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Review Article

DNA Origami–Based Drug Delivery System

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ABSTRACT

DNA origami has emerged as a highly programmable and structurally adaptable framework for next-generation drug delivery systems within the field of nanomedicine. Utilizing precise base-pairing interactions, lengthy single-stranded DNA scaffolds may be folded into specified nanoscale designs with the aid of several short "staple" strands. These structures demonstrate outstanding biocompatibility, structural tunability, and molecular recognition ability, enabling targeted, stimuli-responsive, and high-efficiency therapeutic administration. DNA origami nanocarriers have been examined for the encapsulation, conjugation, or intercalation of anticancer medicines, nucleic acids, proteins, and immunomodulatory substances. Their programmable surfaces allow cell-specific targeting from ligand functionalization and regulated release triggered by pH, enzymes, light, or chemical signals. Despite promising potential, difficulties such as nuclease degradation, high production cost, immunological activation, and scaling restrictions remain impediments to clinical translation. Rapid advancements including chemically stable DNA nanostructures, hybrid nanocarriers, autonomous logic-gated systems, and in vivo imaging-guided delivery are gradually tackling these challenges. This paper gives a complete overview of DNA origami principles, structural classes, drug loading methods, biological applications, limits, and future possibilities. The talk stresses the revolutionary significance of DNA nanotechnology in generating precise, customized, and least hazardous drug delivery systems for next-generation therapeutic interventions.

INTRODUCTION

Recent advances in nanomedicine have driven the development of improved, extremely precise drug delivery devices capable of overcoming the constraints of conventional delivery technologies. Traditional medication formulations frequently demonstrate low solubility, fast clearance, systemic toxicity, and limited target specificity, decreasing therapeutic effectiveness. Nanotechnology-based delivery platforms such as liposomes, dendrimers, polymeric nanoparticles, and inorganic nanomaterials have enhanced

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medication encapsulation and biodistribution to some extent. However, questions relating to toxicity, lack of programmability, restricted loading capacity, and biological instability remain unanswered.

DNA origami provides a completely different paradigm in building drug delivery vehicles. Introduced by Paul Rothemund in 2006, DNA origami employs predictable Watson–Crick base-pairing principles to fold a long viral DNA strand into a desired form using hundreds of synthetic short "staple" strands. This method enables researchers to manufacture nanostructures of practically limitless shapes 2D patterns, 3D polyhedral, tubes, capsules, cages, boxes, sheets, and dynamic reconfigurable machines with nanoscale accuracy. The extraordinary programmability of DNA allows atomic-scale control over size, surface chemistry, mechanical characteristics, and dynamic behaviour.

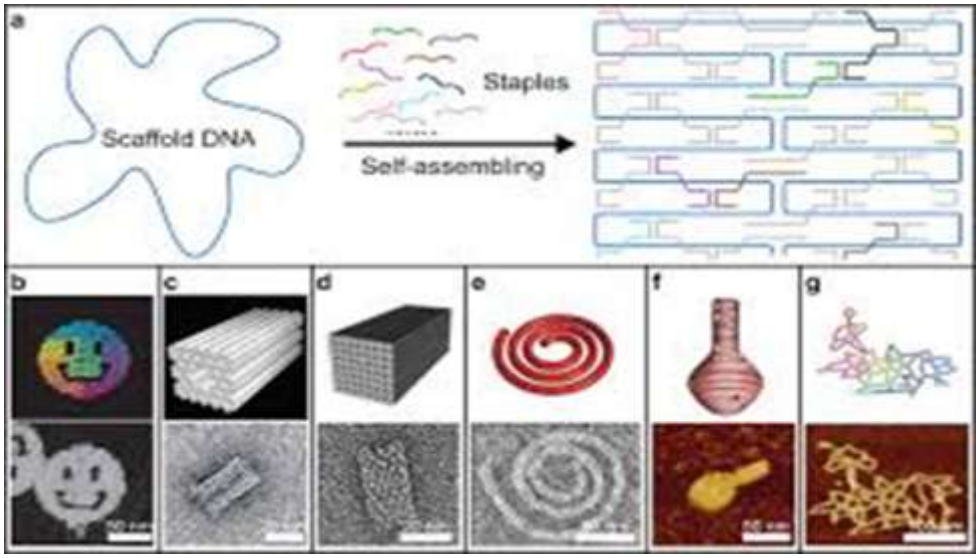

Fig: DNA structure

For medication distribution, these properties translate into numerous advantages: (i) programmable loading of pharmaceuticals at specified places, (ii) molecular recognition-based targeting, (iii) response to environmental cues, (iv) minimum toxicity and immunogenicity, and (v) compatibility with imaging and biosensing modalities. DNA origami nanocarriers have demonstrated encouraging results for chemotherapy delivery (e.g., doxorubicin), gene therapy, immunotherapy, and photodynamic therapies.

