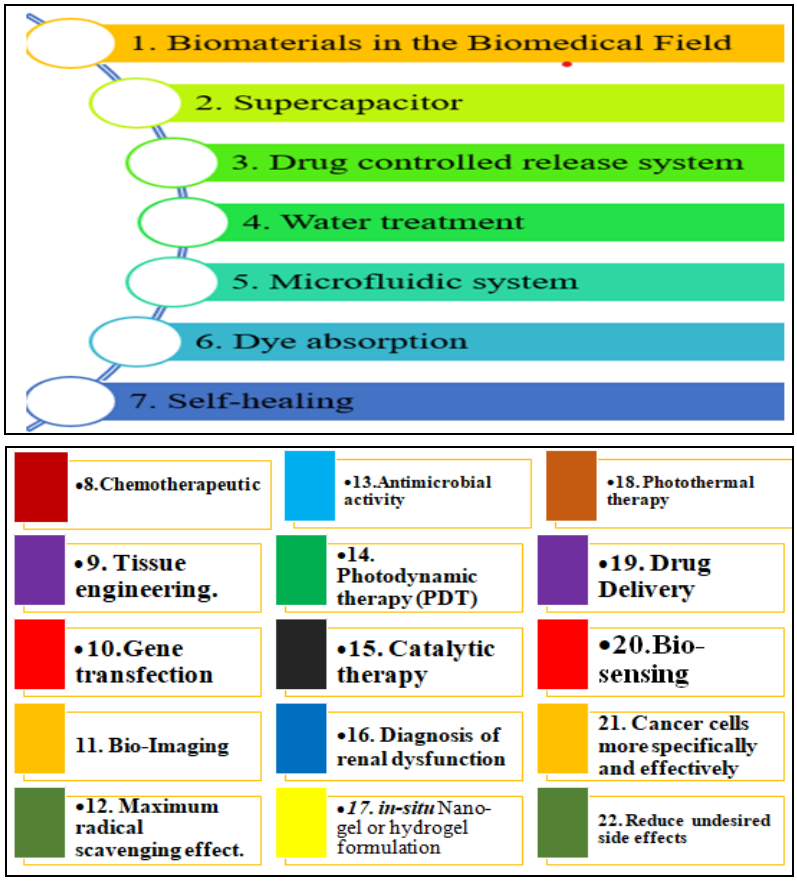


**FIG. 6: APPLICATIONS OF GRAPHENE-BASED SYSTEM**

The integration of graphene and polymers brings electrical conductivity to scaffolds, making them ideal for cardiac or neuronal tissue engineering [39, 40, 41].