This study examines the background of DNA origami, its basic design concepts, structural kinds, drug loading and release processes, medicinal uses, problems, and prospects. The purpose is to give a thorough, accessible, and scientifically sound review of the current state and future directions of DNA origami–based drug delivery systems.

Background

DNA Nanotechnology's Emergence

Ned Seeman developed DNA nanotechnology in the early 1980s after realizing that artificial nanoscale structures might be created using DNA's predictable base pairing. Researchers created DNA tiles, lattices, junctions, and geometric structures during the ensuing decades, establishing the groundwork for the DNA origami method.

DNA Origami's Introduction

DNA origami was first described by Paul W.K. Rothemund in 2006. It involves employing complementary short oligonucleotides called staples to fold a lengthy ssDNA scaffold typically from the M13 bacteriophage into exact forms. This technique transformed the discipline by making it possible to create very intricate, inflexible, and programmable nanostructures with atomic-level accuracy.

Why Use DNA to Deliver Drugs?

Targeting ligands, imaging agents, aptamers, or medicinal compounds may readily functionalize DNA, which is naturally biocompatible, biodegradable, and non-toxic. DNA origami is a particularly appealing material for drug delivery applications because of these characteristics.

Principles of DNA Origami

DNA origami is a nanoscale fabrication technique in which a long single-stranded DNA (commonly derived from bacteriophage M13mp18) is folded into predetermined shapes using hundreds of short complementary oligonucleotides known as "staple strands." This method relies fundamentally on the predictable base-pairing rules of DNA, where adenine pairs with thymine and guanine pairs with cytosine through Watson–Crick hydrogen bonding. By designing staple strands with specific sequences, the long scaffold strand can be precisely guided to fold into two-dimensional (2D) and three-dimensional (3D) nanoscale architectures.

Programmability, or the capacity to directly encode structural information into nucleotide sequences, is the fundamental idea underlying DNA origami. Researchers can precisely make complex nano-objects, designate crossover sites, and organize helices using computational design tools like caDNAno. The ideal geometric orientation and structural stiffness are determined by crossovers, which occur when strands move between neighboring DNA helices. Under carefully regulated thermal annealing conditions, when the mixture is progressively chilled to enable proper hybridization, these preprogrammed interactions guarantee that the final structure self-assembles.

Thermodynamic self-assembly is a fundamental idea. DNA origami structures form spontaneously because the designed hybridization patterns minimize the system's free energy, guiding the scaffold into the lowest-energy conformation. Because of the high degree of cooperation in this self-assembly process, partial motif formation promotes additional binding, resulting in the formation of robust and repeatable architecture.

Addressability is another essential concept. Functional compounds like medications, targeting ligands, fluorophores, or nanoparticles may be precisely positioned since each base pair in the origami structure corresponds to a defined spatial location. DNA origami is incredibly effective for biomedical applications, particularly drug delivery, due to its nanoscale precision.

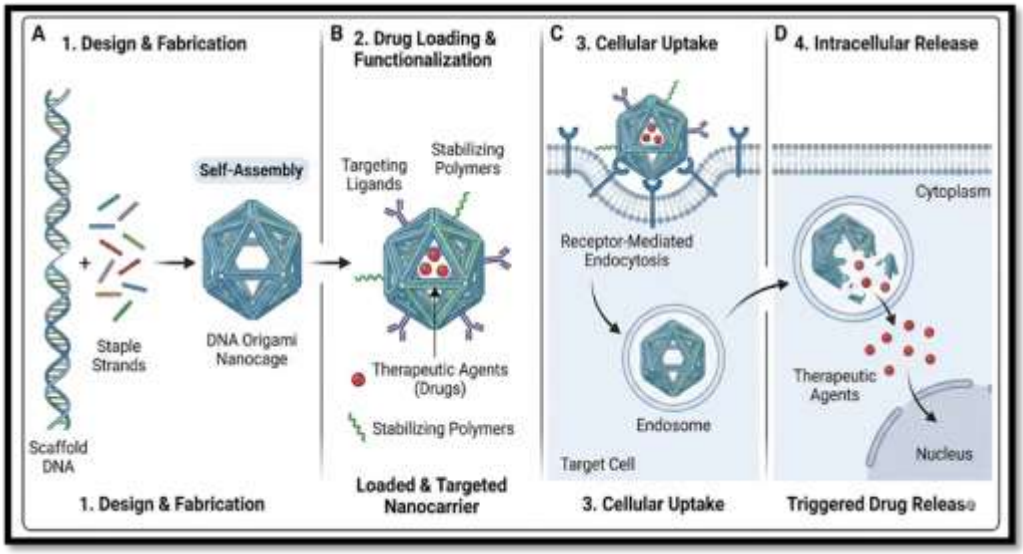

Fig.2: Schematic Representation of DNA Origami Based Drug Delivery System

Types of DNA Origami Structures for Drug Delivery

2D Sheets and Tiles

Flat rectangles or sheets are uncomplicated to create and offer extensive surface area for multivalent ligand display and adsorption of hydrophobic medicines intercalated between base pairs. They are commonly utilized for in vitro cell-surface interactions and as scaffolding for assembling enzymes or multicomponent complexes.

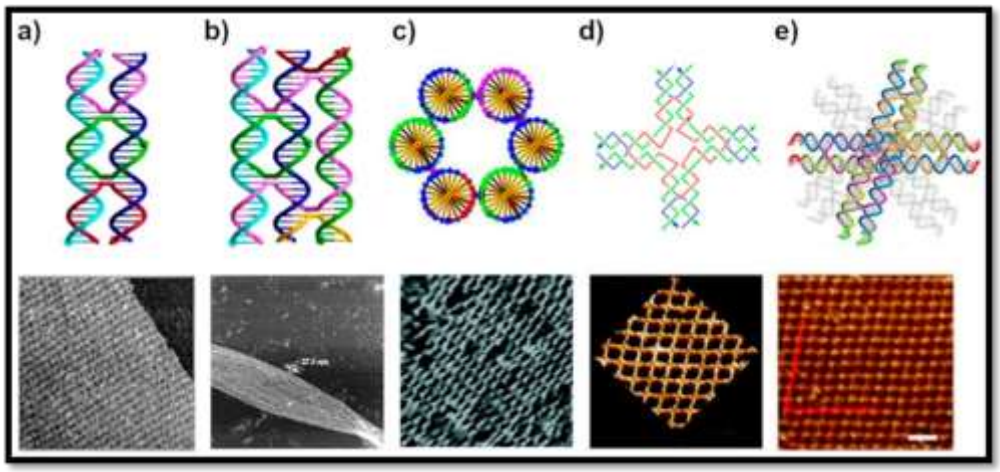

Fig: 2D Sheets and Tiles DNA structure

Hollow 3D Polyhedral (Boxes, Cages)

Closed or semi-closed polyhedral cages (boxes, cubes, tetrahedra) generate internal cavities to encapsulate small-molecule medicines, proteins, or siRNA. Designs generally feature a controlled lid or gate locked by DNA strands that can be opened by strand displacement, aptamer binding, or environmental stimuli allowing triggered release.