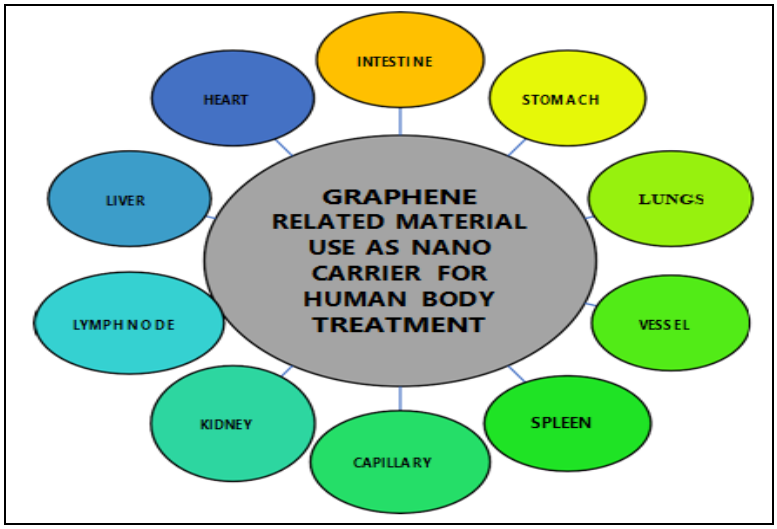


**FIG. 7: GRAPHENE-RELATED MATERIAL CAN BE USED AS A NANOCARRIER IN THE HUMAN BODY FOR TREATMENT**

**CONCLUSION:** This presents an overview of graphene and graphene-related nanomaterial carriers or nano products with superior activity and highly selectivity for any disease. These nanocarriers are very effective in also developing target-related formulations with high selectivity, so graphene can provide a suitable nanocarrier for these target-based or, disease-specific or organ-based delivery system for better therapeutic activity, which reduces the toxicity of drugs compared to other drug delivery systems. Graphene is also part of the novel and nano-grade material and includes how to characterize the study of all nanocarriers; this review provides all specific aspects of graphene and other nanocarriers.

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## REFERENCES:

1. Tyagi N, Song YH and De R: "Recent progress on bio-compatible nanocarrier-based genistein delivery systems in cancertherapy. Journal of Drug Targeting 2018; 1-14.
2. Zhao J, Wang Y, Ma Y, Liu Y, Yan B and Wang L: "Smart nanocarrierbased on PEGylated hyaluronic acid for deacetyl mycoepoxydience: high stability with enhanced bioavailability and efficiency. Carbohydrate Polymers 2019; 203: 356-68.
3. Banerjee S and Kundu A: "Lipid-drug conjugates: a potential nanocarrier system for oral drug delivery applications," DARU Journal of Pharmaceutical Sciences 2018; 26.
4. Borandeh S, Abdolmaleki A, Abolmaali SS and Tamaddon AM: Synthesis, structural and *in-vitro* characterization of β-cyclodextrin grafted L- phenylalanine functionalized grapheme oxide nanocomposite: a versatile nanocarrier for pH-sensitive doxorubicin delivery. Carbohydrate Polymers 2018; 201: 151–161.
5. Joshi K, Mazumder B, Chattopadhyay P, Bora NS, Goyary D & Karmakar S: Graphene family of nanomaterials: reviewing advanced applications in drug delivery and medicine. Current Drug Delivery 2018; 15. doi:10.2174/1567201815666181031163116
6. Chen D, Feng H & Li J: Graphene oxide: Preparation, functionalization, and electrochemical applications. Chem Rev 2012; 112: 6027–6053.
7. Mukherjee S, Bytesnikova Z, Ashrafi AM, Adam V and Richtera L: Graphene oxide as a nanocarrier for biochemical molecules: current understanding and trends. Processes 2020; 8(12): 1636. https://doi.org/10.3390/pr8121636
8. Hu X, Chen S, Gong X, Gao Z, Wang X and Chen P: Synthesis and preparation of biocompatible and pH-responsive cyclodextrin-based nanoparticle. Journal of Nanoparticle Research 2017; 19(3): 109.
9. Marcano DC: Improved Synthesis of Graphene Oxide. ACS Nano 2010; 4: 4806–4814.
10. Dideikin AT & Vul' AY: Graphene oxide and derivatives: The place in graphene family. Front Phys 2019; 6: 149.
11. Tiago José Marques Fraga: 2020 Nano Ex. 1 022002 DOI 10.1088/2632-959X/abb921.
12. Yousefi N, Lu X, Elimelech M and Tufenkji N: Nat. Nanotechnol 2019; 14: 107.
13. Kovaříček P, Vrkoslav V, Plšek J, Bastl Z, Fridrichová M, Drogowska K and Kalbáč M: 2017 Carbon 118 200
14. Wu JB, Lin ML, Cong X, Liu HN and Tan PH: Chem Soc Rev 2018; 47: 1822