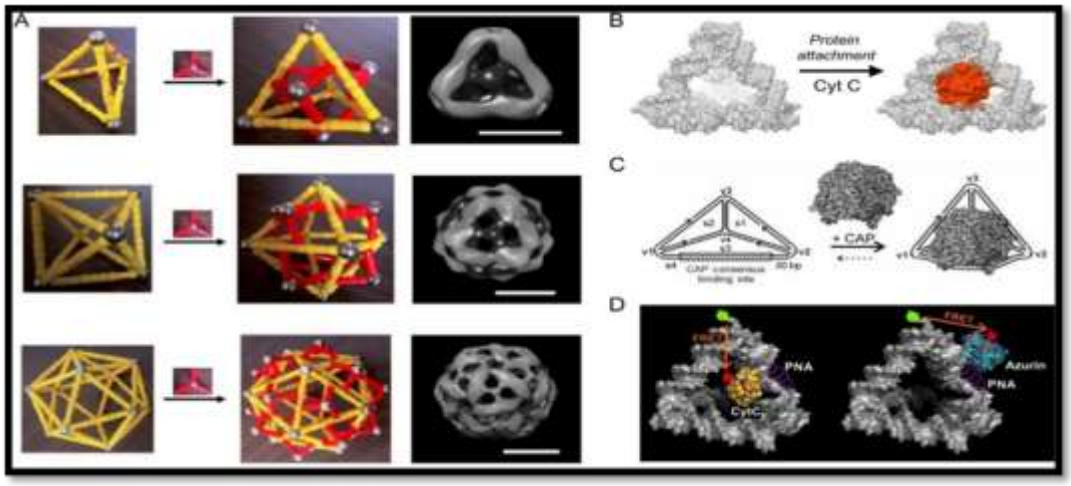

Fig: Hollow 3D Polyhedral DNA structure

Tubular and Rod-like Structures

Rigid rods and tubes (e.g., six-helix bundles) promote cellular absorption and can penetrate tissues more readily. Tubular morphologies can be loaded inside or adorned externally with ligands for cell-targeting.

Wireframe Architectures

Minimal-mass wireframe architectures combine mechanical stability with reduced immunogenic mass and lower nuclease target density. They are advantageous for minimizing clearance and permitting larger internal pockets with less material.

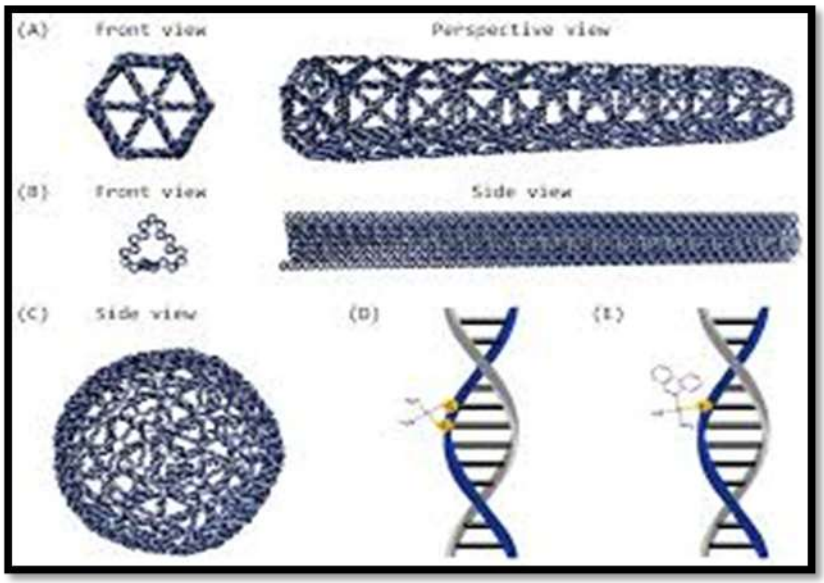

Fig: Wireframe Architectures DNA structure

DNA Origami with Lipid or Polymer Coatings (Hybrid Systems)

Combining DNA origami with lipid bilayers (lipid–DNA hybrids) or polymer shells (PEG, polyethyleneimine) boosts serum stability and improves pharmacokinetics. Coatings also permit membrane fusion or endosomal escape methods.

Mechanisms of Cargo Loading and Release

Understanding how cargos are linked with DNA origami and then released is vital for rational design.

Cargo Loading Mechanisms

Physical Encapsulation

Cages/Boxes: Drugs are contained inside a closed cavity during assembly or loaded post-assembly through removable lids or pores. Encapsulation shields labile cargos (proteins, RNA) from degradation and prevents off-target interactions.

Intercalation and Groove Binding

Small-molecule medicines that intercalate DNA (e.g., doxorubicin) naturally bind to double-stranded regions—this strong yet reversible contact has been used extensively. Intercalation enables substantial drug loading per base pair and inherent regulated release as intercalation equilibria alter in different conditions.

Covalent/Conjugation Attachment

Drugs or prodrugs can be covalently linked to staple strands by cleavable linkers (disulfide, enzymatic). Covalent connection ensures stable transport during circulation; release is mediated by cleavage in target microenvironment (reducing cytosol, particular enzymes).

Hybridization-Based Attachment

Oligonucleotide-functionalized medicines, peptides, or nanoparticles hybridize to single-stranded handles on the origami, permitting modular and reversible loading. Strand-displacement systems can subsequently discharge cargo when activated

Noncovalent Surface Adsorption / Electrostatic Complexing

Charged biomolecules (siRNA) or cationic polymers can electrostatically bind the negatively charged DNA origami. Compaction using polycations can further condense nucleic acid cargos for cellular absorption

Encapsulation via Co-assembly

Protein cargos can be integrated during assembly by linking binding motifs to staples that recruit proteins into internal pockets.

Release Mechanisms

Environmental Stimuli-Triggered Release

pH-responsive systems: Acid-labile locks or i-motif sequences cause opening in acidic endosomal/lysosomal compartments

Redox-sensitive release

Disulfide linkers cleave in the reducing cytosol, freeing covalently bound drugs/oligos.

Enzyme-responsive

Protease-cleavable peptide linkers or nuclease-susceptible strand intended to be destroyed by overexpressed tumor enzymes release cargo preferentially at illness locations.

Strand-Displacement and Oligonucleotide Triggers

An added complementary DNA/RNA strand displaces a locking strand via toehold-mediated strand displacement, opening lids or detaching cargo. This enables excellent specificity and tunability.

Competitive Binding and Dilution Effects

For intercalated medicines, changes in ionic strength, presence of competing DNA, or dilution

in bodily fluids can shift binding equilibria toward release.

Mechanical/Conformational Switching

Conformational changes (e.g., closure to open) mediated by ligand binding can expose internal cargo to external milieu for diffusion-driven release.

External Physical Triggers

Light-responsive groups (photo-cleavable linkers), heat (photothermal agents combined with origami), or magnetic fields (via connected nanoparticles) can enable remote-triggered release.

Cellular Uptake and Intracellular Processing

Uptake methods primarily follow receptor-mediated endocytosis when targeting ligands are provided, or macropinocytosis for bigger forms. Once internalized, escape from endosomes is vital; options include insertion of endosomal escape peptides, pH-triggered membrane-disruptive patterns, or co-delivery with fusogenic lipids. Intracellular release generally relies on redox or enzymatic cues outlined above. Finally, regulated breakdown of the DNA scaffold lowers long-term accumulation

Formulations of DNA Origami–Based Drug Delivery Systems

Doxorubicin-Loaded DNA Origami Nanostructures

Because it naturally intercalates into double-stranded DNA, doxorubicin (DOX) is the most studied DNA origami model treatment. This characteristic makes it perfect for loading efficiency and structural stability testing. Early studies used DOX at controlled molar ratios to create two-dimensional DNA origami sheets and triangles. Every base pair is a possible binding site, thus intercalation increased loading densities. These formulations balance loading and structural integrity: excessive DOX can destabilize local duplex areas or change mechanical rigidity, while modest intercalation preserves origami shape. DOX-loaded origami developed larger solid tumors than free DOX in animal studies due to blood stability and slower renal clearance. The DNA scaffold enabled progressive drug diffusion from tumor vasculature to tumor cells by preventing rapid release.

Using origami tubes and rods to encapsulate DOX in a hollow interior minimizes premature release. These structures circulate longer and have lesser cardiotoxicity than free DOX when coated with PEG or lipids.

Aptamer-Functionalized Targeted Formulations

Aptamer-functionalization is a highly effective targeting strategy. For example, rectangular DNA origami tiles have been decorated with tumor-targeting aptamers, such as AS1411 (nucleolin-targeting) or EpCAM-targeting sequences. By arranging these ligands at precise positions, researchers create multivalent interactions that significantly enhance binding specificity and uptake.

In one formulation example, a DNA origami tube carrying DOX was functionalized with AS1411 aptamers placed symmetrically along its length. This arrangement improved selective uptake in nucleolin-overexpressing cancer cells, resulting in nearly double the cytotoxicity compared to non-targeted DNA origami. Additionally, the aptamer functionalization stabilized the nanostructure by partially shielding it from serum proteins.

Lipid-Coated DNA Origami Nanocarriers

Lipid bilayer coverings mirror natural viral envelopes and dramatically boost stability in physiological fluids. In lipid-coated formulations, DNA origami carriers—usually tubes, boxes or sheets—are enclosed in a supportive lipid membrane comprised of phosphatidylcholine or fusogenic lipids.

One study formed a lipid-coated DNA origami rod loaded with doxorubicin by intercalation. The lipid barrier minimized DNA exposure to nucleases and greatly increased hemocompatibility. Moreover, the lipid formulation permitted greater endosomal escape due to the presence of DOPE (dioleoylphosphatidylethanolamine), a fusogenic lipid that destabilizes endosomal membranes at acidic pH.