15. Malard LM, Pimenta MA, Dresselhaus G and Dresselhaus MS: Phys Rep 2009; 473: 51.
16. Ahmed MS and Kim YB: Carbon 2017; 111: 577.
17. Jiang T, Liu W, Mao Y, Zhang L, Cheng J, Gong M, Zhao H, Dai L, Zhang S and Zhao Q: Chem Eng J 2015; 259: 603.
18. Gao T, Yu J, Zhou Y and Jiang X: J Taiwan Inst Chem Eng 2017; 71: 426.
19. Zhao W, Tang Y, Xi J and Kong J: Appl Surf Sci 2015; 326: 276.
20. Gholampour A, Valizadeh Kiamahalleh M, Tran DN H, Ozbakkaloglu T and Losic D: ACS Appl Mater Interfaces 2017; 9: 43275.
21. Yousefi N, Lu X, Elimelech M and Tufenkji N: Nat Nanotechnol 2019; 14: 107.
22. Spyrou K and Rudolf P: Functionalization of Graphene ed V Georgakilas (Weinheim, Germany: Wiley-VCH Verlag GmbH & Co. KGaA) 2014.
23. Wang J, Salihi E C and Šiller S: Mater. Sci. Eng. C 2017; 72: 1.
24. Sahraei R, Hemmati K and Ghaemy M: RSC Adv 2016; 6: 72487.
25. Mojica-Sepulveda RD, Mendoza-Herrera LJ, Grumel E, Soria DB, Cabello CI and Trivi M: Appl Surf Sci 2018; 447: 587.
26. Zhou Y, Apul OG and Karanfil T: Water Res 2015; 79: 57.
27. Gregg SJ and Sing KSW: Adsorption, Surface Area and Porosity (New York, NY: Wiley 1982.
28. Hackley VA and Stefaniak AB: J Nanoparticle Res 2013; 15: 1742–91.
29. Fan C, Herrera LF, Do DD and Nicholson D: Langmuir 2010; 26: 5610.
30. Ghislandi M, Tkaly E, Alekseev A, Koning C and De with G: Appl Mater Today 2015; 1: 88.
31. Yang K, Feng L, Shi X and Liu Z: Chem Soc Rev 2013; 42: 530.
32. Chandra V and Kim KS: Chem Commun 2011; 47: 3942.
33. McAllister MJ: Chem Mater 2007; 19: 4396.
34. Yaghoubi F, Hosseini Motlagh NS, Naghib SM, Haghiralsadat F, Jaliani HZ & Moradi A: A functionalized graphene oxide with improved cytocompatibility for stimuli-responsive co-delivery of curcumin and doxorubicin in cancer treatment. Scientific Reports 2022; 12. https://doi.org/10.1038/s41598-022-05793-9
35. Abdelnour Alhourani, Jan-Lukas Førde, Mojdeh Nasrollahzadeh, Lutz Andreas Eichacker, Lars Herfindal and Hanne Røland Hagland: Graphene-based phenformin carriers for cancer cell treatment: a comparative study between oxidized and pegylated pristine graphene in human cells and zebrafish. Nanoscale Advances 2022; 4(6): 1668-1680. https://doi.org/10.1039/D1NA00778E

36. Bhattacharjee S, DLS and zeta potential – What they are and what they are not. Journal of Controlled Release 2016; 235: 337–351.
37. Kaya NS, Yadav A, Wehrhold M, Zuccaro L & Balasubramanian K: Binding Kinetics of Methylene Blue on Monolayer Graphene Investigated by Multiparameter Surface Plasmon Resonance. ACS Omega 2018; 3(7): 7133–7140. doi:10.1021/acsomega.8b00689
38. Şenel B, Demir N, Büyükköroğlu G & Yıldız M: Graphene quantum dots: Synthesis, characterization, cell viability, genotoxicity for biomedical applications. Saudi Pharmaceutical Journal 2019. doi:10.1016/j.jsps.2019.05.006
39. Han Y: A simple and effective flexible electrochemiluminescence sensor for lidocaine detection. 2020; 116: 106760.
40. Ghany NAA: Revolution of Graphene for different applications: State-of-the-art 2017; 9: 93-106.
41. Bharech S and Kumar RJJMSME: A review on the properties and applications of graphene 2015; 2(10): 70.

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