This technique links DNA nanotechnology with clinically effective lipid-based delivery (such as liposomes and LNPs), creating a hybrid design that shows great potential for translational development.

APPLICATIONS

DNA origami–based nanostructures have emerged as very adaptable platforms for modern drug delivery, offering programmable architecture, biocompatibility, and precise spatial control over therapeutic molecules. Their unique capacity to self-assemble into preset forms offers applications across cancer therapy, gene regulation, antibiotic administration, biosensing, and customized medicine. One of the most significant uses is tailored cancer medication delivery. DNA origami carriers can be created to encapsulate chemotherapeutic drugs such as doxorubicin (DOX) within their double-helical structures through intercalation. By attaching tumor-specific ligands, aptamers, or antibodies to the external surface, these nanodevices promote selective accumulation in cancer cells while minimizing systemic toxicity. Furthermore, researchers have produced “smart” DNA origami nanorobots that open only in reaction to tumor-specific stimuli—such as pH, enzymes, or nucleic acid markers—releasing medications precisely at the sick spot. Another key application involves gene delivery and gene regulation. DNA origami structures can carry small interfering RNA (siRNA), microRNA (miRNA), and antisense oligonucleotides to control gene expression. Their predictable structure allows precise loading and preservation of nucleic acids against enzyme breakdown in the circulation. In addition, surface modification with cationic polymers or targeting ligands promotes cellular uptake and endosomal escape, hence boosting gene silencing efficacy. Such systems offer promise in treating genetic diseases, viral infections, and cancer.

DNA origami is also commonly employed in immunotherapy. Nanostructures can deliver many antigens or immune-regulating chemicals with nanoscale precision, enabling effective activation of immune cells. For example, DNA origami vaccines can exhibit repeating antigenic patterns that imitate virus architecture, resulting in greater immune responses compared to traditional vaccines. These systems can also distribute adjuvants, cytokines, or immune checkpoint inhibitors in a controlled manner, thus helping cancer immunotherapy and infectious disease management.

In antimicrobial therapy, DNA origami carriers provide a new response to rising antibiotic resistance. Researchers have built nanostructures capable of delivering antimicrobial peptides (AMPs) or medicines directly to bacterial membranes. Their high surface area and changeable design allow multivalent binding to

bacterial targets, improving therapeutic effects even at lower medication doses. Such technologies are particularly effective for biofilm-forming infections, where traditional antibiotics often fail. Another new application is theranostics, where DNA origami constructs integrate both therapeutic and diagnostic activities. By adding fluorescent markers, gold nanoparticles, or quantum dots, these nanodevices enable simultaneous imaging and drug delivery. This dual feature provides real-time monitoring of medication delivery, biodistribution, and treatment response. Theranostic DNA origami technologies are extremely promising for individualized cancer treatment.

Lastly, DNA origami nanostructures have a vital role in penetrating biological barriers, such as the blood–brain barrier (BBB). By attaching BBB-penetrating peptides or receptor-targeting ligands, these carriers can deliver neurologically active medications to the brain, enabling novel therapeutic options for Alzheimer's disease, Parkinson's disease, and brain malignancies.

FUTURE PROSPECTS

Future work should target enhancing serum stability and scalable manufacture, robust in vivo pharmacokinetics and toxicity research, and development of modular platforms that can be swiftly changed for other cargos. Combining DNA origami with biologically derived materials (exosomes, proteins) may increase stealth and targeting. Clinical niches where precise spatial presentation matters—such as immunoengineering (vaccine platforms) or localized cancer therapies—could be early translation targets. Regulatory pathways will require consistent characterisation of structure, purity, and functional performance. Machine learning-guided design may speed optimization of forms for biodistribution and cellular uptake.

CONCLUSION

DNA origami offers an extraordinarily adaptable and programmable framework for next-generation medication delivery systems. Its addressability and aptitude for dynamic, stimuli-responsive behavior enable complex cargo loading and release tactics not attainable with many traditional carriers. Major hurdles—stability in biological settings, scale-up, and in vivo validation—remain but are being aggressively addressed. With continuous multidisciplinary development (nanotechnology, chemistry, pharmacology), DNA origami has realistic potential to translate into specific therapeutic applications where molecular precision gives clear clinical advantage.

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